Biomedical and Biopharmaceutical Research

Abbreviation: Biomed. Biopharm. Res. Volume: 19: Issue: 02 | Year: 2022

Page Number: 78-90



Antibiotic Susceptibility Profiling of Methicillin-Resistant Staphylococcus aureus (MRSA) with Special Reference to Alternative Therapeutic Agents in atertiary care centre in Telangana Dr Shilpa HS

¹Asso professor Dept of Microbiology TRR Medical College

Corresponding Author

Dr Tabia Mir Dr Shilpa HS

¹Asso professor Dept of Microbiology TRR Medical College

Article Received:01-11-2022

Article Accepted:01-12-2022

©2022 Biomedical and Biopharmaceutical Research. This is an open access article under the terms of the Creative Commons Attribution 4.0 International License.

ABSTRACT

Background: Methicillin-resistant *Staphylococcus aureus* (MRSA) is a significant pathogen associated with hospital and community-acquired infections. It poses a major threat due to its resistance to multiple antibiotics, including β-lactams. Though vancomycin remains a cornerstone treatment, the emergence of intermediate and resistant strains necessitates exploration of alternative therapeutic agents such as linezolid, tigecycline, and quinupristin-dalfopristin. Objectives: Assess antibiotic susceptibility of MRSA, including alternative agents, vancomycin MIC, and mupirocin resistance. Methods: A prospective study was conducted over a 12-month period in the microbiology laboratory of a tertiary care hospital. A total of 304 Staphylococcus aureus isolates were obtained from clinical samples. Identification was done using conventional biochemical tests. MRSA was detected using the cefoxitin disc diffusion method. Susceptibility testing was performed using Kirby-Bauer disc diffusion for linezolid, tigecycline, and quinupristin-dalfopristin. Minimum inhibitory concentration (MIC) for vancomycin was assessed using E-test. Detection of inducible clindamycin resistance was done by the D-test. Results: Of 304 S. aureus isolates, 114 (37.5%) were MRSA. Most isolates were from pus (78.94%), followed by blood (12.28%). A male predominance was observed (71.92%). The most affected age group was 40-49 years. All MRSA isolates were sensitive to linezolid and tigecycline. Quinupristin-dalfopristin showed 93.86% sensitivity. MIC values for vancomycin ranged from 0.5 to 16 µg/mL; 5 isolates were VRSA. D-test was positive in 13 isolates (11.4%). Conclusion: Linezolid and tigecycline demonstrated excellent efficacy against MRSA. Vancomycin remains effective, though MIC creep and emergence of VRSA are concerning. Routine susceptibility testing and rational antibiotic use are essential to prevent resistance.

Keywords: MRSA, antibiotic resistance, vancomycin, linezolid, tigecycline, quinupristin-dalfopristin, MIC, cefoxitin, D-test..

INTRODUCTION

Staphylococcus aureus (S. aureus) is a virulent pathogen that is currently the most common cause of infections in hospitalized patients. It causes a broad spectrum of diseases, ranging from skin and soft tissue infections to endocarditis and fatal pneumonia. This pathogenicity is associated with various enzymes and toxins produced by the bacterium such as enterotoxins, exfoliative toxin, toxic shock syndrome toxin, and Panton-Valentine leucocidin (PVL) [1,2]. It also exists as a commensal, colonizing the anterior nares of about one-third of the healthy human population. Asymptomatic nasal carriers are at a higher risk of subsequent S. aureus infection. Carriers are presumed to be an important source of S. aureus that can spread and cause infection among contacts [3].

Although this bug has been naturally susceptible to almost every antibiotic developed so far, it frequently gains resistance by gene mutations and horizontal gene transfer [4]. Unlike penicillin resistance that results from a plasmid-encoded penicillin-degrading enzyme (β -lactamase), methicillin resistance is genetically mediated by staphylococcal cassette chromosome (SCCmec), a mobile genetic element encoding for an altered penicillin-binding protein (PBP2a, mecA) with a decreased affinity to β -lactams [5].

The emergence of MRSA was first described in the 1960s, and this has traditionally been regarded as a nosocomial pathogen endemic in hospitals and healthcare facilities in most countries [6]. Hospital-associated MRSA (HA-MRSA) characteristically colonizes or infects hospitalized individuals with predisposing risk factors such as surgery, presence of indwelling medical devices (IMDs), an immunocompromised state, or prior antibiotic exposure [6]. It is often isolated from cases of wound infections, vascular line-associated bacteremia, and ventilator-associated pneumonia. HA-MRSA strains usually harbour *SCCmec* types I, II, and III, and are multidrug-resistant (MDR) [7].

About three decades after the emergence of HA-MRSA, the organism spilled over into the community, and community-acquired strains (CA-MRSA) evolved either from the hospital strains through genetic changes or were the result of *mec* gene transfer to formerly susceptible subsets in the community [8]. True community-associated MRSA, infecting healthy individuals without any previous healthcare contact, was initially reported in the 1990s in Australia, followed by reports from the United States of America, and is now highly prevalent worldwide [4,6]. CA-MRSA infects healthy individuals without any healthcare contact, harbors smaller and more mobile *SCCmec* types (IV and V), is susceptible to non-β-lactam antimicrobial drugs, and typically manifests as skin and soft tissue infections. Life-threatening conditions, including severe necrotizing pneumonia, osteomyelitis, and fatal sepsis, have also been reported [7].

To effectively treat the infections caused by this organism, it is important to know the local antibiotic susceptibility pattern and prevalence of MRSA in our hospital. This study will also assess the sensitivity of MRSA to alternate antibiotics as well as evaluate the prevalence of mupirocin resistance among the locally isolated strains of MRSA.

OBJECTIVES

Assess antibiotic susceptibility of MRSA, including alternative agents, vancomycin MIC, and mupirocin resistance.

MATERIALS AND METHODS

Study Design and Duration

This was a prospective, cross-sectional study conducted over a period of 12 months in the Department of Microbiology at a tertiary care teaching hospital. Ethical clearance was obtained from the Institutional Ethics Committee prior to the commencement of the study.

Sample Collection and Identification

A total of 304 non-duplicate clinical isolates of *Staphylococcus aureus* were collected from various clinical specimens, including pus, blood, urine, sputum, ascitic fluid, pleural fluid, vaginal swabs, and catheter tips. Standard microbiological techniques were used for isolation and identification, including:

- Colony morphology
- Gram staining
- Catalase test
- Coagulase test (slide and tube)
- Mannitol fermentation on mannitol salt agar
- DNase test

Identification of MRSA

All *S. aureus* isolates were screened for methicillin resistance using the cefoxitin (30 μ g) disc diffusion method on Mueller-Hinton agar. A zone of inhibition of \leq 21 mm was interpreted as MRSA, according to CLSI 2022 guidelines.

Antibiotic Susceptibility Testing

Antimicrobial susceptibility testing of confirmed MRSA isolates was performed using the Kirby-Bauer disc diffusion method on Mueller-Hinton agar. The alternative therapeutic agents tested included:

- Linezolid (30 μg)
- Tigecycline (15 μg)
- Quinupristin-dalfopristin (15 μg)

Zone diameters were interpreted based on CLSI guidelines. The purpose of this testing was to assess the efficacy of alternative antibiotics against MRSA isolates.

MIC Testing for Vancomycin

The minimum inhibitory concentration (MIC) of vancomycin for MRSA isolates was determined using E-test strips on Mueller-Hinton agar supplemented with 2% NaCl. MIC breakpoints were interpreted as per CLSI:

- ≤2 µg/mL: Sensitive
- 4–8 μg/mL: Intermediate (VISA)
- ≥16 µg/mL: Resistant (VRSA)

D-Test for Inducible Clindamycin Resistance

Inducible clindamycin resistance was assessed using the D-zone test. Isolates resistant to erythromycin but sensitive to clindamycin were subjected to the double-disc diffusion method. Flattening of the clindamycin inhibition zone adjacent to the erythromycin disc indicated positive inducible resistance.

Inclusion and Exclusion Criteria

Inclusion Criteria

- All non-duplicate clinical isolates of *Staphylococcus aureus* from patients during the study period.
- Isolates confirmed as methicillin-resistant Staphylococcus aureus (MRSA) by the cefoxitin disc diffusion method.
- Isolates obtained from clinical specimens such as pus, blood, body fluids, and catheter tips.
- Patients of all ages and both sexes, from both inpatient and outpatient departments.

Exclusion Criteria

- Duplicate isolates from the same patient.
- Isolates identified as methicillin-sensitive Staphylococcus aureus (MSSA).
- Contaminated cultures or non-viable growth.
- Environmental samples or screening swabs not related to clinical infection.

RESULTS

Prevalence of MRSA: Out of 304 *S. aureus* isolates, 114 (37.5%) were confirmed to be MRSA using the cefoxitin disc diffusion method.



Figure 1: Colonies of Staphylococcus aureus on 5% sheep blood agar

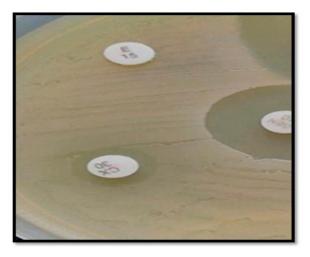


Figure 2: Cefoxitin disc diffusion test for phenotypic detection of MRSA

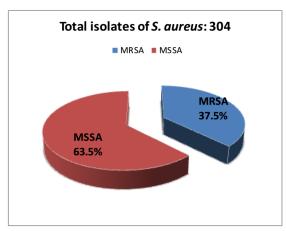


Figure 3: Proportion of MRSA isolated from clinical specimens. (MRSA: Methicillin resistant S. aureus, MSSA: Methicillin sensitive S. aureus)

Table 1: Gender-wise Distribution of MRSA (n = 114)

Gender	Frequency	Percentage (%)
Male	82	71.92%
Female	32	28.07%

Majority of MRSA cases were seen in males (71.92%).

Table 2: Age and Gender-wise Distribution of MRSA

Table 2011go and Gondon Wilso Distribution of Wilson			
Age (Years)	Male (82)	Female (32)	Total MRSA n (%)
0–9	3	5	8 (7.01%)
10–19	4	1	5 (4.38%)
20–29	9	7	16 (14.03%)
30–39	11	4	15 (13.15%)
40–49	24	4	28 (24.56%)
50-59	13	3	16 (14.03%)
60–69	14	6	20 (17.54%)
70+	4	2	6 (5.26%)

Majority of MRSA cases were seen in the 40-49 years age group with male predominance (71.9%).

Table 3: Sample-wise Distribution of MRSA Isolates

Clinical Sample	Number (n)	Percentage (%)
Wound/Exudate	90	78.94%
Blood	14	12.28%
Urine	4	3.5%
Sputum	3	2.63%
Vaginal Swab	1	0.87%
Pleural Fluid	1	0.87%
Ascitic Fluid	1	0.87%

The majority of MRSA isolates (78.94%) were obtained from wound or pus samples, indicating skin and soft tissue as the most common sites of MRSA infection. Blood was the second most common source (12.28%), followed by a few isolates from urine, sputum, and body fluids.

Table 4: Antibiotic Susceptibility Pattern of MRSA

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	51 (44.73%)	63 (55.27%)
Erythromycin	38 (33.34%)	76 (66.66%)
Clindamycin	84 (73.68%)	30 (26.31%)
Cefoxitin	0 (0%)	114 (100%)
Ampicillin	0 (0%)	114 (100%)
Amoxyclav	72 (63.15%)	42 (36.84%)
Cotrimoxazole	78 (68.42%)	36 (31.57%)

Tetracycline	88 (78.07%)	26 (21.93%)
Linezolid	114 (100%)	0 (0%)
Vancomycin	109 (95.62%)	5 (4.38%)
Teicoplanin	102 (89.47%)	12 (10.52%)
Ciprofloxacin	61 (53.51%)	53 (46.49%)
Gentamycin	46 (40.35%)	68 (59.65%)

All isolates were resistant to cefoxitin and ampicillin, confirming methicillin resistance. Highest sensitivity was observed for linezolid (100%), followed by vancomycin (95.62%) and teicoplanin (89.47%). Moderate resistance was noted to macrolides and fluoroquinolones.

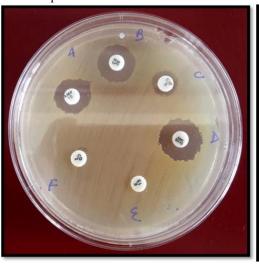




Figure 4: Antibiotic sensitivity tesing by Kirby Bauer disc diffusion method. A: Ampicillin, B: Cefoxitin, C:Ciprofloxacin, D:Amoxyclav, E: Erythromycin, F:Clindamycin, G: Teicoplanin, H: Cotrimoxazole, I: Tetracycline, J: Vancomycin, K:Gentamycin

Table 5: MRSA Susceptibility to Alternative Antibiotics

Antibiotics	Sensitive n (%)	Resistant n (%)
Linezolid	114 (100%)	0 (0%)
Tigecycline	114 (100%)	0 (0%)
Quinupristin-dalfopristin	107 (93.85%)	7 (6.14%)
Mupirocin*	108 (94.73%)	6 (5.26%)

Linezolid and tigecycline demonstrated 100% efficacy against all MRSA isolates, supporting their role as key alternative agents. Quinupristin-dalfopristin showed good sensitivity (93.85%). Although mupirocin was not part of the primary objective, it showed 94.73% sensitivity.

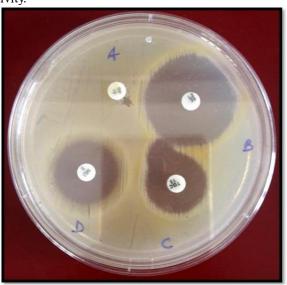


Figure 5: Antibiotic sensitivity testing of alternate antibiotics for MRSA A.Quinpristin-Dalfopristin B.Linezolid C.Tigecycline D.Mupirocin

Table 6: MRSA from Wound/Exudate Samples (n = 90)

Table 0. MKSA from Would/Exhibites (n – 70)			
Antibiotics	Sensitive n (%)	Resistant n (%)	
Azithromycin	38 (42.22%)	52 (57.77%)	
Erythromycin	32 (35.5%)	58 (64.44%)	
Clindamycin	63 (70%)	27 (30%)	
Cefoxitin, Ampicillin	0 (0%)	90 (100%)	
Amoxyclav	57 (63.33%)	33 (36.66%)	
Cotrimoxazole	63 (70%)	27 (30%)	
Tetracycline	67 (74.44%)	23 (25.55%)	
Linezolid	90 (100%)	0 (0%)	
Vancomycin	86 (95.55%)	4 (4.44%)	
Teicoplanin	81 (90%)	9 (10%)	
Ciprofloxacin	48 (53.33%)	42 (46.66%)	
Gentamycin	32 (35.5%)	58 (64.44%)	

Among the 90 MRSA wound isolates, the highest sensitivity was observed for linezolid (100%) and tigecycline, followed by vancomycin and teicoplanin. Resistance to beta-lactams and macrolides was high, similar to the overall pattern.

Table 7: Wound MRSA to Alternate Antibiotics

Antibiotics	Sensitive n (%)	Resistant n (%)
Linezolid	90 (100%)	0 (0%)
Tigecycline	90 (100%)	0 (0%)
Quinupristin-dalfopristin	86 (94.44%)	5 (5.55%)
Mupirocin	86 (95.55%)	4 (4.44%)

All wound-derived MRSA isolates were 100% sensitive to linezolid and tigecycline. Quinupristin-dalfopristin and mupirocin showed >94% sensitivity. These findings reinforce the value of alternative agents in MRSA treatment.

Table 8: MRSA from Blood Samples (n = 14)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	8 (57.14%)	6 (42.85%)
Erythromycin	2 (14.28%)	12 (85.71%)
Clindamycin	13 (92.85%)	1 (7.14%)
Cefoxitin, Ampicillin	0 (0%)	14 (100%)
Amoxyclav	10 (71.42%)	4 (28.57%)
Cotrimoxazole	7 (50%)	7 (50%)
Tetracycline	12 (85.71%)	2 (14.28%)
Linezolid, Vancomycin	14 (100%)	0 (0%)
Teicoplanin	13 (92.85%)	1 (7.14%)
Ciprofloxacin	5 (35.7%)	9 (64.28%)
Gentamycin	9 (64.28%)	5 (35.71%)

Of the 14 blood-derived MRSA isolates, most showed excellent sensitivity to clindamycin, linezolid, vancomycin, and teicoplanin. However, resistance to erythromycin and ciprofloxacin was high, indicating limited use of these agents in bloodstream infections.

Table 9: Blood MRSA to Alternate Antibiotics

Antibiotics	Sensitive n (%)	Resistant n (%)
Linezolid	14 (100%)	0 (0%)
Tigecycline	14 (100%)	0 (0%)
Quinupristin-dalfopristin	14 (100%)	0 (0%)
Mupirocin	13 (92.85%)	1 (7.14%)

All blood MRSA isolates (n=14) were 100% sensitive to linezolid, tigecycline, and quinupristin-dalfopristin. One isolate showed mupirocin resistance. The data supports the use of alternative agents in MRSA bacteremia.

Table 10: MRSA from Other Samples (n = 10)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	5 (50%)	5 (50%)
Erythromycin	4 (40%)	6 (60%)
Clindamycin	8 (80%)	2 (20%)
Cefoxitin, Ampicillin	0 (0%)	10 (100%)
Amoxyclav	5 (50%)	5 (50%)
Cotrimoxazole	8 (80%)	2 (20%)
Tetracycline	9 (90%)	1 (10%)
Linezolid	10 (100%)	0 (0%)
Vancomycin	9 (90%)	1 (10%)
Teicoplanin	9 (90%)	1 (10%)
Ciprofloxacin	8 (80%)	2 (20%)
Gentamycin	5 (50%)	5 (50%)

These included MRSA isolates from urine, pleural fluid, ascitic fluid, and sputum. Linezolid and vancomycin retained 100% and 90% sensitivity respectively. Resistance to beta-lactams was universal. Moderate sensitivity was observed to fluoroquinolones and tetracycline.

Table 11: Other MRSA to Alternate Antibiotics

Antibiotics	Sensitive n (%)	Resistant n (%)
Linezolid	10 (100%)	0 (0%)
Tigecycline	10 (100%)	0 (0%)
Quinupristin-dalfopristin	8 (80%)	2 (20%)
Mupirocin	9 (90%)	1 (10%)

Linezolid and tigecycline again showed 100% sensitivity. Quinupristin-dalfopristin and mupirocin had slightly lower effectiveness in this subgroup, with 80% and 90% sensitivity, respectively.

Table 12: Antibiogram of MRSA Isolates with Vancomycin MIC of $0.5 \mu g/mL$ (n = 2)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	0 (0%)	2 (100%)
Erythromycin	1 (50%)	1 (50%)
Clindamycin	2 (100%)	0 (0%)
Cefoxitin	0 (0%)	2 (100%)
Ampicillin	0 (0%)	2 (100%)
Amoxyclav	1 (50%)	1 (50%)
Cotrimoxazole	2 (100%)	0 (0%)
Tetracycline	1 (50%)	1 (50%)
Linezolid	2 (100%)	0 (0%)
Vancomycin	2 (100%)	0 (0%)
Teicoplanin	2 (100%)	0 (0%)
Ciprofloxacin	1 (50%)	1 (50%)
Gentamycin	1 (50%)	1 (50%)
Tigecycline	2 (100%)	0 (0%)
Quinupristin-dalfopristin	2 (100%)	0 (0%)
Mupirocin	2 (100%)	0 (0%)

These low-MIC isolates were susceptible to all alternate antibiotics. Moderate resistance was seen to first-line agents like azithromycin and cefoxitin.

Table 13: Antibiogram of MRSA Isolates with Vancomycin MIC of 1 μ g/mL (n = 85)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	38 (44.71%)	47 (55.29%)
Erythromycin	30 (35.29%)	55 (64.7%)
Clindamycin	65 (76.47%)	20 (23.53%)
Cefoxitin, Ampicillin	0 (0%)	85 (100%)
Amoxyclav	55 (64.7%)	30 (35.29%)

Cotrimoxazole	53 (62.35%)	32 (37.65%)
Tetracycline	66 (77.64%)	19 (22.36%)
Linezolid, Vancomycin	85 (100%)	0 (0%)
Teicoplanin	79 (92.94%)	6 (7.05%)
Ciprofloxacin	50 (58.82%)	35 (41.17%)
Gentamycin	28 (32.94%)	57 (67.05%)
Tigecycline	85 (100%)	0 (0%)
Quinupristin-dalfopristin	83 (97.64%)	2 (2.35%)
Mupirocin	84 (98.83%)	1 (1.17%)

Most common MIC category. Highest resistance observed to cefoxitin, ampicillin, and gentamycin. Alternate agents showed excellent sensitivity.

Table 14: Antibiogram of MRSA Isolates with Vancomycin MIC of 2 μ g/mL (n = 22)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	12 (54.55%)	10 (45.45%)
Erythromycin	6 (27.27%)	16 (72.73%)
Clindamycin	17 (77.27%)	5 (22.73%)
Cefoxitin, Ampicillin	0 (0%)	22 (100%)
Amoxyclav	14 (63.63%)	8 (36.31%)
Cotrimoxazole	19 (86.36%)	3 (13.36%)
Tetracycline	18 (81.81%)	4 (18.18%)
Linezolid, Vancomycin	22 (100%)	0 (0%)
Teicoplanin	21 (95.45%)	1 (4.54%)
Ciprofloxacin	8 (36.36%)	14 (63.63%)
Gentamycin	10 (45.45%)	12 (54.54%)
Tigecycline	22 (100%)	0 (0%)
Quinupristin-dalfopristin	22 (100%)	0 (0%)
Mupirocin	22 (100%)	0 (0%)

Vancomycin MIC creep evident. Excellent susceptibility retained for all alternate agents.

Table 15: Antibiogram of MRSA Isolates with Vancomycin MIC of 16 μ g/mL (VRSA) (n = 5)

Antibiotics	Sensitive n (%)	Resistant n (%)
Azithromycin	1 (20%)	4 (80%)
Erythromycin	1 (20%)	4 (80%)
Clindamycin	0 (0%)	5 (100%)
Cefoxitin, Ampicillin	0 (0%)	5 (100%)
Amoxyclav	3 (60%)	2 (40%)
Cotrimoxazole	4 (80%)	1 (20%)
Tetracycline	3 (60%)	2 (40%)
Linezolid, Tigecycline	5 (100%)	0 (0%)
Vancomycin	0 (0%)	5 (100%)
Teicoplanin	1 (20%)	4 (80%)
Ciprofloxacin	2 (40%)	3 (60%)
Gentamycin	2 (40%)	3 (60%)
Quinupristin-dalfopristin	0 (0%)	5 (100%)
Mupirocin	0 (0%)	5 (100%)

All 5 VRSA isolates were resistant to vancomycin, cefoxitin, mupirocin, and multiple other agents. Only linezolid and tigecycline maintained 100% effectiveness.

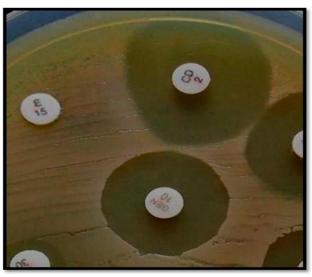
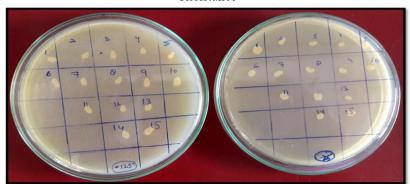
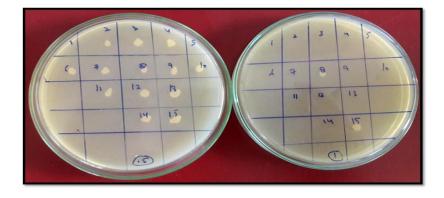
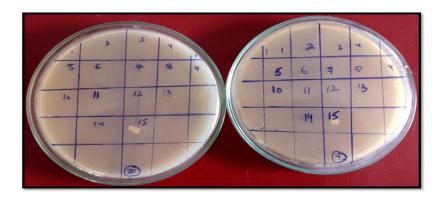


Figure 7: D-zone phenomenon by S. aureus due to inducible clindamycin Resistance







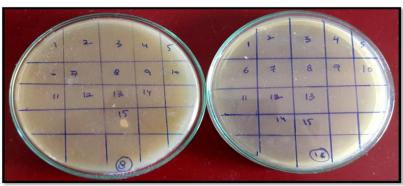


Figure 8: Minimum Inhibitory concentration of Vancomycin among MRSA isolates by Muller hinton agar dilution method

DISCUSSION

The global spread of MRSA constitutes one of the most serious contemporary clinical challenges encountered during the treatment of infections. Infections caused by MRSA are known to contribute significantly to the morbidity and mortality in hospitalized patients worldwide and have been associated with several hospital outbreaks since the late 1970s [8]. This study was designed to provide data on the isolation rate of MRSA from clinical samples in YMCH and to study their vancomycin MIC and sensitivity to alternate antibiotics. This will serve as a reference to strategize and develop a robust in-house antibiotic policy.

In the current study, 37.5% (114/304) of the S. aureus isolates were MRSA, detected using the cefoxitin disc diffusion method. This proportion aligns with national data reported from various regions of India, indicating MRSA prevalence typically ranging between 30% to 50% [9,10]. Comparatively, a pan-European study found 22.5% of SSTI isolates to be MRSA [11]. Globally, MRSA prevalence is highly variable, ranging from as low as 0.4% in Sweden to 48.4% in Belgium [11], and from 2% in the Netherlands to 70% in countries like Japan and Hong Kong [12].

In India, the MRSA isolation rate is reported to vary from 25% to over 80% depending on region, type of healthcare facility, and diagnostic protocols. Rajaduraipandi et al. (2006) observed a rate of 31% in Tamil Nadu [9], while Mohanty et al. (2004) reported 38.56% in Delhi [10]. A declining trend was noted in another Delhi-based hospital where the prevalence dropped from 51.6% in 2001 to 38.44% in 2008 [13]. Studies from tertiary care centres in India such as those by Anupurba et al. (2003) showed a prevalence of 54.82% [14], while Sangeeta et al. (2013) found an average prevalence of 42% across 15 centres in India, with 28% in OPD, 42% in ward, and 43% in ICU [15].

Alarming rates such as 80.89% in Indore [16], 66.84% in Bengaluru [17], 56.7% in Gulbarga [18], and 59.3% in Varanasi [19] have also been reported. Velasco et al. (2005) noted that nosocomial MRSA infections in India generally fall between 20–40% [20]. Differences can be attributed to multiple factors like sample type, patient demographics, type of hospital, antibiotic policies, and surveillance methods [21].

Most MRSA cases in our study were among adults aged 30–60 years (57.03%), consistent with findings from Moran et al. in the UK [22] and Yong Chen et al. in China [23]. This may be due to increased mobility, exposure, and social contact among adults. Male patients comprised 71.92% of MRSA cases compared to females at 28.07%. Male predominance in MRSA has also been reported in studies such as by Chua et al. in Detroit [24], and German studies which associated male gender with higher risk due to factors like diabetes, dialysis, and catheter use [25].

MRSA was most commonly isolated from patients admitted to surgical and medical wards (each 28.07%) followed by orthopaedics (15.78%). This trend aligns with studies by Sahai and Chauhan (2012) who reported highest MRSA

prevalence in medicine (41.7%), surgery (38.1%), and OBG wards (35.7%) [26]. Kaur et al. (2015) also observed high rates in OBG (33.33%), surgery (30.56%) and medicine (19.44%) [27].

Among the MRSA isolates, a majority (78.94%) were from exudates, which may reflect the predominance of wound and soft tissue infection samples received for culture. Other Indian studies also found high MRSA rates in pus/exudate samples: Anupurba et al. (70%) [14], Gayathri et al. (63%) [28], Lahari et al. (47%) [29], and Namita et al. (47%) [30]. Tripathi et al. found 36.18% of MRSA strains from pus, 33.33% from blood and sputum/throat swabs [31]. Mehta et al. similarly reported 33% MRSA isolation from pus [32] while Ringberg et al. concluded that throat carriage is a significant reservoir and suggested its use in screening [33].

Most MRSA isolates were sensitive to vancomycin (95.61%) and teicoplanin (89.47%). Sensitivity to other antibiotics was as follows: clindamycin (73.68%), tetracycline (78.07%), cotrimoxazole (68.42%), amoxicillin-clavulanate (63.15%), and ciprofloxacin (53.5%). Resistance was high against azithromycin (55.2%), erythromycin (66.66%), and gentamicin (59.64%).

Alternative drugs like linezolid and tigecycline showed excellent efficacy, with 100% sensitivity. Only 6.14% were resistant to quinupristin-dalfopristin. Perala et al. (2016) also found 90.9% sensitivity to linezolid and strong responses to levofloxacin and amikacin [34]. Joshi et al. reported low ciprofloxacin sensitivity: 53% in MSSA and 21% in MRSA [15]. Tripathi et al. observed resistance to gentamicin (100%), amikacin (71.42%) and erythromycin (57.14%), with universal sensitivity to glycopeptides [31]. Surpur et al. and Goyal et al. confirmed full sensitivity to linezolid and tigecycline [35,36].

The INSAR group in 2013 also found strong activity of linezolid and tigecycline against MRSA [37]. Khalili et al. in Iran also reported complete susceptibility to tigecycline [38]. Given their excellent oral bioavailability and broad Grampositive coverage, linezolid and tigecycline are promising alternative agents [3,4].

Although vancomycin remains the mainstay for MRSA treatment, evidence of increasing MICs or "MIC creep" has emerged globally [39]. In our MIC testing, 2 isolates had MIC of $0.5\mu g/ml$, 85 had MIC of $1\mu g/ml$, and 22 isolates had MIC of $2\mu g/ml$. Five isolates (4.38%) were confirmed as VRSA.

All VRSA isolates showed resistance to mupirocin and quinupristin-dalfopristin, and also to azithromycin, erythromycin, clindamycin, teicoplanin, and ciprofloxacin. However, they were fully sensitive to linezolid and tigecycline. A study by Thati et al. also showed high rates of resistance among VRSA isolates in ICUs [40].

With the increase in MRSA and VRSA cases, there is growing concern about therapeutic options. Hence, glycopeptides and linezolid should be reserved. For MSSA, de-escalation to beta-lactam antibiotics (such as cefazolin or oxacillin) is advised as they are more effective than vancomycin [41].

VRSA isolates were found to be multidrug-resistant, compromising treatment success and increasing the risk of morbidity and mortality. However, linezolid and tigecycline remain effective even against glycopeptide-resistant MRSA, supporting their use in reserve situations.

CONCLUSION

This study highlights the necessity of robust antibiotic stewardship programs. Linezolid and tigecycline remain potent agents for MRSA and VRSA infections, but their use should be preserved for confirmed resistant cases. The presence of vancomycin MIC creep and VRSA isolates is concerning and underscores the need for ongoing surveillance, rational antibiotic use, and strict infection control protocols.

REFERENCES

- Akpaka P, Monecke S, Swanston W, Rao A, Schulz R, Levett P. Methicillin sensitive Staphylococcus aureus producing Panton-Valentine leukocidin toxin in Trinidad & Tobago: a case report. J Med Case Reports 2011; 5:157
- 2. Moellering, Jr.R. Current Treatment Options for Community-Acquired Methicillin-Resistant Staphylococcus aureus Infection. Clin Infect Dis 2008; 46(7):1032-1037.
- 3. DeLeo F, Otto M, Kreiswirth B, Chambers H. Community-associated meticillinresistant Staphylococcus aureus. The Lancet. 2010; 375(9725):1557-1568.
- 4. Chambers H, Deleo F. Waves of resistance: Staphylococcus aureus in the antibiotic era. Nature Rev Microbiol 2009; 7: 629-41.
- 5. Lencastre H, Oliveira D, Tomasz A. Antibiotic resistant Staphylococcus aureus: a paradigm of adaptive power. Current Opinion Microbiol 2007; 10: 428-35.
- 6. Otter J, French G. Molecular epidemiology of community-associated meticillinresistant Staphylococcus aureus in Europe. The Lancet Infect Dis. 2010; 10: 227-39.
- 7. Gordon R, Lowy F. Pathogenesis of Methicillin resistant Staphylococcus aureus infection. Clin Infect Dis 2008; 46 Suppl 5: S350-9.
- 8. Cimolai N. Methicillin-resistant Staphylococcus aureus in Canada: a historical perspective and lessons learned. Can J Microbiol. 2010;56(2):89–120.

- 9Rajaduraipandi K, Mani KR, Panneerselvam K, Mani M, Bhaskar M, Manikandan P. Prevalence and antimicrobial susceptibility pattern of methicillin resistant Staphylococcus aureus: A multicentre study. Indian J Med Microbiol. 2006;24:34–8.
- 10. Mohanty S, Kapil A, Dhawan B, Das B. Bacteriological and antimicrobial susceptibility profile of soft tissue infections from Northern India. Indian J Med Sci. 2004;58:10–5.
- 11. Sader HS, Farrell DJ, Jones RN. Antimicrobial susceptibility of Gram-positive cocci isolated from skin and skin-structure infections in European medical centres. Int J Antimicrob Agents. 2010;36:28–32.
- 12. Fluit AC, Wielders CL, Verhoef JF, Schmitz J. Epidemiology and susceptibility of 3,051 Staphylococcus aureus isolates from 25 university hospitals participating in the European SENTRY Study. J Clin Microbiol. 2001;39:3727–32.
- 13. Tiwari HK, Sapkota D, Sen MR. High prevalence of multidrug-resistant MRSA in a tertiary care hospital of northern India. Infect Drug Resist. 2008;1:57–61.
- 14. Anupurba S, Sen MR, Nath G, Sharma BM, Gulati AK, Mohapatra TM. Prevalence of methicillin-resistant Staphylococcus aureus in a tertiary referral hospital in eastern Uttar Pradesh. Indian J Med Microbiol 2003; 21(1):49–51
- 15. Joshi S, Ray P, Manchanda V, Bajaj J, Gauatm V, Goswami P, et al. Methicillin-resistant Staphylococcus aureus (MRSA) in India: prevalence and susceptibility pattern. Indian J Med Res 2013; 137(2):363–9
- 16. Tahnkiwale SS, Roy S, Jalgaonkar SV. Methicillin resistance among isolates of Staphylococcus aureus: antibiotic sensitivity pattern and phage typing. Indian J Med Sci 2002; 56(7):330–4.
- 17. Yadhav K, Gayathri J, Panicker J. Prevalence and antibiogram of methicillin resistant Staphylococcus aureus isolates at a tertiary care hospital in Bangalore, South India. Int J Curr Res. 2014;6(17):37–40.
- 18. Chandrashekar DK, et al. Prevalence of Methicillin Resistant Staphylococcus aureus in a tertiary care hospital in Gulbarga, Karnataka. J Pharm Biomed Sci. 2012;19(19):1–3.
- 19. Arora S, Devi P, Arora U, Devi B. Prevalence of methicillin resistant Staphylococcus aureus (MRSA) in a tertiary care hospital in Northern India. J Lab Physicians 2010; 2(2):78–81.
- Velasco D, del Mar Tomas M, Cartelle M, Beciero A, Perez A, Molina F, et al. Evaluation of different methods for detecting methicillin (oxacillin) resistance in Staphylococcus aureus. J Antimicrob Chemother. 2005;55:379– 382.
- 21. Akpaka PE, Kissoon S, Swanston WH, Monteil M. Prevalence and antimicrobial susceptibility pattern of methicillin resistant Staphylococcus aureus isolates from Trinidad & Tobago. Ann Clin Microbiol Antimicrob. 2006;5(16):1–6.
- 22. Moran GJ, et al. Methicillin-Resistant S. aureus infections among patients in the emergency department. N Engl J Med. 2006;355:666–674.
- 23. Chen Y, Liu Z, Duo L, Xiong J, Gong Y, Yang J, et al. Characterization of Staphylococcus aureus from distinct geographic locations in China: an increasing prevalence of spa-t030 and SCCmec Type III. 2014;9(4):e96255.
- 24. Chua T, Moore CL, Perri MB, Donabedian SM, Masch W, et al. Molecular epidemiology of methicillin-resistant Staphylococcus aureus bloodstream isolates in urban Detroit. J Clin Microbiol. 2008;46:2345–2352.
- 25. Kupfer M, Jatzwauk, Monecke S, Möbius J, Weusten A. MRSA in a large German university hospital: Male gender is a significant risk factor for MRSA acquisition. GMS Krankenhaushygiene Interdisziplinär. 2010;5(2):11.
- 26. Sahai S, Chauhan S. Study of Methicillin-resistant Staphylococcus aureus in indoor patients of a tertiary care hospital in North India. Indian J Basic App Med Res. 2012;3(1):245–252.
- 27. Kaur DC, Chate SS. Study of antibiotic resistance pattern in methicillin resistant Staphylococcus aureus with special reference to newer antibiotics. J Glob Infect Dis. 2015;7(2):78–84.
- 28. Arakere G, Nadig S, Swedberg G, Macaden R, Amarnath SK, Raghunath D. Genotyping of methicillin-resistant Staphylococcus aureus strains from two hospitals in Bangalore, South India. Clin Microbiol. 2005;43(7):3198.
- 29. Lahari S, Nath R, Choudhury B, Sarkar M. Prevalence and antimicrobial susceptibility pattern of methicillin-resistant Staphylococcus aureus in Assam. Indian Crit Care Med. 2009;13:156–8.
- 30. D'Souza N, Rodrigues C, Mehta A. Molecular characterization of Methicillinresistant Staphylococcus aureus with emergence of epidemic clones of sequence type (ST) 22 and ST 772 in Mumbai, India. J Clin Microbiol 2010; 48: 1806-11
- 31. Tripathi A. Prevalence and antimicrobial susceptibility pattern of methicillin resistant Staphylococcus aureus in central India. Ind Med J. 2015;2(1):45–48.
- 32. Mehta AP, Rodrigues C, Sheth K, Jani S, Hakimiyan A, Fazalbhoy N. Control of methicillin resistant Staphylococcus aureus in a tertiary care centre a five year study. J Med Microbiol. 1998;16:31–4.
- 33. Ringberg H, Peterson AC, Walder N, Hugo Johanson PJ. The throat: an important site for MRSA colonization. Scand J Infect Dis. 2006;38(10):888–93.
- 34. Perala BMK, Koripella RL, Vani T, Lakshmi N. Prevalence of Methicillin Resistant Staphylococcus aureus in a Tertiary Care Hospital. J Dent Med Sci. 2016;29–31.

- 35. Surpur RR, Patil VM, Rao A, Hegadi S, Kalpana. Prevalence of Methicillin resistant Staphylococcus aureus and antibiotic susceptibility pattern among patients admitted at Navodaya Medical College, Hospital and Research Center, Raichur. Int J Recent Trends Sci Technol. 2013;9(1):152–154.
- 36. Goyal A, Diwakar MK, Bhooshan S, Goyal S, Agrawal A. Prevalence and antimicrobial susceptibility pattern of methicillin-resistant Staphylococcus aureus (MRSA) isolates in a tertiary care hospital in Agra, North India a systematic annual review. J Dent Med Sci. 2013;11(6):80–84.
- 37. Indian Network for Surveillance of Antimicrobial Resistance (INSAR) group, India. Methicillin-resistant Staphylococcus aureus (MRSA) in India: prevalence and susceptibility pattern. Indian J Med Res. 2013;137:363–369.
- 38. Khalili H, Dashti-Khavidaki S, Khaleghi SH, Maleki Z, Rasoolinejad M. Evaluation of tigecycline activity against methicillin-resistant Staphylococcus aureus isolated from biological samples. Iranian J Pharm Res. 2010;9(1):61–65.
- 39. Maclayton D, Suda K, Coval K, York C, Garey K. Case-control study of the relationship between MRSA bacteremia with a vancomycin MIC of 2 μg/mL and risk factors, costs, and outcomes in inpatients undergoing hemodialysis. Clin Ther. 2006;28(8):1208–1216.
- 40. Thati V, Shivannavar CT, Gaddad S. Vancomycin resistance among methicillin resistant Staphylococcus aureus isolates from intensive care units of tertiary care hospitals in Hyderabad. Indian J Med Res. 2011;134(5):704–708.
- 41. Liu C, Bayer A, Cosgrove SE, Daum RS, Fridkin SK, Gorwitz RJ, et al. Clinical practice guidelines by the infectious disease society of America for the treatment of methicillin resistant Staphylococcus aureus infections in adults and children. Clin Infect Dis. 2011;52:e18–e55.