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Original Research

Molecular Epidemiology of Methicillin-Resistant Staphylococcus aureus (MRSA) in Tertiary Care Centers.

¹M Mamatha Reddy, ²Syeda Nazia Fathima, ³Hari kumar kuragayala.

¹Assistant Professor, Department of Microbiology, Surabhi Institute of Medical Sciences, Siddipet, Telangana State, India, 502375.

²Post Graduate, Department of Microbiology, Shadhan Institute of Medical Sciences, Peerancheru, Hyderabad, Telangana State, India, 500091.

³Hari kumar kuragayala, Hingeclinica private limited. Cybertowers Hyderabad, Telangana State, India, 500081.

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Abstract

Background: Methicillin-resistant Staphylococcus aureus (MRSA) remains a major cause of healthcare-associated and community-associated infections worldwide. The emergence of diverse MRSA clones possessing distinct resistance and virulence determinants has complicated infection control and therapeutic management. Molecular epidemiological surveillance is essential for understanding the transmission dynamics and genetic diversity of circulating MRSA strains, particularly in regions where comprehensive molecular data remain limited. **Objective:** To investigate the molecular epidemiology, antimicrobial resistance patterns, and clonal distribution of MRSA isolates recovered from a tertiary care centre in Telangana, India. **Methods:** A prospective cross-sectional study was conducted in the Department of Microbiology of a tertiary care teaching hospital in Telangana between March 2020 and August 2020. A total of 180 non-duplicate clinical Staphylococcus aureus isolates obtained from various clinical specimens were included. Phenotypic identification of MRSA was performed using cefoxitin disk diffusion according to Clinical and Laboratory Standards Institute guidelines. Statistical analysis was performed using SPSS version 26.0, and p-values <0.05 were considered statistically significant. **Results:** Among the 180 S. aureus isolates, 74 (41.1%) were identified as MRSA, while 106 (58.9%) were methicillin-susceptible S. aureus (MSSA). Molecular analysis demonstrated mecA positivity in 70 (94.6%) MRSA isolates. SCCmec type III was the predominant genotype (37.1%), followed by type IV (31.4%), type V (14.3%), type II (8.6%), and type I (5.7%), whereas 2.9% of isolates were untypable. Spa typing revealed substantial genetic diversity, with t037 (28.6%) emerging as the dominant lineage, followed by t657 (18.6%), t852 (12.9%), and t6574 (11.4%). **Conclusions:** The study demonstrates a high prevalence of MRSA and considerable molecular diversity among circulating isolates in a tertiary care center in Telangana.

Keywords: Methicillin-resistant Staphylococcus aureus; MRSA; Molecular epidemiology; mecA; SCCmec typing; spa typing; Antimicrobial resistance.

Introduction

Methicillin-resistant Staphylococcus aureus (MRSA) remains one of the most significant multidrug-resistant bacterial pathogens worldwide and continues to pose a major challenge to healthcare systems. Since its first identification shortly after the introduction of methicillin in 1961, MRSA has evolved into a globally disseminated pathogen responsible for a wide spectrum of infections ranging from skin and soft tissue infections to severe invasive diseases such as bacteremia, pneumonia, endocarditis, and sepsis [1]. The remarkable adaptability of MRSA is largely attributed to its ability to acquire and disseminate antimicrobial resistance determinants, particularly the mecA gene, which encodes an altered penicillin-binding protein (PBP2a) with reduced affinity for β -lactam antibiotics [2]. Consequently, MRSA infections are associated with prolonged hospital stays, increased healthcare costs, treatment failures, and elevated mortality rates [3].

The epidemiology of MRSA has undergone substantial transformation over the past two decades. Historically, MRSA was considered primarily a healthcare-associated pathogen; however, the emergence of community-associated MRSA (CA-MRSA) has significantly altered transmission dynamics [4]. Distinct molecular lineages have emerged in both healthcare and community settings, demonstrating differences in virulence, antimicrobial resistance profiles, and epidemiological behavior. The global dissemination of epidemic clones such as ST239, ST5, ST22, ST8 (USA300), and ST59 has highlighted the importance of molecular surveillance in understanding the evolution and spread of MRSA strains [5]. The increasing movement of patients, healthcare workers, and populations across regions further facilitates the international transmission of successful MRSA clones, emphasizing the need for region-specific epidemiological investigations [6].

India has witnessed a substantial increase in MRSA prevalence over the last two decades. Multiple multicentric surveillance studies have reported MRSA prevalence rates ranging from 25% to 54% among clinical S. aureus isolates, although considerable regional variation exists [7]. Factors contributing to the increasing burden include widespread antibiotic misuse, inadequate infection control practices, overcrowded healthcare facilities, and limited antimicrobial stewardship programs [8]. Several studies from different regions of India have documented the circulation of diverse MRSA clones, including ST22-MRSA-IV, ST239-MRSA-III, and emerging community-associated lineages [9,10]. The coexistence of hospital-associated and community-associated strains has created a complex epidemiological landscape that necessitates continuous molecular monitoring.

The emergence and dissemination of specific MRSA molecular clones are primarily driven by mobile genetic elements, particularly the staphylococcal cassette chromosome mec (SCCmec). SCCmec is a large genomic island carrying the mecA gene and additional resistance determinants that contribute to the adaptability and survival of MRSA in different ecological niches [11]. To date, several SCCmec types have been identified, with types I, II, and III commonly associated with healthcare-associated MRSA and types IV and V frequently linked to community-associated strains [12]. SCCmec typing therefore provides valuable insights into the origin, transmission patterns, and evolutionary

relationships of MRSA isolates.

In addition to SCCmec characterization, *spa* typing has emerged as a highly discriminatory molecular epidemiological tool for investigating MRSA diversity. *Spa* typing is based on sequencing of the polymorphic X region of the protein A (*spa*) gene and allows rapid identification of clonal lineages with excellent reproducibility and inter-laboratory comparability [13]. Compared with conventional typing methods, *spa* typing offers a standardized approach for global surveillance and outbreak investigations. The combined application of SCCmec and *spa* typing enables comprehensive characterization of circulating MRSA strains and facilitates comparison of local epidemiological patterns with national and international trends [14].

Telangana is one of the rapidly developing states in southern India with increasing healthcare utilization and patient referrals from neighboring regions. Despite the growing burden of antimicrobial resistance, limited information is available regarding the molecular epidemiology of MRSA in tertiary care centers within Telangana. Most published studies from India have focused primarily on phenotypic antimicrobial resistance patterns, while detailed molecular characterization of circulating MRSA clones remains relatively scarce. Furthermore, data regarding SCCmec distribution, *spa* type diversity, and the prevalence of virulence-associated genetic markers among clinical MRSA isolates in Telangana are insufficient [9,15].

Addressing these knowledge gaps is essential for understanding local transmission dynamics, identifying emerging epidemic clones, and strengthening infection prevention strategies. Therefore, the present study was undertaken to investigate the molecular epidemiology of MRSA isolates obtained from a tertiary care center in Telangana through molecular detection of *mecA*, SCCmec characterization, *spa* typing, and analysis of antimicrobial resistance profiles. The findings are expected to contribute to regional surveillance efforts and provide evidence-based guidance for infection control and antimicrobial stewardship programs.

Address for Correspondence Dr. M Mamatha Reddy, Assistant Professor, Department of Microbiology, Surabhi Institute of Medical Sciences, Siddipet, Telangana State, India, 502375

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Materials and Methods

Study Setting

A prospective cross-sectional observational study was conducted in the Department of Microbiology of a tertiary care teaching hospital located in Telangana, India, over a six-month period from March 2020 to August 2020. The hospital serves as a major referral center catering to urban, semi-urban, and rural populations across Telangana states. Clinical specimens received from both inpatient and outpatient departments were included in the study.

During the study period, a total of 180 non-duplicate clinical isolates of *Staphylococcus aureus* were recovered from various clinical specimens submitted to the microbiology laboratory for routine diagnostic investigations. The specimens included pus, wound swabs, blood, sputum, urine, endotracheal aspirates, catheter tips, and other sterile body fluids. Only the first isolate obtained from each patient was included to avoid duplication and ensure accurate epidemiological assessment.

The study aimed to determine the prevalence, antimicrobial resistance patterns, and molecular characteristics of methicillin-resistant *Staphylococcus aureus* (MRSA) isolates circulating within the tertiary care center. Demographic and clinical data, including age, sex, ward location, specimen type, hospitalization status, previous antibiotic exposure, and underlying comorbidities, were collected from medical records and laboratory requisition forms using a structured data collection format.

Inclusion and Exclusion Criteria

Inclusion Criteria

1. Clinical specimens yielding *Staphylococcus aureus* during the study period.
2. Non-duplicate isolates obtained from individual patients.
3. Isolates recovered from both inpatient and outpatient departments.
4. Isolates from patients of all age groups and both sexes.
5. Adequately preserved isolates suitable for phenotypic and molecular characterization.

Exclusion Criteria

1. Duplicate isolates obtained from the same patient during the same episode of infection.
2. Contaminated cultures or mixed bacterial growths that precluded accurate identification.
3. Isolates with incomplete demographic or clinical information.
4. Non-viable bacterial cultures unsuitable for molecular analysis.
5. Environmental or surveillance isolates not associated with clinical infections.

Laboratory Procedures

Isolation and Identification of *Staphylococcus aureus*

Clinical specimens were processed according to standard microbiological protocols. Samples were inoculated onto Blood Agar and Mannitol Salt Agar (HiMedia Laboratories, Mumbai, India) and incubated aerobically at 35–37°C for 18–24 hours. Colonies exhibiting characteristic morphology suggestive of *S. aureus* were subjected to further identification.

Presumptive isolates were identified based on colony morphology, Gram staining characteristics, and biochemical reactions. Gram-positive cocci arranged in clusters were tested for catalase production using 3% hydrogen peroxide. Catalase-positive isolates were subsequently subjected to slide and tube coagulase tests using rabbit plasma. Mannitol fermentation on Mannitol Salt Agar was also assessed. Identification of *S. aureus* was confirmed using standard microbiological criteria recommended by the Clinical and Laboratory Standards Institute (CLSI).

Phenotypic Detection of MRSA

All confirmed *S. aureus* isolates were screened for methicillin resistance using the cefoxitin disk diffusion method. A 0.5 McFarland bacterial suspension was prepared and inoculated onto Mueller-Hinton Agar plates. A 30 µg cefoxitin disk was placed on the inoculated agar surface, and plates were incubated at 35°C for 18–24 hours.

Interpretation of zone diameters was performed according to CLSI 2020 guidelines. Isolates exhibiting inhibition zones of ≤21 mm were classified as MRSA, while those showing zones ≥22 mm were considered methicillin-susceptible *S. aureus* (MSSA).

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed using the Kirby-Bauer disk diffusion method on Mueller-Hinton Agar following CLSI recommendations. The antibiotics tested included penicillin (10 units), erythromycin (15 µg), clindamycin (2 µg), ciprofloxacin (5 µg), gentamicin (10 µg), cotrimoxazole (25 µg), tetracycline (30 µg), doxycycline (30 µg), linezolid (30 µg), and vancomycin.

Inducible clindamycin resistance was determined using the D-test among erythromycin-resistant isolates. Vancomycin susceptibility was further confirmed by broth microdilution minimum inhibitory concentration (MIC) testing whenever required.

Quality control procedures were performed using *Staphylococcus aureus* ATCC 25923 and *Staphylococcus aureus* ATCC 43300 reference strains.

Molecular Methods

DNA Extraction

Genomic DNA was extracted from all phenotypically confirmed MRSA isolates using a commercially available bacterial genomic DNA extraction kit according to the manufacturer's instructions. Briefly, overnight bacterial cultures were suspended in lysis buffer and subjected to enzymatic digestion followed by purification through silica membrane-based spin columns. DNA concentration and purity were assessed spectrophotometrically and stored at -20°C until further analysis.

Detection of *mecA* Gene

Polymerase chain reaction (PCR) was performed to detect the presence of the *mecA* gene, the principal genetic determinant responsible for methicillin resistance. Amplification reactions were carried out in a thermal cycler using specific primers targeting the *mecA* gene. The PCR mixture contained template DNA, primers, deoxynucleotide triphosphates, Taq DNA polymerase, magnesium chloride, and reaction buffer.

Amplified products were visualized by electrophoresis on 1.5% agarose gel stained with ethidium bromide and examined under ultraviolet illumination. Detection of the expected amplicon size confirmed the presence of the *mecA* gene.

SCCmec Typing

Molecular characterization of MRSA isolates was performed using multiplex PCR targeting SCCmec elements. Specific primer sets were employed to identify SCCmec types I, II, III, IV, and V based on characteristic amplification patterns.

Classification of isolates into SCCmec types enabled differentiation between healthcare-associated and community-associated MRSA strains and provided insight into the genetic diversity of circulating clones within the study population.

spa Typing

The polymorphic X-region of the protein A (*spa*) gene was amplified using PCR. Purified PCR products were subjected to DNA sequencing using an automated genetic analyzer.

Obtained sequences were analyzed and assigned corresponding *spa* types using the RidomSpaServer database. *Spa* typing was utilized to determine clonal relationships among MRSA isolates and identify predominant circulating lineages.

Detection of Panton-Valentine Leukocidin (PVL) Genes

The presence of the *lukS-PV* and *lukF-PV* genes encoding Panton-Valentine leukocidin was investigated using PCR. Amplification products were analyzed by agarose gel electrophoresis. Isolates harboring PVL genes were categorized as PVL-positive MRSA and evaluated for their association with community-acquired infections and specific molecular lineages.

Statistical Analysis

Data were entered into Microsoft Excel and analyzed using Statistical Package for the Social Sciences (SPSS) version 26.0 (IBM Corp., Armonk, NY, USA).

Continuous variables were summarized using means and standard deviations or medians and interquartile ranges, depending on data distribution. Categorical variables were expressed as frequencies and percentages.

Associations between categorical variables were evaluated using the Chi-square test or Fisher's exact test whenever appropriate. Odds ratios (ORs) with 95% confidence intervals (CIs) were calculated to identify factors associated with MRSA infection.

Multivariate logistic regression analysis was performed to determine independent predictors of MRSA acquisition after adjusting for potential confounding variables. Variables demonstrating a p-value of <0.20 in univariate analysis were included in the regression model.

A two-tailed p-value of <0.05 was considered statistically significant. Results were presented in the form of tables, graphs, and molecular epidemiological profiles to facilitate comprehensive interpretation of findings.

Results

Table 1. Demographic Characteristics of Study Participants (n=180)

Variable	n	%
Male	112	62.2
Female	68	37.8
<20 years	24	13.3
21-40 years	78	43.3
41-60 years	54	30.0
>60 years	24	13.3

Table 1. A total of 180 patients with culture-confirmed *Staphylococcus aureus* infections were included in the study. Among them, 112 (62.2%) were males and 68 (37.8%) were females, yielding a male-to-female ratio of approximately 1.6:1. The majority of patients belonged to the 21-40 years age group (43.3%), followed by the 41-60 years age group (30.0%). Patients younger than 20 years and older than 60 years each accounted for 13.3% of the study population. The predominance of young and middle-aged adults suggests that economically productive age groups constituted the major burden of *S. aureus* infections in the tertiary care setting.

Table 2. Distribution of Clinical Specimens (n=180)

Specimen	n	%
Pus/Wound swab	82	45.6
Blood	32	17.8
Sputum	24	13.3
Urine	18	10.0
Endotracheal aspirate	12	6.7
Catheter tip	6	3.3

Other body fluids	6	3.3
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Table 2 Among the 180 clinical specimens yielding *S. aureus*, pus and wound swabs constituted the largest proportion (45.6%), indicating that skin and soft tissue infections were the predominant clinical presentation. Blood cultures accounted for 17.8% of isolates, highlighting the importance of *S. aureus* as a cause of bloodstream infections. Respiratory specimens, including sputum and endotracheal aspirates, contributed 20.0% of isolates, while urinary tract infections accounted for 10.0%. Catheter tips and other sterile body fluids together represented 6.6% of the isolates. These findings demonstrate the diverse clinical manifestations of *S. aureus* infections and emphasize its role as a significant pathogen across multiple body sites.

Table 3. Prevalence of MRSA and MSSA (n=180)

Isolate	n	%
MRSA	74	41.1
MSSA	106	58.9

Table 3 Phenotypic characterization using the cefoxitin disk diffusion method identified 74 isolates (41.1%) as methicillin-resistant *Staphylococcus aureus* (MRSA) and 106 isolates (58.9%) as methicillin-susceptible *Staphylococcus aureus* (MSSA). The observed MRSA prevalence of 41.1% indicates a substantial burden of methicillin resistance within the tertiary care center. This prevalence is comparable to rates reported from several tertiary healthcare institutions across India and reflects the ongoing challenge posed by antimicrobial resistance among clinical *S. aureus* isolates.

Table 4. Antibiotic Susceptibility Pattern among MRSA (n=74)

Antibiotic	Susceptible n (%)
Penicillin	0 (0)
Erythromycin	22 (29.7)
Clindamycin	34 (45.9)
Ciprofloxacin	18 (24.3)
Gentamicin	39 (52.7)
Cotrimoxazole	43 (58.1)
Tetracycline	46 (62.2)
Doxycycline	52 (70.3)
Linezolid	74 (100)
Vancomycin	74 (100)

Table 4 Antimicrobial susceptibility testing revealed high levels of multidrug resistance among MRSA isolates. Complete resistance to penicillin was observed, consistent with the intrinsic resistance profile of MRSA strains. Low susceptibility rates were recorded for ciprofloxacin (24.3%) and erythromycin (29.7%), indicating widespread resistance to fluoroquinolones and macrolides. Moderate susceptibility was observed for clindamycin (45.9%), gentamicin (52.7%), cotrimoxazole (58.1%), and tetracycline (62.2%).

Table 5. Distribution of mecA-positive Isolates (n=74 MRSA)

Molecular Result	n	%
mecA Positive	70	94.6
mecA Negative	4	5.4

Table 5 Molecular analysis revealed that 70 of the 74 phenotypically identified MRSA isolates (94.6%) carried the *mecA* gene, while four isolates (5.4%) were *mecA*-negative. The high concordance between phenotypic cefoxitin resistance and molecular detection of *mecA* confirms the reliability of conventional laboratory methods used for MRSA identification.

Table 6 SCCmec typing was successfully performed for all *mecA*-positive MRSA isolates. SCCmec type III was the predominant genotype, accounting for 37.1% of isolates, followed by SCCmec type IV (31.4%). SCCmec type V represented 14.3% of isolates, whereas SCCmec types II and I accounted for 8.6% and 5.7%, respectively. Two isolates (2.9%) could not be assigned to any recognized SCCmec type and were classified as untypable.

Table 6. SCCmec Typing Distribution (n=70 mecA-positive isolates)

SCCmec Type	n	%
Type I	4	5.7
Type II	6	8.6
Type III	26	37.1
Type IV	22	31.4
Type V	10	14.3
Untypable	2	2.9

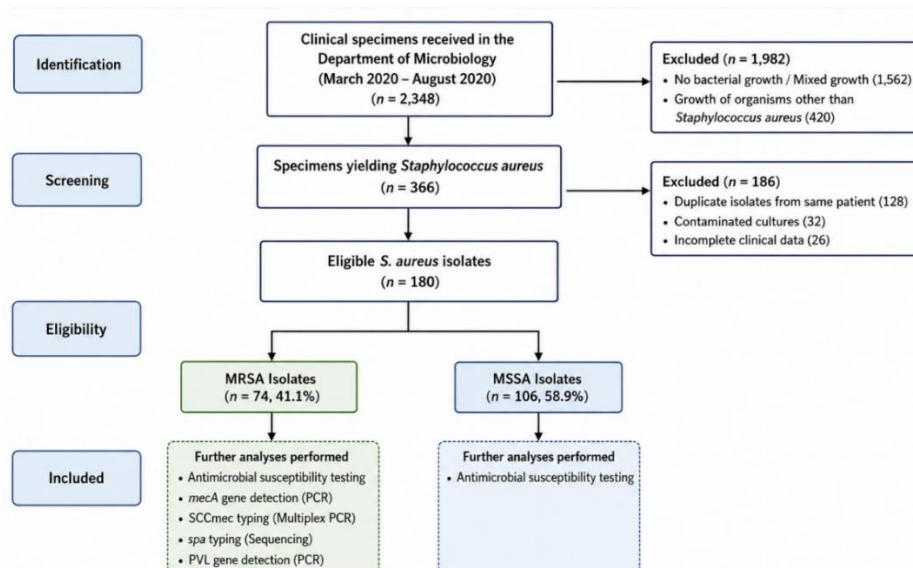


Figure 1. STROBE Flow Chart of study Participant and Isolates

Figure 1 The STROBE flow chart summarizes the selection process of clinical specimens and bacterial isolates included in the study. During

the study period (March 2020 to August 2020), a total of 2,348 clinical specimens were received in the Department of Microbiology for routine diagnostic investigations. After excluding specimens showing no bacterial growth, mixed growth, or growth of organisms other than *Staphylococcus aureus*, 366 *S. aureus* isolates were initially identified.

Subsequently, duplicate isolates from the same patient, contaminated cultures, and isolates with incomplete clinical information were excluded. Following application of the eligibility criteria, 180 non-duplicate *S. aureus* isolates were included in the final analysis.

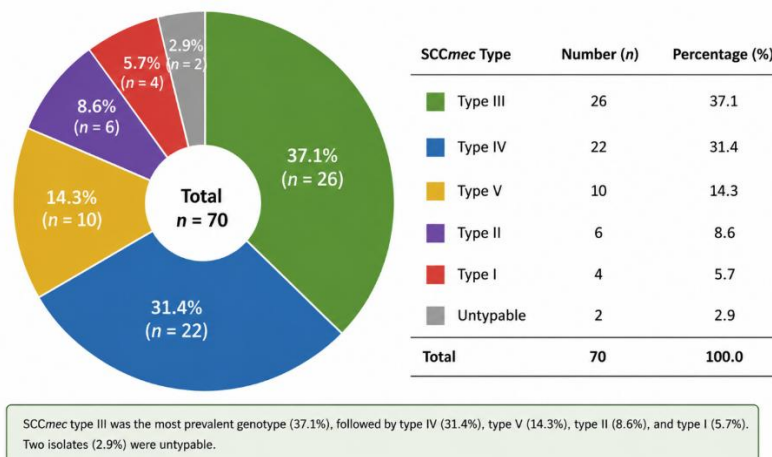


Figure 2. Distribution of SCCmec Types among mecA-Positive Methicillin-Resistant *Staphylococcus aureus* (MRSA) Isolates

Figure 2 illustrates the distribution of staphylococcal cassette chromosome mec (SCCmec) types among the 70 mecA-positive MRSA isolates recovered during the study period. SCCmec type III was the predominant genotype, accounting for 26 isolates (37.1%), followed by SCCmec type IV, which was identified in 22 isolates (31.4%). SCCmec type V was detected in 10 isolates (14.3%), while SCCmec type II and type I were identified in 6 (8.6%) and 4 (5.7%) isolates, respectively. Two isolates (2.9%) could not be assigned to any recognized SCCmec type and were therefore categorized as untypable.

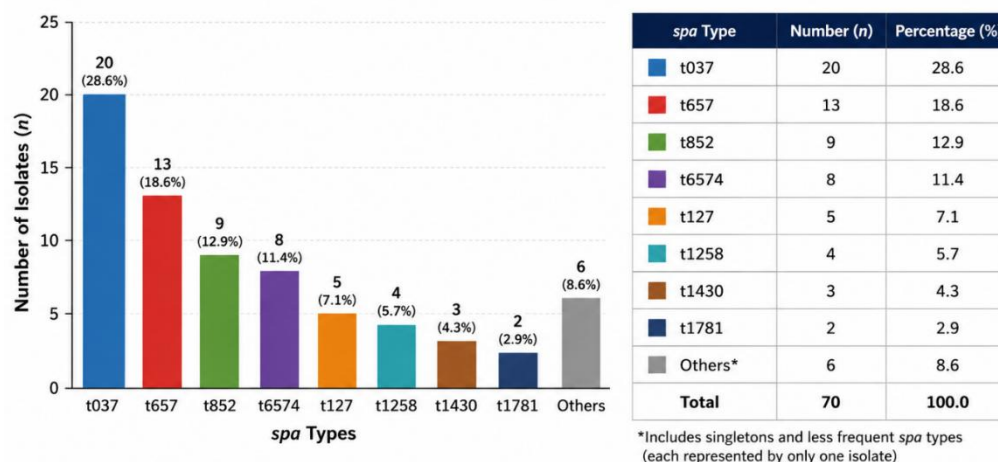


Figure 3. Distribution of Prevalent spa Types among Methicillin-Resistant *Staphylococcus aureus* (MRSA) Isolates (n = 70).

Figure 3 illustrates the distribution of spa types identified among 70 mecA-positive MRSA isolates recovered from clinical specimens during the study period. Spa typing revealed considerable genetic diversity, with a total of 14 distinct spa types detected among the isolates.

Discussion

The present study provides a comprehensive assessment of the molecular epidemiology of MRSA isolates recovered from a tertiary care center in Telangana, India. Among the 180 clinical *Staphylococcus aureus* isolates analyzed, 41.1% were identified as MRSA, indicating a substantial burden of methicillin resistance within the study setting. Molecular characterization revealed a high prevalence of mecA-positive isolates (94.6%), predominance of SCCmec type III, considerable spa type diversity, and the coexistence of both healthcare-associated and community-associated MRSA lineages. These findings contribute valuable regional data to the growing body of evidence regarding MRSA epidemiology in India.

The MRSA prevalence observed in the present study is consistent with previous reports from India, where prevalence rates have ranged from approximately 25% to 54% depending on geographic location, patient population, and healthcare setting [16,17]. The prevalence recorded in our study closely resembles findings from multicenter surveillance studies conducted across tertiary healthcare institutions in India, which have highlighted the persistent challenge posed by MRSA despite improvements in infection control practices [7,16].

Internationally, MRSA prevalence varies considerably. Lower prevalence rates have been reported in several European countries due to effective infection control programs and antimicrobial stewardship initiatives, whereas higher rates continue to be observed in many Asian and developing countries [18]. Such variations emphasize the influence of regional healthcare infrastructure, antibiotic prescribing practices, and local transmission dynamics on MRSA epidemiology.

The high proportion of mecA-positive isolates observed in the present study is comparable to findings reported from India and other Asian countries, where mecA remains the principal determinant of methicillin resistance [19]. The strong concordance between phenotypic and genotypic methods further validates the utility of ceftoxitin-based screening for routine MRSA detection.

One of the most important findings of the study was the predominance of SCCmec type III, which accounted for 37.1% of the mecA-positive MRSA isolates. SCCmec type III has historically been associated with hospital-associated MRSA (HA-MRSA) strains characterized by

multidrug resistance and prolonged persistence in healthcare environments [20]. Similar observations have been reported from several Indian studies where SCCmec type III remained the dominant genotype among hospital-acquired infections [9,20].

Interestingly, SCCmec type IV and type V collectively accounted for nearly half of the characterized isolates. Traditionally, these SCCmec types are associated with community-associated MRSA (CA-MRSA) strains that possess enhanced transmissibility and virulence but often exhibit lower levels of multidrug resistance [21]. The increasing detection of SCCmec IV and V isolates within hospital settings suggests an evolving epidemiological pattern in which community-associated strains are increasingly encountered in healthcare environments. Similar observations have been reported from Europe, North America, and Asia [22].

The coexistence of multiple SCCmec types within a single institution indicates substantial genetic heterogeneity among circulating MRSA strains. Such diversity may arise through repeated introductions of new clones, horizontal gene transfer, and ongoing evolutionary adaptation under antibiotic selection pressure [23].

Spa typing demonstrated the presence of 14 distinct spa types among the MRSA isolates, highlighting considerable clonal diversity. Spa type t037 emerged as the predominant lineage, accounting for 28.6% of isolates. This finding is consistent with previous reports from India and other Asian countries where t037 has been frequently associated with multidrug-resistant ST239-MRSA lineages [24]. The ST239-t037 clone is recognized as one of the most successful epidemic hospital-associated MRSA clones globally and has been implicated in numerous healthcare-associated outbreaks [25].

The detection of additional spa types including t657, t852, and t6574 suggests the circulation of diverse genetic backgrounds within the study population. Similar observations have been reported in molecular surveillance studies demonstrating the gradual replacement of traditional epidemic clones by emerging lineages possessing distinct resistance and virulence characteristics [26].

The observed spa diversity may reflect increasing patient mobility, inter-hospital referrals, and adaptation of community-associated strains to healthcare environments. Continuous monitoring of spa type distribution is therefore essential for identifying emerging clones and tracking transmission pathways within healthcare facilities.

The molecular findings of this study have important implications for infection prevention and control. The predominance of SCCmec type III indicates the persistence of endemic hospital-associated MRSA clones, suggesting ongoing nosocomial transmission within healthcare settings. At the same time, the substantial proportion of SCCmec IV and V isolates demonstrates that community-associated strains are increasingly contributing to hospital infections.

These observations underscore the need for strengthened infection control strategies, including active surveillance, hand hygiene compliance, contact precautions, environmental decontamination, and prompt identification of colonized or infected patients [27]. Molecular typing methods such as SCCmec and spa typing can facilitate outbreak investigations by identifying transmission networks and monitoring clonal dissemination within hospitals.

The coexistence of multiple MRSA lineages further emphasizes the importance of integrating molecular epidemiological surveillance into routine infection control programs. Such approaches allow healthcare institutions to detect shifts in circulating clones and implement targeted interventions before widespread dissemination occurs [28].

The antimicrobial susceptibility profile observed in the present study revealed extensive resistance to several commonly used antibiotics, including penicillin, erythromycin, and ciprofloxacin. These findings are consistent with the multidrug-resistant nature of hospital-associated MRSA clones and highlight the therapeutic challenges associated with their management [29].

Importantly, all isolates remained susceptible to vancomycin and linezolid, indicating that these agents continue to represent reliable treatment options for severe MRSA infections. Nevertheless, increasing reliance on last-line antimicrobials raises concerns regarding the potential emergence of glycopeptide- and oxazolidinone-resistant strains in the future [30].

The molecular epidemiological data generated in this study reinforce the need for robust antimicrobial stewardship programs aimed at optimizing antibiotic utilization, minimizing unnecessary antimicrobial exposure, and reducing selective pressure that drives the emergence and spread of resistant clones [31]. Combining molecular surveillance with stewardship initiatives can improve patient outcomes while simultaneously limiting the expansion of multidrug-resistant MRSA populations.

Overall, the findings of this study demonstrate a complex and evolving MRSA epidemiology characterized by the coexistence of healthcare-associated and community-associated clones, significant genetic diversity, and substantial multidrug resistance. Continuous molecular surveillance and evidence-based infection control interventions remain essential for mitigating the burden of MRSA in tertiary healthcare settings.

Conclusion

The present study provides important insights into the molecular epidemiology of methicillin-resistant *Staphylococcus aureus* (MRSA) in a tertiary care center in Telangana, India. A high prevalence of MRSA (41.1%) was observed among clinical *S. aureus* isolates, highlighting the continued burden of methicillin resistance in healthcare settings. Molecular characterization demonstrated a strong correlation between phenotypic methicillin resistance and the presence of the *mecA* gene, confirming the reliability of conventional screening methods for MRSA detection.

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