

Original Article

Prevalence of community-acquired methicillin-resistant *Staphylococcus aureus* in Taif social correctional center, Saudi Arabia

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Abstract

Introduction: Community-acquired methicillin-resistant strains of *Staphylococcus aureus* are primarily distinguished through their genetic characteristics. These strains carry the smaller types of staphylococcal cassette chromosome, specifically types IV and V. These infections occur mostly in healthy younger patients, and have been linked to such severe clinical conditions as necrotizing pneumonia and sepsis. A higher risk of methicillin-resistant *Staphylococcus aureus* contagion exists among incarcerated sub-populations; therefore, this study investigated colonization rate and risk factors among the residents of the Taif Social Correctional Center

Methodology: The study included 93 inmates and 19 employees. Specimens were collected from participants' noses and hands and from different environmental locations. The isolated organisms were identified according to standard microbiological methods. Methicillin resistance was evaluated using the standard cefoxitin disk diffusion method and oxacillin screen agar procedure. Methicillin resistance was further confirmed by multiplex polymerase chain reaction.

Results: High methicillin-resistant *Staphylococcus aureus* colonization rate was found among the center residents (24.7%) and employees (15.8%). Long duration of residence in the correctional institution and bad hand hygiene emerged as prominent risk factors for this colonization. An antibiogram categorized the isolated strains into six phenotypes, with a predominance of two antibiotic-resistant patterns suggesting cross-contamination and the presence of local foci of dissemination.

Conclusions: Taif Social Correctional Center residents exhibited a higher prevalence of methicillin-resistant *Staphylococcus aureus* colonization than was found in similar institutions. Poor personal hygiene and infection control measures are likely the major contributors to the problem.

Key words: Community-acquired; correctional institutions; methicillin-resistant *Staphylococcus aureus*.

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Introduction

Staphylococcus aureus is one of the most commonly isolated human pathogens in the clinical setting. This common pathogen, which colonizes the mucosal and/or skin surfaces of individuals of various age groups, is responsible for local soft tissue infections and systemic infections up to septicemia [1]. The notable ability of *S. aureus* to develop antimicrobial resistance constitutes a major public health problem. In the early 1940s, penicillin was an effective therapeutic agent for *S. aureus* infections; however, *S. aureus* soon developed a plasmid-encoded penicillinase enzyme that destroys the beta-lactam ring [2]. Since the mid-1990s, more isolates of *S. aureus* have shown an increasing range of antimicrobial resistance to the penicillinase-

resistant beta-lactams (such as methicillin and similar antibiotics) through harboring the *mecA* gene. Methicillin-resistant strains of *Staphylococcus aureus* (MRSA) were initially confined largely to hospital environments [2]. Subsequently, community-acquired MRSA (CA-MRSA) infections were reported among populations lacking access to healthcare systems. The Centers for Disease Control and Prevention (CDC) has defined CA-MRSA as “any MRSA infection diagnosed for an outpatient or within 48 hours of hospitalization if the patient lacks the following healthcare-associated MRSA (HA-MRSA) risk factors: hemodialysis, surgery, residence in a long-term healthcare facility or hospitalization during the previous year, the presence of an indwelling catheter or a percutaneous device at the

time of culture” [3,4]. CA-MRSA and HA-MRSA can be distinguished using a molecular-based analysis approach. CA-MRSA isolates carry the smaller staphylococcal cassette chromosome *mecA* (SCC *mecA*) elements (type IV or V), in comparison to HA-MRSA strains, which harbor the larger types of SCC *mecA* (types I, II, or III). Additionally, CA-MRSA strains commonly carry the genes for Pantone–Valentine leukocidin (PVL) [5].

CA-MRSA has been implicated in particular clinical syndromes among various subpopulations of the general public. Youthful and immunocompetent individuals who have no history of admission into healthcare settings are the most frequent victims of CA-MRSA infections [6]. Two conditions found to manifest in severe CA-MRSA infections are sepsis and the necrotizing type of pneumonia [7]. The standard infection control guidelines regarding MRSA prevention rely on the fact that, antibiotic-resistant pathogens are sensitive to routinely used disinfectants. Thus, optimum cleaning and disinfection of surfaces must be deployed throughout healthcare settings [6,8]. Specific interventions must be followed, such as the isolation of colonized individuals, active surveillance of MRSA colonization among high-risk populations, MRSA decolonization protocol, and environmental disinfection [9,10]. One effective protocol that was developed to attain full decolonization of MRSA comprises twice-daily intranasal application of mupirocin and chlorhexidine baths for 5 days. [11]. However, recent research has reported the development of bacterial resistance against both chlorhexidine and mupirocin in hospitals. However, as no detailed investigation of such resistance outside the hospital setting has yet been conducted, the protocol is still recommended in cases of MRSA colonization [9,12].

Many challenges are involved in controlling CA-MRSA in correctional facilities, such as inadequate housing, deficient hygiene standards and other infection control measures [13,14]. Federal Bureau of Prisons guidelines have discouraged routine attempts to eliminate MRSA colonization among U.S. inmates as a part of the standard guidelines for controlling MRSA infections. Two contexts have, however, been identified as warranting decolonization: recurrence or outbreaks of MRSA infections [6,14]. According to the guidelines, improving personal hygiene is the ultimate imperative. Recommendations include the use of soap and alcohol-based hand sanitizers. Moreover, educational campaigns, consciously urgent care, and wound clinics are possible interventions to prevent the spread of MRSA in incarceration settings [15,16].

Other guidelines recommend active surveillance after the diagnosis of a single case of MRSA and targeted examination of the infected inmate’s close contacts [17,18]. To the best of our knowledge, no published research has investigated CA-MRSA among the incarcerated population in Saudi Arabia. As a part of Taif University’s effort to achieve its vision for serving the local community, the main objective of this study was to investigate the prevalence and risk factors associated with the incidence of CA-MRSA colonization in the sub-population of the incarcerated residents of Taif Social Correctional Center (TSCC), aiming to improve control efforts and standardize the existing prevention protocols.

Methodology

Study design and population

This observational study was conducted in TSCC from September 2019 to April 2020. TSCC is a governmental institution for underage adolescents who have committed criminal acts, with a view to providing detention for care and psychological and social rehabilitation. The study was conducted in TSCC in cooperation with Taif University in accordance with the ethical consideration policy of both Taif University and TSCC. The study included 93 male inmates and 19 employees. In addition, 60 specimens were obtained to screen different TSCC environmental compartments, including food tables, bed linen, doorknobs, sinks, living room furniture, and shared sports equipment.

Specimen collection and identification

Demographic data and risk factors were collected via a structured questionnaire. Sterile cotton-tipped swabs were used to obtain samples from the subject’s nose (anterior nares) and hands. When obtaining environmental specimens, the swabs were moisturized with sterile saline. Swab samples were transported immediately to the clinical microbiology laboratory and plated onto blood agar, nutrient agar, MacConkey agar, and mannitol salt agar, and the growth was identified according to standard clinical microbiology procedures. *S. aureus* identification was confirmed phenotypically by a tube coagulation test and DNase production test. Methicillin resistance was evaluated using a standard oxacillin screen agar procedure where oxacillin screen agar plates (Becton, Dickinson and Co., BD Life Sciences, Maryland, USA) were spot inoculated with 10 μ L of trypticase soy broth suspension of *S. aureus* using a micropipette and incubated at 35 °C for 24 hours. Trypticase soy broth suspension turbidity was calibrated to a 0.5 McFarland turbidity standard [19]. *S.*

aureus isolates were also subjected to a cefoxitin (30 µg) disk diffusion test as described by the Clinical and Laboratory Standards Institute (CLSI). A 0.5 McFarland suspension was inoculated on Mueller–Hinton agar (MHA) plate and incubated at 37 °C for 18 hours. An inhibition zone diameter of ≤ 21 mm was reported as MRSA [19].

Molecular confirmation

MRSA was further confirmed as described in a previous study [20] by multiplex polymerase chain reaction (PCR). This multiplex PCR enabled *Staphylococcus* genus confirmation through detecting *16S rRNA* and identification of *S. aureus* species through detection of the *nuc* gene (to distinguish *S. aureus* from coagulase-negative staphylococci) while simultaneously detecting methicillin resistance through detection of the *mecA* gene. Table 1 provides a summary of the primer sets and PCR amplicon sizes. The amplification cycling conditions were as described before [20] briefly; initial denaturation at 95 °C for 5 minutes, 10 cycles of 94 °C for 30 seconds, 60 °C for 30 seconds, and 72 °C for 45 seconds, followed by 25 cycles of 94 °C for 30 seconds, 50 °C for 30 seconds, and 72 °C for 45 seconds, final extension step at 72 °C for 7 minutes. The PCR amplicons were visualized after electrophoresis on a 2% agarose gel stained with 0.5% ethidium bromide. Confirmed MRSA isolates were further subjected to antibiogram testing to sort the isolated strains into different antibiotic-resistant patterns.

Antimicrobial susceptibility

Antimicrobial susceptibility testing was carried out using the disk diffusion method on MHA (Becton, Dickinson and Co.) employing the following antibiotics (Oxoid, Thermo Fisher Scientific, Basingstoke, UK): penicillin (10 µg), oxacillin (30 µg), cefoxitin (30 µg), gentamicin (10 µg), erythromycin (15 µg), chloramphenicol (30 µg), tetracycline (30 µg), trimethoprim (5 µg), rifampicin, levofloxacin (5 µg), clindamycin (2 µg), ceftaroline (30 µg), and linezolid (30 µg). Susceptibility to teicoplanin and vancomycin was determined by the MIC method. The results were interpreted according to CLSI breakpoints M100-S16 [19]

Statistical analysis

Statistical analysis was performed by SPSS Version 20. Antibiogram frequencies were directly counted. The results of MRSA colonization of both TSCC residents and employees were analyzed by a chi-square test (χ^2).

A p -value < 0.05 was considered statistical significance.

Results

A total of 112 subjects (93 residents and 19 employees) were included in this study. The TSCC employees had a higher level of education than the residents, most of whom had less than a university education level. A statistically significant difference emerged between the hand hygiene of the TSCC employees and residents ($p = 0.009$). No significant difference in smoking rate was observed between the two groups, but diabetes was more prevalent among the TSCC employees. History of intake of immunosuppressive drugs revealed no significant difference between employees and residents ($p > 0.05$) (Table 2).

From the collected samples, 346 bacterial strains were isolated. The most prevalent was coagulase-negative staphylococci (125 isolates, 36.1%), followed by *S. aureus* (123 isolates, 35.5%). Forty-one *S. aureus* isolates were found to be MRSA, while 82 were methicillin-sensitive (Table 3). Twenty-three (24.7%) TSCC residents and three (15.8%) employees were colonized by MRSA. Moreover, 15 (25%) MRSA isolates were recovered from the TSCC environmental samples. The oxacillin screening agar method showed a lower MRSA detection sensitivity (95.1%) than the *mecA* PCR method, where 39 of 41 isolates were oxacillin (methicillin) resistant. Cefoxitin disk diffusion method results were identical to those obtained via *mecA* PCR, with 100% sensitivity and specificity. An antibiotic sensitivity test grouped the isolated MRSA strains into six distinctive patterns. The third phenotype displayed the most frequent pattern (Table 4). All isolates were resistant to penicillin, cefoxitin, and oxacillin (excluding the two hetero-resistant isolates on oxacillin screen agar that were classified as distinctive pattern 6). All MRSA isolates were susceptible to clindamycin, ceftaroline linezolid, teicoplanin, and vancomycin.

Discussion

The clinical spectrum of MRSA infection outcomes ranges from asymptomatic colonization to overwhelming fulminant infections [21]. The gold standard for confirmed MRSA diagnosis is *mecA* gene identification via PCR [22]. The current study also employed the cefoxitin disk diffusion method for the diagnosis of MRSA, with 100% sensitivity when compared to the PCR results.

Table 1. Primers set and PCR amplicons sizes of the multiplex PCR.

Gene target	Primer Sequence	Product size
Staph <i>16SrRNA</i>	F:AACTCTGTTATTAGGGAAGAACA R:CCACCTTCCTCCGTTTGTACC	756 bp
Nuc	F:GCGATTGATGGTGATACGGTT R:AGCCAAGCCTTGACGAATAAAGC	279 bp
<i>mecA</i>	F:GTGAAGATATACCAAGTGATT R:ATCAGTATTTCACCTTGTCG	112 bp

Table 2. The demographic data and risk factors of the study population.

Variable	Residents (n = 93)	Employee (n = 19)	p value
Age (Mean ± SD)	13 ± 4.2	30 ± 17	< 0.0001
Education:			
Less than high school	63 (67.7 %)	9 (47.4 %)	< 0.0001
High school	30 (32.3 %)	2 (10.5 %)	
University level	0	8 (42.1 %)	
Hand hygiene			
Infrequent	62 (66.7 %)	5 (26.3 %)	< 0.05
Average	23 (24.7%)	10 (52.6 %)	
Frequent	8 (8.6 %)	4 (21 %)	
Smoking:			
Yes	73 (78.5 %)	12 (63.2 %)	> 0.05
No	20 (21.5 %)	7 (36.8 %)	
Diabetes			
Yes	3 (3.2 %)	7 (36.8%)	< 0.0001
No	90 (96.8 %)	12 (63.2 %)	
Immunosuppressive Drugs intake:			
Yes	2 (2.2 %)	3 (15.8 %)	> 0.05
No	91 (97.8 %)	16 (84.2 %)	

Table 3. The prevalence of the isolated organisms from the collected specimens.

Organisms	Residents	Employee	Environment	Total
	n (%)	n (%)	n (%)	n (%)
<i>Staphylococcus aureus</i>	69 (37.1)	10 (34.5)	44 (33.6)	123 (35.5)
MRSA	23 (12.4)	3 (10.3)	15 (11.5)	41 (11.8)
MSSA	46 (24.7)	7 (24.1)	29 (22.1)	82 (23.7)
CoNS	71 (38.2)	15 (51.7)	39 (29.8)	125 (36.1)
<i>Escherichia coli</i>	29 (15.6)	1 (3.4)	23 (17.6)	53 (15.3)
<i>Klebsiella</i> spp	11 (5.9)	0 (0)	12 (9.2)	23 (6.6)
<i>Enterobacter</i> spp	2 (1.1)	1 (3.4)	4 (3.0)	7 (2.0)
<i>Enterococcus</i>	1 (0.5)	0 (0)	3 (2.3)	4 (1.2)
Others	3 (1.6)	2 (6.9)	6 (4.6)	11 (3.2)
Total	186	29	131	346

Table 4. The antibiogram of the isolated MRSA strains.

Antibiotic resistance pattern	Nose	Hand	Environment	Total
	(n =18)	(n =8)	(n =15)	
(1) PEN, OX, CEF, GN, TET, TS, RF	2	1	2	5
(2) PEN, OX, CEF, ERY, CHL, TET, TS, LEV	5	2	4	11
(3) PEN, OX, CEF, ERY, CHL, TET, TS, LEV	6	3	4	13
(4) PEN, OX, CEF, GN, ERY, TET, TS	4	1	3	8
(5) PEN, OX, CEF, GN, ERY, TET, TS	1	1	0	2
(6) PEN, CEF, ERY, CHL, TET, TS, LEV	0	0	2	2

PEN: penicillin; OX: oxacillin; CEF: cefoxitin; GN: gentamicin; ERY: erythromycin; CHL: chloramphenicol; TET: tetracycline; TS: trimethoprim; RF: rifampicin; LEV: levofloxacin.

The oxacillin screen agar method demonstrated lower sensitivity (95.1%). The aforementioned results are consistent with the findings of Anand *et al.* [23], who posited that the cefoxitin disk diffusion method could provide the best option for phenotypic identification of MRSA. The authors had a lower recommendation for the adoption of oxacillin screen agar due to the inability of oxacillin screen methods to detect hetero-resistant strains that possess the *mecA* gene with a low rate of expression of resistance [23]. The *mecA* gene expression can be enhanced by cefoxitin. Furthermore, the cefoxitin disk diffusion method can be used for phenotypic detection of the newly described MRSA harboring the *mecC* gene, which necessitates specific primers that differ from those used for *mecA*. In addition, *mecC*-containing MRSA produces PBP2a with a higher affinity for oxacillin than for cefoxitin [22].

Although incarcerated populations are at a higher risk of CA-MRSA contagion, few studies from this sub-population have focused on targeted surveillance and control efforts for CA-MRSA infections, even though outbreaks have been reported widely among many incarcerated sub-populations [6,24,25]. Furthermore, CA-MRSA has become an endemic pathogen and the predominant etiology of soft tissue infections and sepsis [26–29]. Studies set in the United States have reported high rates of CA-MRSA infections (12 per 1,000 prisoner/years). Among the risk factors identified in these studies were female gender, White race, and young age. Furthermore, the percentage of soft tissue infections caused by CA-MRSA increased dramatically from 29% in 1997 to 74% in 2012 among the incarcerated populations [30]. The results of the present study showed a high MRSA colonization among both TSCC residents and employees (24.7% and 15.8%, respectively). These results are in accordance with similar findings from Baltimore City (Maryland, USA) correctional institutions, where detainees showed a high MRSA nasal colonization rate of about 17% [31]. A previous Colombian study reported comparable results, finding 17.52% of the incarcerated population to be MRSA-colonized [23]. Meanwhile, a higher prevalence rate (35%) was observed in a study conducted in the Los Angeles County (California, USA) jail. That study suggested that the high prevalence of MRSA colonization reported might be attributed to the high-risk population group subjected to incarceration (e.g., low socioeconomic class, frequent drug abuse, and lack of shelter) rather than incident colonization during incarceration [32]. A previous study conducted in correctional institutions revealed that nasal MRSA

colonization was less prevalent (4.9%) among inmates. However, the colonization rate was higher in the female population compared to the male population, with rates of 5.9% and 2.5%, respectively [33]. Many risk factors have been reported for MRSA colonization and spread among incarcerated persons. As recommended in the CDC-conducted study, any inmate incarcerated for longer than 36 days should be targeted for MRSA decolonization [25].

In the current study, a long duration of residence in the correctional institution (more than 2 months) was recorded for the majority of the residents (71/93). These results are matched with a previous study in which long-term detainees had a higher (sevenfold) MRSA colonization rate compared to those incarcerated for a shorter time [25]. Statistical significance was found in the difference between TSCC employees and residents regarding hand hygiene ($p < 0.05$). Unsurprisingly, considering that this institution serves a youthful, most of the TSCC residents (63/93) had an educational level below high school. This low educational level coincides with a potential risk factor reported in a previous study [32]. Regarding other risk factors (smoking and immune-suppressive drug intake), no significant differences were observed between the incarcerated people and TSCC employees. However, diabetes was more prevalent ($p < 0.05$) among the employees, which could be explained by the older overall age of the employees compared to the residents. These studies reveal that the prevention of MRSA among inmates in incarceration institutions remains a challenging task [6,34].

Antibiogram (phenotyping) results of the 41 MRSA isolates (Table 4) showed six phenotypes with two predominant antibiotic-resistant patterns: phenotype 2 (11 isolates) and phenotype 3 (13 isolates). This clustering of a few phenotypes is highly suggestive of cross-contamination and the presence of local foci of MRSA dissemination. Corresponding results have been elicited in other studies done in correctional institutions in Maryland (USA) and Canada [35,36].

Conclusions

In conclusion, this study showed a higher occurrence of CA-MRSA colonization among TSCC residents than in other similar institutions. Long duration of incarceration, poor personal hygiene, and inadequate infection control measures are likely the major contributors to the problem. Improvement of infection control measures in addition to health education and awareness programs should be applied. The generalizability of this research is limited by its

small sample size of incarcerated residents. Further studies could be conducted on a larger scale to provide further insight into this health problem.

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