

Original Research Article

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Prevalence and Antibiotic Resistance Pattern of Methicillin Resistant *Staphylococcus aureus* Isolates in a Tertiary Care Hospital

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ABSTRACT

Methicillin resistant *Staphylococcus aureus* (MRSA) is one of the most important etiology of community and hospital acquired infections. With an increasing incidence of Methicillin resistant *Staphylococcus aureus* (MRSA), the aim of the present study was to determine the prevalence of Methicillin resistant *Staphylococcus aureus* strains and their antibiotic susceptibility pattern in a tertiary care Hospital between July 2017 and July 2018. In this prospective cross sectional study, 100 *Staphylococcus aureus* were isolated and identified conventionally from various clinical specimens collected from different departments of the hospital. Subsequently, antimicrobial susceptibility test was performed by Kirby Bauer disc diffusion method as per Clinical and Laboratory Standards Institute (CLSI) guidelines. Among the 100 *S. aureus* isolates, MRSA was found to be 54% by cefoxitin (30 µg) disk diffusion method out of which maximum numbers were isolated from the age group of more than 60 years i.e. 14 isolates (25.92%) predominantly in males. Among the 54% of MRSA isolated, maximum 44.44% were from pus, 18.53% from blood, 11.11% each from sputum, urine and drain and 1.85% each from Ascitic fluid and tracheal aspirate. Among the MRSA isolated, 33.33% were from Surgery which was the highest followed by 27.78% from IMCU, 18.52% from Orthopaedics, 9.26% from Dermatology and 5.56% from Burns ward. All the isolates were resistant to Penicillin and Cephalosporin, followed by Tetracycline (22 isolates), Erythromycin (21 isolates), Clindamycin and Cotrimoxazole 19 isolates each, Gentamicin (18 isolates), Ciprofloxacin (17 isolates) and Amikacin (15 isolates). All the isolates were sensitive to Vancomycin, Linezolid and Teicoplanin. Inducible Clindamycin resistance was detected in 13 isolates (24%) among MRSA isolates. Due to increasing development of resistance to multiple antibiotics, there is an increased need to find out the prevalence of MRSA and their current antimicrobial profile in order to provide appropriate therapy to the patients.

Keywords

Methicillin resistant *Staphylococcus aureus* (MRSA) Community and Hospital acquired infections, Disk diffusion Method, Antibiotic susceptibility testing

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Introduction

The emergence of antibiotic resistance is a health problem worldwide and has affected the management and outcome of wide spectrum of infections. It contributes to significant mortality and morbidity and remains a hindrance to the control of infectious diseases. It leads to increase in health associated expenses and also acts as a barrier in the healthcare security of countries.

Globally, *Staphylococcus aureus* (*S.aureus*) is considered as one of the most common cause of nosocomial infections (Rajesh Bhatia, 2008). This remains as the hardiest of the non-sporing bacteria and can survive well in the environment under both moist and dry conditions. The high prevalence of *S.aureus*, together with its propensity to infiltrate tissues, colonize foreign material, form abscesses and produce toxins, makes it by far the most feared micro-organism in healthcare-associated infections.

In recent times, there is a steady rise in the number of *S.aureus* isolates that show resistance to Methicillin and has evolved as a serious problem since resistance to this drug indicates resistance to all β -lactam antibiotics. (Multiple use of antibiotics and prolonged hospitalisation are important factors which make hospital an ideal place for transmission and perpetuation of Methicillin Resistant *S.aureus* (MRSA) (Blot *et al.*, 2002). For these above reasons, accuracy and promptness in the detection of Methicillin resistance plays a key role for good prognosis of infections and hence abrupting its transmission (Chambers, 1997).

Hence the present study was undertaken to determine the prevalence of Methicillin resistant *Staphylococcus aureus* strains and their antibiotic susceptibility pattern in a tertiary care Hospital.

Materials and Methods

Study population

Inclusion criteria

Culture positive for *Staphylococcus aureus*. Only samples from hospitalized patients are included in the study.

Exclusion criteria

Samples from out patients are excluded from the study.

Samples from paediatric populations (less than 12 years)

Sample collection and processing

A total of 100 *Staphylococcus aureus* isolates from blood, urine, sputum, pus, wound swab, throat swab, nasal swab and endotracheal aspirates were taken for the study. All the samples were collected under aseptic precautions by standard procedures. They were then processed according to the standard guidelines. The specimens were inoculated on to nutrient agar (NA), MacConkey agar (MA), Blood agar (BA) and Mannitol Salt Agar (MSA). The media were incubated at 37^o C overnight. The growth was identified by its characteristic colony morphology, Gram staining (gram positive cocci in clusters) and Coagulase test positivity.

All the isolated *S.aureus* strains were subsequently tested for methicillin resistance based on Kirby-Bauer disk diffusion method on Muller Hinton Agar using Cefoxitin (30 μ g) disk obtained from Hi Media laboratories, Mumbai, India. The isolates were considered to be Methicillin resistant if the zone of inhibition is equal to 22 mm or less. Further, DD method was performed with the following antibiotic disks like Penicillin (10U), Cefoxitin (30 μ g), Erythromycin (15 μ g), Clindamycin (2 μ g), Gentamicin

(10µg), Amikacin (30µg), Vancomycin (30µg), Ciprofloxacin (5µg), Cotrimoxazole (1.25/23.75µg), Teicoplanin (30µg), Linezolid (30µg). Finally, the data were recorded and analysed at the completion of the study as per CLSI guidelines. *S.aureus* ATCC 25923 was used as a reference strain for the standardization of antibiotic susceptibility testing (CLSI, 2009).

Results and Discussion

Among 100 *S. aureus* isolates 46 isolates (46%) were MSSA and 54 isolates (54%) were MRSA (Table 1). Among the 100 isolates, maximum number of MRSA maximum numbers was isolated from the age group of more than 60 years i.e. 14 isolates (25.92%) followed by age group of 41-50 years i.e.11 isolates (20.37%). Males outnumbered the females in the rate of isolation of MRSA. The sex ratio (male: female) was 1.25: 1 for MRSA (Table 1 and 2).

Among the 54% of MRSA isolated,44.44% were from pus,18.53% from blood,11.11% each from sputum, urine and drain and 1.85% each from Ascitic fluid and tracheal aspirate (Table- 3). Among the 54% of MRSA isolated,33.33% were from Surgery which was the highest followed by 27.78% from IMCU,18.52% from Orthopaedics,9.26% from Dermatology, 5.56% from Burns ward and 1.85% each from Urology, Thoracic Medicine and Ophthalmology wards. There were no isolates from Nephrology ward (Table 4).

Antibiotic resistance pattern of MRSA strains

All the isolates were resistant to Penicillin and Cephalixin, followed by Tetracycline (22 isolates), Erythromycin (21 isolates), Clindamycin and Cotrimoxazole 19 isolates each, Gentamicin (18 isolates), Ciprofloxacin (17 isolates) and Amikacin (15 isolates). All the isolates were sensitive to Vancomycin, Linezolid and Teicoplanin (Table-5).

Inducible Clindamycin resistance among MRSA isolates which was found to be 13.A MRSA isolate is considered to be multi drug resistant if resistance was noted among 3 or more than 3 drugs. Among the MRSA isolates, 14 (25.93%) were resistant to more than or equal to 3 drugs, 12 isolates (22.22%)for 4 drugs, 5 isolates (9.26%) for 5 drugs and 7 isolates (12.96%) were resistant for more than 6 drugs.

Antibiotic resistance among *Staphylococcus aureus* have been increasing day by day. The outbreaks of nosocomial infections and emergence of antimicrobial resistance and its epidemiological complexity have made resistant strains of *S.aureus* a remarkable organism. They are now considered as one of the important nosocomial pathogens. Resistance pattern among this pathogen may vary widely from place to place even within the same country over time. Hence the early detection of Methicillin resistance is of prime importance in prevention of nosocomial outbreaks (Salgado *et al.*, 2003).

Table.1 Percentage of MSSA and MRSA among *Staphylococcus aureus* isolates

<i>Staphylococcus aureus</i>	Number	Percentage (%)
MSSA	46	46
MRSA	54	54
Total	100	100

Table.2.1 Analysis by age and gender

Age group (In years)	MRSA	
	Number	Percentage(%)
<20	3	5.56
21-30	8	14.81
31-40	9	16.67
41-50	11	20.37
51-60	9	16.67
>60	14	25.92
Total	54	100

Table.2.2 Distribution of MRSA among gender

<i>S.aureus</i>	Male		Female		Total
	Number	Percentage(%)	Number	Percentage(%)	
MRSA	30	55.56	24	44.44	54

Table.3 Percentage of MRSA isolated from clinical samples

Specimen	MRSA	
	Total	Percentage(%)
Pus	24	44.44
Sputum	6	11.11
Urine	6	11.11
Blood	10	18.53
Tracheal aspirate	1	1.85
Drain	6	11.11
Ascitic fluid	1	1.85
Total	54	100

Table.4 Percentage of distribution of MRSA in wards

Ward	MRSA	
	Total	Percentage(%)
IMCU	15	27.78
Surgery	18	33.33
Orthopaedics	10	18.52
Burns ward	3	5.56
Urology	1	1.85
Thoracic Medicine	1	1.85
Dermatology	5	9.26
Nephrology	-	-
Ophthalmology	1	1.85
Total	54	100

Table.5 Antibiogram of MRSA

Antibiotic	MRSA	
	Sensitive	Resistant
Penicillin	0	54
Cefoxitin	0	54
Erythromycin	33	21
Clindamycin	35	19
Gentamicin	36	18
Amikacin	39	15
Ciprofloxacin	37	17
Cotrimoxazole	35	19
Tetracycline	32	22
Cephalexin	0	54
Vancomycin	54	0
Linezolid	54	0
Teicoplanin	54	0

Among the 100 *S. aureus* isolated from clinical samples, 46% was found to be MSSA and 54% was found to be MRSA isolates which was comparable with the study conducted by Majumer *et al.*, from Assam, observed the prevalence of MRSA to be 52.9% Maximum number of MRSA were more than 60 years i.e.14 isolates (25.92%) followed by age group of 41-50 years i.e. 11 isolates(20.37%).Males outnumbered the females in the rate of isolation of MRSA. The sex ratio (male:female) for MRSA was 1.25:1. This was similar to the study by Sharma *et al.*, where males 30% constituted most of the MRSA isolates while 10% was from females.

Among the 54% of MRSA isolated, 44.44% were from pus,18.53% from blood,11.11% each from sputum, urine and drain and 1.85% each from Ascitic fluid and tracheal aspirate which was comparable to the study by Terry Ali *et al.*, where majority of the MRSA isolates were from pus (21.4%).

Among the 54% of MRSA isolated, 33.33% were from Surgery followed by 27.78% from

IMCU, 18.52% from Orthopaedics,9.26% from Dermatology and 5.56% from Burns ward which was comparable with the study by Arora *et al.*, who had found highest prevalence from surgical units (54.8%). Among the MRSA strains isolated, all the isolates were resistant to Penicillin and Cephalexin, followed by Tetracycline (22 isolates), Erythromycin (21 isolates), Clindamycin and Cotrimoxazole 19 isolates each, Gentamicin (18 isolates), Ciprofloxacin (17 isolates) and Amikacin (15 isolates). All the isolates were sensitive to Vancomycin, Linezolid and Teicoplanin. Inducible Clindamycin resistance was detected in 13 isolates (24%) among MRSA isolates.

This study highlights the prevalence of MRSA among clinical samples especially in hospitalized patients. Antibiogram of Methicillin sensitive and resistant isolates differs and susceptibility testing is mandatory for clinical isolates of *S. aureus* before initiation of treatment as few antibiotics exist for serious MRSA infections. An approach to eliminate MRSA in the hospitals and community needs to be integrated by creating

awareness among people and good hygienic practices and effective barrier precautions are to be adapted to prevent further transmission.

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