Vancomycin Susceptibility Pattern of Methicillin Resistant *Staphylococcus aureus* Isolates

Venkatakrishna Rao Illuri, Kishore G Bhat, Vidya Pai, Sunil Rao Padmaraj, Manjula Shantaram

**Background:** The antimicrobial chemotherapy for *Staphylococcus aureus* infections has been empirical as this species has overcome most of the therapeutic agents. *S. aureus* is among the most common organisms in hospital and community acquired infections. In recent years, a large and continuing increase in the prevalence of methicillin resistant *S. aureus* (MRSA) has been observed. In addition, the emergence of *S. aureus* with reduced susceptibility or resistance to vancomycin also has been reported.

**Objective:** To evaluate vancomycin susceptibility in clinical isolates of Methicillin resistant *Staphylococcus aureus* (MRSA) for a period of three years (2009-2011) in a tertiary care hospital.

**Materials and Methods:** A total of 400 clinical isolates of *S. aureus* from in-patients (pus, sputum, blood and body fluids) at Yenepoya University Hospital from July 2009 to June 2011 were included in the study. The specimens from the patients were cultured on blood agar. The isolates were identified by Gram’s stain, catalase test, mannitol fermentation test, free coagulase and deoxyribonuclease test according to standard techniques. Antibiogram and minimum inhibitory concentration (MIC) of all the MRSA isolates for vancomycin were determined by Kirby Bauer’s disc diffusion method and macro broth dilution procedure respectively, according to the 2009 CLSI guidelines. Results were interpreted as susceptible if the MIC was ≤ 2µg/ml, 4-8µg/ml as intermediate and ≥16 µg/ml as resistant.

**Results:** A total of 400 strains of *S. aureus* collected over three year period were included in the study. Of these, 104 (26%) were MRSA. All the MRSA isolates were susceptible to vancomycin by disc diffusion method as well as MIC. 30 (28.8%) were with MIC of 0.5 µg/ml, 67 (64.4%) were with 1µg/ml and 07 (6.7%) with 2µg/ml. There was an increase in the percentage of isolates from 0.5µg/ml to 1µg/ml during the study period of three years.

**Conclusion:** There is an increase in the number of strains with MICs of 1µg/ml. This may indicate a step towards development of VISA and possibly VRSA. Continuous monitoring of MIC levels of vancomycin in MRSA is warranted.

**Keywords:** Vancomycin, MRSA, MIC
INTRODUCTION

The antimicrobial chemotherapy for Staphylococcus aureus infections has been empirical as this species has overcome most of the therapeutic agents that have been developed in the recent years. S. aureus is among most common organisms in hospital and community acquired infections. In recent years, a large and continuing increase in the prevalence of methicillin resistant S. aureus (MRSA) has been observed. In addition, the emergence of S. aureus with reduced susceptibility or resistance to vancomycin also has been reported.

Vancomycin is the antibiotic of choice for the treatment of severe infections caused by MRSA. However, vancomycin treatment failure in MRSA bacteraemia is not uncommon, even when MRSA is susceptible to vancomycin. Recently, a direct impact of vancomycin Minimum inhibitory concentration (MIC) on the morbidity associated with MRSA bacteraemia was described even when the MICs were within the susceptibility range. There are reports of an increase in MICs of vancomycin over the time.

Although, there are many studies reported MIC on MRSA, only limited studies are presently available on MRSA isolates with reduced susceptibility to vancomycin in clinical laboratories. In the present study, vancomycin MICs of MRSA were determined for the period of three years from 2009-2011 and analyzed to determine the trends.

MATERIALS AND METHODS

Between July 2009 and June 2011, a total of 400 S. aureus strains were isolated on sheep blood agar from the patients’ specimens. Confirmation of the strains was done by catalase test, tube coagulase test, mannitol fermentation test and deoxyribonuclease test according to the standard techniques. The protocol of this study was approved by Yenepoya University Ethics Committee.

All the isolates were subjected to antibiotic sensitivity testing by Kirby-Bauer disc diffusion method for oxacillin using a 1μg disc, cephalothin using 30μg disc and vancomycin using 30μg disc on Mueller- Hinton Agar (MHA). The plates were incubated at 35°C for 24 hours and zones of inhibition were measured. An inhibition zone of ≥13 mm was considered as susceptible, 11-12 mm was considered as intermediate and ≤10 mm as resistant for oxacillin. An inhibition zone of ≥20 mm was considered as susceptible and ≤19 mm resistant for cephalothin. An inhibition zone of ≤15 mm in diameter was considered as vancomycin resistant.

Control strains, ATCC 29213 for MRSA and ATCC 25923 for MSSA were used in the antibiotic sensitivity testing. Minimum inhibitory concentration (MIC) for all the strains identified as MRSA by disc diffusion method was tested for vancomycin by standard macro broth dilution method as per Clinical and Laboratory Standards Institute (CLSI) 2009 guidelines. Results were interpreted as vancomycin ≤ 2µg/ml as susceptible, 4-8 µg/ml as intermediate and ≥16µg/ml as resistant.

RESULTS

Of the total 400 strains included in the study, 104 strains (26%) were MRSA. The number of strains isolated in each year for three years was 38, 33 and 33 respectively. The year wise number of strains with different vancomycin MICs is presented in Table 1.

All the MRSA isolates were confirmed as vancomycin susceptible by MIC detection. The MRSA isolates with a vancomycin MIC of 0.5µg/ml was 44(42.3%), 1µg/ml was 50(48.07%) and 2µg/ml was 10(9.6%). There is an increase in the percentage of isolates with vancomycin MIC of 1µg/ml; 44.4% in 2009, 48.48% in 2010 and 51.51% in 2011.

DISCUSSION

Studies on vancomycin MICs have shown conflicting results. Though all the strains of S. aureus were found susceptible to vancomycin as per the breakpoints, increase in the MIC from ≤0.5µg/ml to 1 µg/ml was observed for MRSA isolates.

A study by Holmes et al showed no vancomycin MIC movement in bacteremia MRSA isolates from 1999 to 2006 in Texas, USA. Vancomycin activity remained stable in terms of inhibitory and bactericidal activity. In hospital settings with a widespread use of vancomycin to treat MRSA infections, there might be a selection pressure for emergence of vancomycin intermediate S. aureus (VISA) and vancomycin resistant S. aureus (VRSA).
In the present study, although vancomycin usage in our hospital in the past three years has been minimal, we found that vancomycin MIC of MRSA increased from 0.5µg/ml to 1µg/ml. Similar results have been reported by other investigators, who observed an increase in MIC to vancomycin, inspite of all the strains being within the CLSI breakpoints. The increase in the number of such strains may not be as high as reported by some studies. This may indicate a step towards development of VISA and possibly VRSA. Continuous monitoring of MIC levels of vancomycin in MRSA is warranted.

CONCLUSION

There is an increase in the number of strains with MICs of 1µg/ml which may indicate a step towards development of VISA and possibly VRSA. This may be a matter of concern because there are barely any treatment options available. This study stresses the need for continuous monitoring of MIC levels of vancomycin in MRSA, and the infection control practices to prevent transmission.

CONFLICTS OF INTEREST

None declared

REFERENCES


For More Information Log on to www.ijhrs.com