Full Length Research Paper

# Prevalence of methicillin - resistant *Staphylococcus aureus* at the Hospital of the medical University of Bushehr in Iran

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Staphylococcus aureus is Gram positive cocci which causes different infections, some of these strains are resistant to Methicillin (MRSA) and can be found in hospitals. *S. aureus* is one cause of nosocomial infections. The aim of this study was to find MRSA and Vancomycin resistant *S. aureus* (VRSA) in Fatemeh Zahra Hospital. Clinical samples were isolated from confined patients in the hospital were cultured on blood agar and mannitol salt agar and the positive coagulase and catalase colonies which were able to ferment the mannitol and trehalose were considered as *S. aureus*. These organisms were antibiogramed by disc diffusion method and E. test. In this research 72 species of *S. aureus* were collected from various clinical samples in hospital, the rate of resistance to different antibiotics is as follow: oxacillin, 59.72%; ciprofloxacin, 22.22%; Ceftizoxime, 51.38%; co-trimoxazole, 38.88%; tetracycline, 52.77% and all the strains were sensitive to vancomycin. In this study prevalence of oxacillin or methicillin resistant was 59.72% and vancomycin resistance or intermediate *S. aureus* was not detected. Treatment based on antibiogram test results and rational antibiotics therapy can prevent increase in the number of resistant species.

Key words: Staphylococcus aureus, methicillin-resistant Staphylococcus aureus (MRSA), vancomycin-resistant Staphylococcus aureus (VRSA), methicillin, vancomycin.

## INTRODUCTION

Ogston Scottish surgeon, in 1882 first observed grape – like cluster cocci and called them Staphylococcus (Ogston, 1882) and in the 1884 Anton J. Rosenbach was able to culture in the solid medium made the white and golden colonies and the name aureus was selected (Liu et al., 2005). Scale skin syndrome, toxic shock syndrome, impetigo (Ladhani et al., 1999), food poisoning (Hennekinne et al., 2011), skin infection, folliculitis, bacteremia, pneumonia, osteomyelitis, mediastinitis, endocarditis, urinary tract infection, enterocolitis (Lina et al., 1999) and infection of surgical site (Khorvash et al., 2008) are diseases which may caused by *Staphylococcus aureus*. Although this organism is one of the nosocomial infections but sometimes it is acquired from community.

In the 1980s, methicillin-resistant *S. aureus* (MRSA) were emerged and were spread in many hospitals of the world (Centers for Disease Control and Prevention, 2002). Vancomycin is the drug of choice for treatment of species of *S. aureus* which are resistant to Methicillin. There are new reports about the resistance of this species even to Vancomycin (VRSA) and Vancomycin Intermediate *S. aureus* (VISA). In 1996, for the first time Vancomycin Intermediate Resistant *S. aureus* (VISA) was reported in Japan (Centers for Disease Control and

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Sample	Number of sample	Frequency in percentage
Blood	20	26.66
Urine	16	21.33
Wound	14	18.66
Abscess	5	6.66
Catheter	1	1.33
Cerebral spinal fluid	1	1.33
Eye	1	1.33
Hospital wards	3	4
Sputum	12	16

Table 1. Frequency percentage of clinical isolates.

Prevention, 1997), and later in Europe, Asia and America (Appelbaum, 2006). In 2002, the first fully resistant species of *S. aureus* was reported in Michigan in United States (The Centers for Disease Control, 2002). At present numbers of nosocomial infections caused by *S. aureus* has been increasing. It can be due to improper methods of diagnosis and treatment of the infected patients and improper methods of infection's control in the hospitals as well. The aim of this study was to investigate the occurrence of MRSA and Vancomycin-Resistant *S. aureus* among confined patients of Fatemeh Zahra Hospital Bushehr, Iran.

#### MATERIALS AND METHODS

In this research, the total of 72 *S. aureus* collected between 2007 to 2009 were studied. They were isolated from clinical samples and were antibiogramed. All the *S. aureus* were segregated from the clinical samples of hospitalized patients of different wards.

Clinical samples were cultured on blood agar and mannitol salt agar and doubtful colonies were stained with gram stain. The Gram positive cocci in cluster were further analyzed. Catalase and coagulase positive cocci were resistant to 0.04  $\mu$ g bacitracin and sensitive to five microgram novobiocin and (ONPG)  $\beta$ -Galactosidase and D-xylose negative was considered as *S. aureus*.

Isolated species were antibiogramed using six different antibiotics as follow 1  $\mu$ g oxacillin, 30  $\mu$ g vancomycin, 5  $\mu$ g ciprofloxacin, 23.75  $\mu$ g co-trimoxazole, 30  $\mu$ g tetracycline and 30  $\mu$ g ceftizoxime and by the use of Kirby- Bauer disc diffusion method (0.5 Mcfarland) and mueller-hinton medium. For vancomycin both method disc diffusion and minimum inhibitory concentration (MIC) (E- test) were applied.

Discs were controlled with standard species of *S. aureus* and after confirmation antibiogram test was conducted. Sensitivity and resistance of isolates were determined based on the NCCLS standard, for vancomycin only zone with 15 mm and above was considered as sensitive and for intermediate and resistant species MIC were measured. According to NCCLS standard MIC range for VSSA was 0.5 to 2 µg/ml and for VRSA strains  $\geq$ 16 µg/ml were considered. While as methicillin in the test culture media is unstable, oxacillin can be used and the results can be considered for methicillin.

## RESULTS

In this study antibiogramed strains were isolated from

clinical samples collected from patients in the hospital from their sputum, urine, blood, abscesses, spinal fluid, sore eyes and catheter. The rate of resistant to Oxacillin or methicillin were 59.72%, highest rate of isolated *S. aureus* was from blood (26.66%) (Table 1). Among the 43 MRSA strains in six cases (blood = one, wounds= two, urine = one and sputum = two) were resistant to all antibiotics except vancomycin.

Resistance to antibiotics such as ciprofloxacin, ceftizoxim, co-trimoxazol and tetracycline were 22.22, 51.38, 38.88 and 52.77%, respectively. It is noted that the resistance to the tetracycline and ceftizoxime was the same. The results of sensitivity test are tabulated in Table 2. Comparison of antimicrobial susceptibility among Oxacillin-resistant *S. aureus* (MRSA) and Oxacillinsensitive *S. aureus* (MSSA) are expressed in Table 3.

Sensitivity to vancomycin by disc diffusion method was 55.5% and MIC by using E- test was 44.4%, also resistance or intermediate isolates to vancomycin was not found.

## DISCUSSION

This microorganism causes wide spectrum of diseases mostly in hospitalized patients or those with chronic underlying disease. Species of MRSA is found in much hospitals in the world. In this study the rate of resistance to Oxacillin (MRSA) was 59.7%. About 33 to 55% of United States hospitals are infected with methicillinresistant strains (Appelbaum, 2006). In another study in eight university-affiliated hospitals in Korea the methicillin resistance rate was reported 64% (Kim et al., 2004). In a study in the Netherlands in 2007, thirty percent of staphylococcus isolated from humans was resistant to methicillin (Huijsdens et al., 2009). In a study done in the Imam-Khomeini Hospital in Tehran the rate of prevalence of S. aureus resistant to methicillin was estimated 41.85% (Aligholi et al., 2007). In a review which based on the global reports the rate of MRSA have been expressed between 10 to 77% (Orrett and Land, 2006). Based on these studies our finding is not unexpected.

Vancomycin is the drug of choice for treatment of

Antibiotic	Sensitive (%)	Intermediate (%)	Resistant (%)
Oxacillin	29.16	11.11	59.72
Vancomycin	100	0	0
Ciprofloxacin	58.33	19.44	22.22
Ceftizoxime	38.88	9.72	51.38
Co-trimoxazole	58.33	2.77	38.88
Tetracycline	34.72	12.5	52.77

**Table 2.** Frequency percentage of sensitivity of isolated S. aureus to different antibiotics.

 Table 3. Frequency percentage of resistance MRSA, MISA and MSSA to different antibiotics.

	Oxacillin			
Antibiotic	Resistant (%)	Intermediate (%)	Sensitive (%)	
	(n = 43)	(n = 8)	(n = 21)	
Vancomycin	0	0	100	
Ciprofloxacin	25.58	37.5	9.52	
Ceftizoxime	55.81	37.5	47.6	
Co-trimoxazole	48.83	25	23.8	
Tetracycline	53.48	62.5	47.6	

MRSA, first report on VRSA in Iran was in 2005 when five cases were identified in Tehran (Saderi et al., 2005). In the present study all the strains were sensitive to vancomycin by disc diffusion method and MIC (E- test). In disc diffusion method only 55.5% of strains were sensitive to vancomycin and sensitivity of the remaining isolates, were determined by MIC. According to the results use for disc diffusion method alone is not suitable for determination of sensitivity to vancomycin.

Ciprofloxacin also can be used for treatment of strains resistant to methicillin. In this study, 25.58% of MRSA strains were resistant. The greatest rate of resistance of MRSA isolates were seen for Ceftizoxime 55.81% followed by Tetracycline 53.48% and Co-trimoxazole 48.83%. MRSA strains compared with MSSA have a higher resistance (Table 3).

Carefully antibiotic therapy by physicians and appropriate medication and avoiding their indiscriminate use can prevent development of resistant strains in the future. Also critical care to discover for MRSA strains and ongoing studies periodically by the hospital infection control groups or research centers, can prevent the epidemic spread of MRSA in the hospitals.

## Conclusion

The purpose of the current study was to determine occurrence of MRSA and VRSA in the Fatemeh Zahra Hospital in Bushehr. This study showed that 59.7% of isolates were Methicillin-Resistant *S. aureus* (MRSA) and all the *S. aureus* were sensitive to Vancomycin.

Therefore based on the results of the study rational use of antibiotics is recommended.

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