

## ANTIMICROBIAL RESISTANCE PATTERN OF *STAPHYLOCOCCUS AUREUS* ISOLATES FROM DAKSHINA KANNADA

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### ABSTRACT

Methicillin resistant *Staphylococcus aureus* (MRSA) is an important cause of infections in hospitals and pose a great challenge to the treating clinicians; even emergence of vancomycin resistance has been reported. Therefore the knowledge of prevalence of MRSA and their antimicrobial profile becomes necessary. This study is aimed to determine prevalence of MRSA and their antimicrobial sensitivity pattern in Dakshina Kannada.

Clinical specimens and carrier samples were cultured as per standard methods. The isolates were identified by using catalase test, coagulase tube test, mannitol fermentation and DNAase test. Antimicrobial susceptibility test was done for the isolates as per Kirby-Bauer disc diffusion method; the isolates were also tested for methicillin resistance using oxacillin and cefoxitin discs.

A total of 250 isolates were tested (200 clinical isolates and 50 from carriers) and 67 MRSA isolates were obtained (52 clinical samples and 15 from carriers). The degree of resistance to penicillin, ampicillin, ciprofloxacin, clindamycin and erythromycin were 100%, 100%, 53-56%, 14-16 % and 45-48% respectively. Resistance to vancomycin was not found. As the degree of resistance of MRSA towards antibiotics varies from region to region, *in vitro* susceptibility testing of every isolate of MRSA in clinical laboratories is inevitable.

**KEY WORDS:** MRSA, antibiotic, prevalence, carrier

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### INTRODUCTION

The antimicrobial chemotherapy for *Staphylococcus aureus* has been empirical as this species has overcome most of the therapeutic agents that have been developed in the recent years<sup>1</sup>. Resistance to methicillin was reported just one year after the launch of this antibiotic<sup>2</sup>. Many of these methicillin resistant *Staphylococcus aureus* (MRSA) isolates are multidrug resistant and are susceptible only to glycopeptide antibiotics such as vancomycin<sup>3</sup>. Emergence of vancomycin resistance also has been reported<sup>4</sup>. The development of resistance to multiple antibiotics and control of disease transmission by MRSA isolates in hospitals have been recognized as major challenges as the bacterial population that express the resistance phenotype varies according to environmental conditions<sup>2</sup>. Therefore the knowledge of

prevalence of MRSA and their current antimicrobial profile become necessary in the selection of appropriate empirical treatment of these infections. We determined the prevalence of MRSA in clinical specimens and carrier screening samples and their *in vitro* susceptibility pattern to various antimicrobial agents to record the current status of MRSA response to commonly used anti *Staphylococcal* antibiotics.

### MATERIALS AND METHODS

The study was conducted between April 2009 and January 2011 in our teaching hospital with a tertiary care facility having 700 beds. A total 250 isolates of *Staphylococcus aureus* were included in the study. Out of these 200 were obtained from various clinical specimens like pus, sputum, blood, fluids and urine, 50 from carrier screening samples from anterior nares.

The specimens were cultured on blood agar, MacConkey's agar and mannitol salt agar and incubated at 37°C for 24 hours. The colonies of Gram positive cocci in clusters were further confirmed for the production of free coagulase enzyme, mannitol fermentation and DNAase according to standard methods<sup>5</sup>.

All the confirmed *Staphylococcus aureus* strains were subsequently tested for methicillin resistance based on Kirby-Bauer disc diffusion method using oxacillin discs (1µg) and ceftioxin (30µg) discs [Hi Media, Mumbai]. The Mueller Hinton agar plates with lawn culture of the isolates (equivalent to 0.5 McFarland turbidity standards) on which oxacillin and ceftioxin discs were applied; incubated aerobically for 24 hours at 37°C. Isolates with inhibition zone of  $\leq 10$ mm and  $\leq 19$  mm around oxacillin and ceftioxin discs respectively were considered MRSA strains. Further the susceptibility pattern of MRSA was determined by modified Kirby-Bauer disc diffusion method and results interpreted as per CLSI criteria<sup>6</sup>. The antimicrobials tested were penicillin, ampicillin, erythromycin, clindamycin, ciprofloxacin and vancomycin. *Staphylococcus aureus* ATCC 29213 was used as control strain for antimicrobial susceptibility testing. All antibiotic discs and culture media were procured from Hi Media Mumbai, India.

## RESULTS

A total number of 250 *Staphylococcus aureus* strains were tested of which 200 and 50 were from clinical specimens and carrier screening samples respectively. The test report on the methicillin resistance by these isolates confirmed the presence of 67 MRSA (52 from clinical specimens and 15 from carrier screening samples) and the remaining were considered as methicillin sensitive *Staphylococcus aureus* (MSSA). The prevalence rate of MRSA was 26% among patient samples and 30 % among carrier samples.

The antimicrobial resistance pattern of MRSA from clinical samples was almost similar to that of carrier sample isolates (**Table 1**). However all isolates from both categories were sensitive to vancomycin.

## DISCUSSION

MRSA infections causing significant morbidity and mortality are well documented<sup>7</sup>. The important reservoirs of MRSA in hospitals are infected patients and transient nasal carriers among hospital staff<sup>8</sup>. In India, the MRSA infections emerged as a problem in the 80s and 90s. The MRSA isolates are often resistant to several other antibiotics.

In the past 15 years, the appearance and world wide spread of many such isolates have posed major therapeutic challenges in many hospitals and have led to diversion of considerable resources to attempts at controlling their spread<sup>9</sup>.

Studies show that epidemiology of MRSA is not uniform in different parts of India and also has variations with time at the same hospitals<sup>10</sup>. We isolated 52 and 15 MRSA strains from 200 clinical samples and 50 carrier screening samples respectively. This amounts to a prevalence rate of 26% among patients and 30% among carriers. Various investigators have reported a prevalence rate of MRSA ranging from 26 % to 51.6 % in India<sup>3, 4</sup>. A significant observation in our study was the increased isolates of MRSA from carrier screening samples. Literature shows a highly variable carrier rate ranging from 0% to 29%<sup>7, 3, 10</sup>. The carrier rate was found to be 30 % in our study which is similar to other reports<sup>11</sup>. All the strains were resistant to penicillin and ampicillin. Resistance to clindamycin was 14 -16%, erythromycin 45- 48 % and ciprofloxacin 53.5 - 56%.

The other contemporary reports state higher resistance rate for fluoro-quinolones, with two studies reporting 90% to 98.9%<sup>2, 10</sup>. In contrast, we have 53.5– 56% of the strains resistant to ciprofloxacin. We observed that the resistance rates of carrier strains were not much different from that of hospital patient strains. This is in contrast to other studies where higher susceptibility rate was found among the strains obtained from carrier screening samples<sup>12</sup>.

## CONCLUSION

The degree of resistance of MRSA towards commonly used antimicrobials is recognized to be diverse from region to region. Uniform sensitivity was shown only to vancomycin as 100 %. It is inevitable to do the *in vitro* susceptibility testing of every isolate of MRSA in clinical laboratories before choosing the antibiotic for therapy.

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**Table 1:** ANTIBACTERIAL RESISTANCE PATTERN OF THE *STAPHYLOCOCCUS AUREUS* ISOLATES

Antibiotic disc	Number of resistant isolates with percentage (From patients) n(%)	Number of resistant isolates with percentage (From carriers) n(%)
Penicillin(10 units)	200 (100%)	50 (100%)
Ampicillin(10µg)	200 (100%)	50 (100%)
Oxacillin (1µg)	52 (26%)	15 (30%)
Cefoxitin (30µg)	52 (26%)	15 (30%)
Vancomycin (30µg)	00 (0 %)	00 (0%)
Clindamycin (2µg)	28(14%)	08 (16%)
Erythromycin (15µg)	90(45%)	24 (48%)
Ciprofloxacin (10µg)	107(53.5%)	28 (56%)

n = number of resistant isolates

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