A study on the prevalence of Methicillin Resistant Staphylococcus aureus (MRSA) isolates from various clinical samples in a tertiary care hospital

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Abstract
Background: Staphylococcus aureus is the most common cause of hospital and community acquired infections. The increasing prevalence of Methicillin Resistant Staphylococcus aureus (MRSA) has led to the widespread usage of Vancomycin which in turn leads to the emergence of resistance to glycopeptides antibiotics. Hence this study is aimed at analyzing the prevalence of Methicillin Resistant Staphylococcus aureus isolates from various clinical specimens.
Methodology: A retrospective study was conducted over a period of one year in a tertiary care hospital from various clinical samples received during the study period. Staphylococcus aureus isolates were identified by phenotypic methods and MRSA isolates were identified by Cefoxitin disk diffusion method as per CLSI guidelines.
Results: Out of 10842 different types of samples processed, 212 isolates of Staphylococcus aureus were identified. Among them 59 (27.83%) were found to be Methicillin Resistant Staphylococcus aureus (MRSA) and 153 (72.17%) were found to be Methicillin Sensitive Staphylococcus aureus (MSSA).
Conclusion: The above study concludes that it is a must to implement appropriate diagnostic methodology for isolating MRSA strains and also to ensure strict antibiotic policy in all the healthcare institutions with precautionary measures before starting the patient on Vancomycin.

Keywords: Staphylococcus aureus, MRSA, Vancomycin, MSSA, CLSI.

Introduction
Staphylococcus aureus is the most common cause of hospital and community acquired infections. It has been reported that the incidence of MRSA, in an endemic country like India is between 25 to 50% based on geographic location¹. Studies have also shown that one in three people (³3%) carry Staphylococcus aureus in their nose, usually without any illness but these could be constant sources of infection to other people. CDC estimated that two in hundred people (2%) carry MRSA strain². In another study which compiled data from 15 Indian tertiary care centers over a period of two years, the overall prevalence of MRSA was 41%³. However, there is no proper statistical data in many tertiary care centers about the prevalence of MRSA as well as protocol for screening MRSA carrier state. In epidemiological point of view, the one more disturbing statistic with regards to MRSA is Asia represents one among the regions with the highest prevalence rates of healthcare-associated Methicillin Resistant Staphylococcus aureus (HA-MRSA) and community-associated Methicillin Resistant Staphylococcus aureus (CA-MRSA) in the world⁴. CA-MRSA is another potential threat, because the so-called resistant strains have been isolated from community itself, which were initially constrained to the health care associated areas. Thus it increases the burden of MRSA strains, its spread and the way to treat those resistant strains. The accuracy of prevalence of MRSA is a must to implement strict antibiotic policy and to overcome the resistance, constant surveillance should be done on all isolates with appropriate diagnostic methodology. Hence this study was conducted to estimate the prevalence of Methicillin Resistant Staphylococcus aureus (MRSA) isolates from various clinical specimens.

Materials and Methods
The study was a retrospective study conducted over a period of one year in a tertiary care hospital. Staphylococcus aureus isolates from various clinical samples like pus, blood, wound swab, throat swab, vaginal swab and urine received during the study period (October 2014 to October 2015) were included in the study.

Out of 10842 clinical samples received and processed, identification of Staphylococcus aureus was done by the morphology of the colonies, pigment production, catalase test, slide and tube coagulase test, urease test and mannitol fermentation. MRSA isolates were identified by cefoxitin (30μg) disk diffusion method. The suspension for inoculation was prepared...
from the colonies from an overnight growth on nutrient agar plate. A suspension of the overnight growth was prepared with 0.5 ml of sterile saline and the turbidity was adjusted to 0.5 McFarland’s units. A sterile swab dipped into this suspension and the excess of inoculum was removed by pressing it against the sides of the tube. The swab was then inoculated on Mueller-Hinton agar plate. Cefoxitin disc (Hi-Media, Mumbai) was applied within 15 min after the inoculation. The plates were incubated for 24hrs at 37°C. The diameter of the zone around the disc was measured and the results were interpreted according to the CLSI guidelines. The isolates with a zone of inhibition less than 22mm were reported as MRSA strains. *Staphylococcus aureus* ATCC 25923 was used as the control strain\(^4\).

**Results**

Out of 10842 samples processed over a period of one year, 212 number of *Staphylococcus aureus* was isolated. Among them 59 (27.83%) were found to be Methicillin Resistant *Staphylococcus aureus* (MRSA) and 153 (72.17%) were found to be Methicillin Sensitive *Staphylococcus aureus* (MSSA).[Fig. 1]

![Distribution of S.aureus (MSSA and MRSA)](image1)

**Fig. 1: Distribution of S.aureus (MSSA and MRSA)**

The age distribution of the study population was analyzed and it showed that highest number of MRSA isolates was identified in the age group of 51 to 60 years (34.09%) followed by 21 to 30 years (29.78%).\((\text{Fig. 2})\)

![Distribution of isolates with respect to age](image2)

**Fig. 2: Distribution of isolates with respect to age**

The distributions of sex among the isolated sample were studied and it showed 116 (54.72%) were male and 96 (45.28%) were females among the study population. Among the males, 86 (74.14%) presented with MSSA infections and 30 (25.98%) presented with MRSA infections. Among the females, 67 (69.8%) presented with MSSA infections and 29 (30.2%) presented with MRSA infections. \((\text{Fig. 3})\)
The isolates were analyzed with respect to the clinical samples and it showed the following findings. 44 isolates of MRSA from pus, 9 from wound swab, 3 from high vaginal swab, 1 from sputum, 1 from tracheal swab and 1 from Cerebrospinal fluid (CSF) were isolated. 

![Figure 3: Distribution of isolates with respect to sex](image)

**Discussion**

MRSA is a major nosocomial pathogen which causes significant morbidity and mortality in all health care levels. The importance of detecting MRSA strains is mainly attributed to its colonization in patients and health care workers, which serves as a constant source of infection and for transmission. Even though MRSA isolates were identified as early as 1960's, the significance of MRSA in India is still under process and the need to screen all the *Staphylococcus aureus* isolates must be implemented at all health care levels. In addition to routine diagnostic services, there is a must to screen all the health care workers for the colonization at a periodic basis also. It is also advisory to communicate with the hospital personnel regarding the prevalence of MRSA in respective departmental infectious control officers on a periodic basis.

In our study, the prevalence of MRSA isolates obtained from different clinical samples was determined and its correlation with age, sex and the clinical materials were studied. Out of 10842 samples processed over a period of one year, 212 number of *Staphylococcus aureus* was isolated. Among them 59 (27.83%) were found to be MRSA and 153 (72.17%) were found to MSSA. Our study states that majority of the isolates obtained were MSSA and hence the clinicians must be warned not to use higher level of antibiotics like glycopeptides without the appropriate
screening results. Inadvertent use of higher antibiotics leads to emerging drug resistant strains like VISA (Vancomycin intermediate susceptibility Staphylococcus aureus) and VRSA (Vancomycin Resistant Staphylococcus aureus). On analysis of clinical samples, 74.58% of the MRSA isolates were obtained from pus samples which indicate the need to screen those surgical wards, surgical health care workers. Similar observation was made by Qureshi et al(6) from Pakistan who has isolated 83% of MRSA isolates from pus and Fitzroy A orrett et al(7). But many other studies have showed varied numbers of MRSA prevalence due to variation in region and antibiotic usage.

**Conclusion**

The emergence of resistance to vancomycin is a threat to the already challenging therapy of MRSA. So it is must to implement antibiotic policy in all the health care institutions and precautionary measures should be ensured before starting the patient on Vancomycin. The clinicians should seek advisory from microbiologists to determine the Methicillin susceptibility of such strains from suspected patients and suspected carriers, so that the emergence of vancomycin resistance can be prevented. In addition, all the clinical microbiology laboratories should routinely do testing for MRSA strains for appropriate treatment of patients and for implementation of infection control measures to prevent the emergence of resistance.

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**Bibliography**


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