

# ISOLATION AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF METHICILLIN RESISTANT AND METHICILLIN SENSITIVE STAPHYLOCOCCUS AUREUS

REHANA AKHTER, KHALID MAHMOOD A KHAN, FURQAN HASAN

## ABSTRACT

*Objective* To find out frequency and antibiotic sensitivity pattern of methicillin resistant and methicillin sensitive staphylococcus aureus.

*Study design* Descriptive study.

*Place & Duration of study* Microbiology section of Department of Pathology in National Institute Of Child Health Karachi, from March 2008 to April 2009.

*Patients and Methods* All the clinical samples received in laboratory were processed. All staphylococcus aureus isolates were then identified by conventional method. The isolates were tested by the modified Kirby Baur disc diffusion method and results were interpreted as outlined by National Committee for Clinical Laboratory Standard criteria.

*Results* Eighty seven isolates of staphylococcus aureus were recovered from various clinical samples. Sixty six (75.86%) were isolated from various swabs and 21(24.13%) from blood. Twenty isolates (22.9%) were methicillin resistant. In this group high resistant was found to cloxacillin (100%), cephadrine (100%), co-trimoxazole (95%), erythromycin (70%), chloromycetin (65%), gentamicin (55%) and low resistance was observed to ciprofloxacin (30%).

In MSSA 0% resistance was seen to meropenem, ciprofloxacin and chloromycetin and high resistance found to co-trimoxazole (98.5%) and pencillin (73.13%). Low resistance was also observed with amoxiclav (20.89%). Both MRSA and MSSA were 100% sensitive to vancomycin.

*Conclusions* In this study high resistance pattern to non b-lactam antibiotic was found in MRSA leaving restricted choice for the treatment. Good hygiene practices, infection control and emphasis on hand washing etc may slow down the process of resistance.

*Key words* MRSA, MSSA, Antibiotic sensitivity pattern.

## INTRODUCTION:

Methicillin resistant staphylococcus aureus (MRSA) infection has increased over the years, becoming great threat in developing and non-developing countries. In

the past it was usually found in hospital setting but now it has moved out into the community.<sup>1</sup> MRSA can cause minor infection to serious ailment that sometimes become fatal.<sup>2</sup> Center for Drug Control report says that 20-30% population are colonized with staphylococcus aureus, although not all of them are infected. Approximately 1% are colonized with MRSA.<sup>3</sup>

## Correspondence:

Dr. Rehana Akhter  
Department of Pathology  
National Institute of Child Health  
Karachi.

MRSA are commonly seen among those who have weak immune system. Children have relatively underdeveloped

immune system. This population is at high risk for infection.<sup>3</sup> It includes patients in hospitals, nurseries and other health care centres and those who are on dialysis.<sup>4</sup> MRSA spreads by contact from person to person. One can get infection by touching another who has it on skin or by touching objects that have the bacteria on them.<sup>5</sup> It can survive on domestic animals such as dogs, cats and horses. It can cause bumble foot in chicken and mastitis in dairy cows. MRSA can be transferred from human to pets (dogs, cats, horses) and vice versa. An uninfected pet can cause infection to humans because it is present on their healthy skin.<sup>6,7,8</sup>

Incidence of MRSA is rising through out the world. In USA, MRSA causes 60% of staphylococcus aureus infections.<sup>9</sup> For methicillin different mechanisms are involved in its resistance. The most important is the production of unique penicillin-binding protein (PBP) with low affinity for b-lactam antibiotics and whose effects are determined by many structural genes (*mec*, *mecR1*, *mec1*). Other mechanism is production of PBPs, with modified affinities for b-lactam drugs, and hyperproduction of penicillinase enzymes, as cited by Fitzroy.<sup>10</sup> This study was undertaken to determine the frequency of methicillin resistant staphylococcus and drug resistance pattern of MRSA and MSSA (methicillin sensitive staphylococcus aureus) in a paediatric tertiary care hospital.

## METHODOLOGY

This study was carried out in Microbiology section of Department of Pathology at National Institute of Child Health Karachi over a period of 13 months from March 2008 to April 2009. This study was carried out on clinical samples received in the laboratory. All clinical samples were first collected in sterile container taking all aseptic measures. For blood samples, 2cc blood was collected in BHI broth and kept in incubator at 37°C. The specimen were inoculated on sheep blood agar, Mac-Conkey agar and blood chocolate agar and kept at 37°C in incubator for 24 hours, for seven days in case of blood culture. ATCC strain used for control was staphylococcus aureus 25923. All staphylococcus aureus isolates recovered from different specimens were identified by conventional method and confirmed by colony morphology, Grams staining, catalase, coagulase and mannitol fermentation test. All the isolates were then tested by the modified Kirby Bauer disc diffusion technique and results were interpreted as outlined by National Committee for Clinical Laboratory Standards criteria.<sup>11</sup> The antibiotic discs tested were penicillin(10), methicillin(10), amoxiclav (30), co-trimoxazole(25), cloxacillin(5), cephradine (30), meropenem(10), gentamicin(30), ciprofloxacin(5), erythromycin(15), chloromycetin (30) and vancomycin(30).

## RESULTS:

Eighty seven isolates of staphylococcus aureus were recovered from different clinical samples. Maximum samples were from various swabs sample and the breakup was, abscess 51, ear swab 3, wound 3, umbilical swab 2, pleural tap 2 and endotracheal tube 5. Out of 87 isolates, 21 were from blood (table I). All 87 isolates were analysed for antimicrobial susceptibility. Twenty (22.9%) isolates were identified as methicillin resistant. There was a marked difference in sensitivity pattern of MRSA (n 20 - 22.9%) and MSSA (n 67 - 77.1%). Penicillin resistance to MRSA was 100% while considerably high resistance was observed in MSSA (73.13%) too. By definition, all MRSA isolates were considered resistant to all b-lactam antibiotic. Of the MSSA isolates tested, b-lactam antibiotics and non b-lactam antibiotics exhibited low resistance to amoxiclav 20.89%, cloxacillin 2.98%, cephradine 1.49%, erythromycin 22.38%, gentamicin 20.89% and 0% resistance to meropenem, ciprofloxacin, chloromycetin. Of MRSA, ciprofloxacin 30% and chloromycetin 65%, erythromycin 70%, gentamicin 55% were resistant.. The greatest prevalence of resistant was found to co-trimoxazole in both MRSA (95%) and MSSA (98.5%). Fortunately vancomycin was 100% sensitive in both the groups. High resistance pattern has been observed in b-lactam antibiotics in MRSA as compared to MSSA (table II).

**Table I: Isolation of MRSA Staphylococcus aureus in various clinical specimens**

Clinical Samples	Staph aureus	MRSA	%
Abscess	51	8	15.68%
Ear Swab	03	1	3.33%
Wound	03	2	66.66%
Umbilical Swab	02	0	0%
Pleural tap	02	1	50%
Endotracheal tube	05	0	0%
Blood	21	8	38.09%

## DISCUSSION:

Methicillin resistant staphylococcus aureus has become an enormous problem for health care providers because it is hard to treat and is sometimes called super bug.<sup>9</sup> Multiple studies have been carried out on growing concern over multidrug resistance including Pakistan. MRSA is becoming a problem in pediatric population including hospital setting. The rate of prevalence of

**Table II: Antimicrobial Drug Resistance in Methicillin Resistant and (MRSA) and Methicillin Sensitive Staphylococcus aureus (MSSA)**

	MRSA (n=20)	Percentage	MSSA (n=67)	Percentage
Penicillin V	20	100	49	73.13
Amoxiclav	18	90	14	20.89
Co-trimoxazole	19	95	66	98.5
Cloxacillin	20	100	2	2.98
Cephradine	20	100	1	1.49
Meropenem	4	20	0	0
Gentamicin	11	55	14	20.89
Ciprofloxacin	6	30	0	0
Erythromycin	14	70	15	22.38
Chloromycetin	13	65	0	0
Vancomycin	0	0	0	0

MRSA isolates have increased over the years as reported by a study where they found 85.9% MRSA in 2003, decreasing to 57.8% in 2005% and again increasing to 90.8% in 2006.<sup>12</sup> In this study the frequency of MRSA is 22.9% which is less than that reported from Iran (35.3%), Karachi 43% and Lahore (63.64%).<sup>13,14,15</sup> Another study showed high rate of CA-MRSA (85%).<sup>16</sup> A study from Eritrea revealed low MRSA (9%) resistance.<sup>17</sup> Frequency of MRSA in this study is comparable with another study done in Kashmir-India, where they found 23.9% MRSA.<sup>18</sup>

MRSA isolates are often multidrug resistant and according to NCCL they are resistant to all b-lactam antimicrobials.<sup>11</sup> High resistant pattern was observed in MRSA isolates in present study. The greatest prevalence of resistance were seen with co-trimoxazole (95%) in MRSA and 98.5% in MSSA. Similar results have been observed by other authors where 100%, 97%, 77.2% resistance to co-trimoxazole is reported.<sup>15,19,20</sup> Another study showed 100% sensitivity to co-trimoxazole in MRSA.<sup>21</sup> This varies from place to place, time to time and depends upon some factors like self medication and empiric use of antibiotics. Our study revealed high resistance to widely used non b-lactam antibiotic erythromycin (70%) in MRSA. The prevalence was more than what was reported from Iran (27.7%) and less than what was reported from Pakistan (95%,89.7%) and Trinidad (86.7%).<sup>14,20,22</sup> Our result was close to 62.4% resistance to erythromycin, reported by Mehdinejad.<sup>23</sup>

In the present study, MRSA showed 65% chloromycetin, 55% gentamicin and 30% ciprofloxacin resistance, whereas MSSA showed 0% resistance to chloromycetin and ciprofloxacin and comparatively low resistance to gentamicin (20.89%). Geographical variation has been observed when comparatively higher rate of resistance to chloromycetin (93.33%) from India and lower rate (38.3%) from Pakistan were reported.<sup>19,20</sup> High rate of resistance to gentamicin (96.2%) was reported from Kashmir India and 93% and 97.8 % from Pakistan as compared to our study.<sup>14,18,20</sup> Our results are close to the study done in Assam India where 70.3% gentamicin resistance found in MRSA.<sup>24</sup>

Low resistance to ciprofloxacin in MRSA was reported by others authors (15% and 22.8% respectively),<sup>15,24</sup> as compared to our study (30%), while high resistance was observed in a study from Canada which was 87%.<sup>25</sup> In our study all MRSA and MSSA were 100% sensitive to vancomycin. Similar findings have been observed by many authors.<sup>16,18,19,20</sup> It shows that the glycopeptide agent vancomycin is currently the drug of choice for the treatment of life threatening infection caused by multidrug resistant MRSA. In our study MSSA showed higher rates of susceptibility to most of the antimicrobials except penicillin (73.13%) and co-trimoxazole (98.5%) to which they were resistant.

#### CONCLUSIONS:

In this study the frequency of MRSA is considerably high and it showed high resistance to b-lactam

antibiotics. Apart from adopting infection control practices, prevention and treatment of MRSA, awareness about this super bug in children should be promoted.

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