Original Article

DOI: NJLM/2013/6900:1996

Microbiology Section

Evaluation of Methicillin Resistant *Staphylococcus Aureus* Colonization in Patients and Nursing Staff of Intensive Care Units of A Tertiary Care Hospital in A Rural Area

RAJESH BAREJA, KANU GOEL, SHINU POTTATHIL, VINOD KUMAR NARANG, VARSHA ASHOK SINGH, PREM SINGH GROVER

ABSTRACT

Context: Methicillin resistant *Staphylococcus aureus* (MRSA) is an increasing problem in health care facilities. Rising colonization rates lead to increased infection rates in the community and in hospitals. MRSA infection in critically ill patients is associated with prolonged intensive care unit stay, increased medical cost and high mortality.

Aim: The prevalence and rate of acquisition of methicillin-resistant *S. aureus* in nursing staff and patients admitted to the intensive care units (ICUs) were studied in order to estimate the possible risk for those, who are initially free of the organism, of acquiring MRSA infection while maintained in the ICU.

Settings and Design: This was a cross sectional study conducted in the Department of Microbiology, M.M.I.M.S.R. Mullana, Ambala, Haryana, India.

Material and Methods: A total of 400 specimens of hands, throat and nares from the fifty patients and fifty nursing staff of ICUs were subjected to bacteriological examination. All the specimens were processed on blood agar and MacConkey agar then incubated at 37°C in incubator for 24 hours. Suspicious colonies were identified as *Staphylococcus aureus* by various biochemical tests. Methicillin resistant was confirmed with cefoxitin disk (30µg) susceptibility test and Oxacillin Salt Agar method.

Results: Of these, 234 (58.5%) isolates were found to be positive for MRSA. MRSA strains isolated from hands, throat and nares of patients and nursing staff were 74 (37%) and 78 (39%), 39 (39%) and 41 (41%), 2 (2%) and 0% respectively. Surgery ICU had the highest burden (30.25%) followed by Medicine ICU (17.75%) and Paediatric ICU (10.5%). Antibiotic sensitivity testing identified 31.63% MRSA strains to be resistant to vancomycin.

Conclusion: The results indicate, screening of ICU staff and patients for methicillin resistant *S. aureus* colonization and infection, accompanied by antibiotic sensitivity testing of cultured isolates, is important to understand its epidemiology and to develop preventive measures and treatment strategies.

Keywords: Cefoxitin disk, ICU, MRSA, Oxacillin salt agar

INTRODUCTION

Infections with methicillin resistant *Staphylococcus aureus* (MRSA) remain a major concern. MRSA was first described in the 1960s [1]. It is presently endemic in many hospitals [2]. The burden of MRSA continues to rise, with a rate of 14% of all *Staphylococcus aureus* strains. The differentiation between Community-Acquired MRSA (CA-MRSA) and hospital-acquired MRSA (HA-MRSA) has been difficult, since CA-MRSA can also spread into hospitals [3]. Intensive Care Units (ICUs) are high-risk areas for the selection and transmission of multi-drug-resistant bacteria and surveillance data show that MRSA is endemic in ICUs in all German regions [4-6]. The possible risk of acquiring MRSA infection in ICUs is dependent upon the severity of illness, length of stay, use of intravascular devices and the intensity of exposure to infected patients [7-10]. Overall, intrinsic together with extrinsic risk factors make the ICU patient extremely vulnerable to HA-MRSA infections. Since direct contact with medical staff members who are colonized with MRSA is a main route of transmission, the surveillance of the medical staff is also important. However,

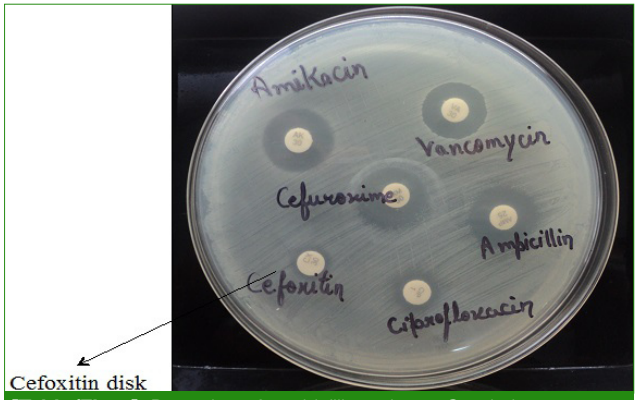
to our knowledge, few surveillance studies on medical staff personnel have been performed. Therefore, this study was conducted to evaluate the detection of MRSA in patients admitted to and medical staff posted in intensive care units.

MATERIAL AND METHODS

Test specimen and identification: The cross sectional study was carried out in a tertiary care hospital over a period of eleven months. During the study 400 specimens were subjected to bacteriological examination from the fifty patients and fifty nursing staff of ICUs. For newly admitted patients in ICU, the screening for MRSA was carried out within the first 48 hours. Four swabs from hands, nasal and throat of the patients and nursing staff were collected with moist sterile cotton-tipped sticks and transported immediately to the Microbiology department of M.M.I.M.S.R, Mullana, Haryana, India for processing. All the specimens thus obtained were processed on blood agar and MacConkey agar. All the inoculated plates were incubated at 37°C in incubator for 24 hours. Suspicious colonies were identified as *S. aureus* by colony morphology, Gram staining, catalase production, clumping test, tube coagulase test, deoxyribonuclease test and mannitol fermentation test [11]. Ethical clearance for the study was obtained from the institutional ethical committee.

Screening for methicillin resistance: Methicillin resistant was confirmed with cefoxitin disk (30µg) susceptibility test [Table/Fig-1] and Oxacillin Salt Agar method (Mueller-Hinton agar containing 4% NaCl and 6.0 mg/litre oxacillin) [12,13].

Antibiotic susceptibility tests: Sensitivity to relevant antibiotics was determined by the Kirby-Bauer disk diffusion method as per the Clinical and Laboratory Standards Institute (CLSI) guidelines, using the commercially available antibiotic disks from Hi-Media (Mumbai, India) [14]. NCCLS (National Committee for Clinical Laboratory Studies) reference strains, *Staphylococcus aureus* ATCC (American Type Culture Collection, Manassas, VA, USA) 25923 and MRSA strain ATCC 43300 were included as control strains.



[Table/Fig-1]: Detection of methicillin resistant *Staphylococcus aureus* (MRSA) by using cefoxitin disk

RESULTS

Fifty patients and fifty nursing staff of ICUs underwent screening with specimens assessed for left hand (n=50), right hand (n=50), throat (n=50) and nasal (n=50) MRSA colonization. Of these, 234 (58.5%) isolates were found to be positive for MRSA. Out of two hundred thirty four isolates, 115 (49.14%) and 119 (50.85%) were from patients and nursing staff respectively. MRSA strains isolated from hands, throat and nares of patients and nursing staff were 74 (37%) and 78 (39%), 39 (39%) and 41 (41%), 2 (2%) and 0% respectively [Table/Fig-2].

Out of 50 patients, 35 (70%) were positive for MRSA. Of 35 patients more than half were community acquired 20 (57.14%), whereas 15 (42.85%) were hospital acquired MRSA. Sex wise distribution of MRSA strains in patients and nursing staffs was also observed. In males and females it was 30 (50%) and 10 (25%) respectively. MRSA strains were found more in males than females (2:1). Surgery ICU had the highest burden (30.25%) followed by Medicine ICU (17.75%) and Paediatric ICU (10.5%) [Table/Fig-3].

Antibiotic sensitivity: Antibiotic sensitivity testing identified that the vancomycin had the best spectrum of antimicrobial activity in patients and nursing staff as well. However, vancomycin exhibited the 31.63% resistant against methicillin resistant strains. The sensitivity pattern to relevant drugs tested against the MRSA isolates is depicted in [Table/Fig-4].

Individuals (n=100)	Hand swabs (n=200)	Throat swabs (n=100)	Nasal swabs (n=100)
Patients	74 (37%)	39 (39%)	2 (2%)
Nursing staffs	78 (39%)	41(41%)	0 (%)

[Table/Fig-2]: Frequency of MRSA strains in patients and nursing staff

ICU Wards	Left hand swabs (n=100)	Right hand swabs (n=100)	Throat swabs (n=100)	Nasal swabs (n=100)
Medicine ICU	23 (5.75%)	23 (5.75%)	25 (6.25%)	0 (0%)
Surgery ICU	40 (10.0%)	40 (10.0%)	41 (10.25%)	0 (0%)
Pediatric ICU	13 (3.25%)	13 (3.25%)	14 (3.5%)	2 (0.5%)
Total (234)	76	76	80	2

[Table/Fig-3]: Ward wise distribution of MRSA strains (n=400)

Antibiotic disks	Patients	Nursing staff	Total (%)
Ciprofloxacin	56	57	113 (48.29%)
Ampicillin	66	92	158 (67.52%)
Vancomycin	75	85	160 (68.37%)
Amikacin	84	66	150 (64.10%)
Cefuroxime	45	40	85 (36.32%)

[Table/Fig-4]: Antibiotic susceptibility pattern of MRSA strains in patients and nursing staff samples (n=234)

DISCUSSION

Emergence of MRSA has not only caused therapeutic problems in hospitals but also created tremendous pressure on resources for controlling their spread. The resistance of MRSA to a wide range of antibacterial is well documented. Multiple prolonged uses of antibiotics and hospitalization are other important factors that make hospitals an ideal place for transmission and perpetuation of MRSA. Methicillin resistant *S. aureus* has become an enormous problem for health care providers because it is a major nosocomial isolate in hospitals and difficult to treat, called super bugs.

The MRSA prevalence rates were differ in various countries across the world, which is in between 26%-66.8% and in India it is 12%-77.9% [15-17]. Rising colonization rates lead to increased infection rates in the community and in hospitals. The consequence to the health care system is longer hospital stays and greater costs, which approximately double the expenditure per patient [18]. In the present study the MRSA positivity is 58.5%. The prevalence of MRSA in hospitals varies considerably from region to region. The important reservoirs of MRSA in hospitals are infected patients and transient nasal carriers among hospital staff. There is hardly any study involving the nursing staff. The present study includes both nurses and patient that may be the reason for higher positivity.

Majumdar et al., (2009) reported that the prevalence of MRSA in the community of East Sikkim was 11.1% while other authors reported CA-MRSA prevalence 30.2% and 55% [19-21]. In the present study community acquired methicillin resistant *S. aureus* positivity is 40%. This study was conducted in rural area where joint families are more and they live in close proximity that may be the reason for transmission of infection by close contact. The results of the present study showed that the prevalence of MRSA is more in males (50%) than the females (25%). Mahmood et al., (2010) and Zer et al., (2009) had supported the present study by showing the male predominance in their study [22,23] while Sharma et al., (2011) in contrast to present study reported a higher prevalence in females (68.9%) than the males (39.13%) [17].

In the present study, strains of MRSA isolated from majority of patients who were in the age group of 0-15 years (50%) followed by the 16-45 years (39%) and of >45 years (25%). This result indicates that the extreme of the ages were more prone to get MRSA infection due to weak immune system. Sharma et al., (2011) and Mohd et al., (2011) also noted that extreme of the ages were prone to get MRSA infection [17,24].

The resistance of MRSA to a wide range of antibacterial is well documented. In the present study MRSA strains exhibited the vancomycin resistance (31.63%), which is an alarming signal. While other workers showed the 100% sensitivity of MRSA strains against vancomycin [25,26]. The reduced susceptibility in some strains of MRSA such a strategy will require careful risk-assessment.

According to National Nosocomial Infection Surveillance System (NNIS) report, 50% of hospital-acquired infections in ICUs in the USA are due to MRSA [27]. The frequency of MRSA in hospitals varies considerably from ward to ward. In the present study maximum isolates of MRSA are from Surgery ICU (30.25%), followed by Medicine ICU (17.75%) and Paediatric ICU (10.5%). Mahmood et al., (2010) in contrast to our study also reported that incidence of MRSA was higher in Medicine ICU (54.34%) followed by Medical and allied (21.89%) and then Surgery ICU (9.43%) [22]. The highest incidence in Surgery may be because of surgical intervention and exposed to antimicrobials as prophylaxis leads to compromised immune system as compare to Medicine ICU. This finding confirms the effect of close contact and the transmission of bacteria from personnel to patients. After introduction within hospital MRSA spreads rapidly by hands of medical personnel. Colonized employees of the hospital such as asymptomatic nasal, throats carriers and infected patients act as reservoirs, are important sources to spread this organism. Uemura et al., (2004) in Japan revealed that the throat as well as the nasal vestibulum was the habitat of *S. aureus* and the real incidence rate of *S. aureus* from the throat (15%) might be higher than that of nose (4%) because sampling from the throat is more difficult than from the nose [15]. This may indicate cross contamination of MRSA between personnel and patients. This situation may be related with both excessive workload and poor sanitation measures within this group. Another risk factor related to colonization has been duration of employment of healthcare workers in ICUs.

LIMITATIONS

First, this study was carried out in a single institution. Because there is epidemiologic variation in healthcare-associated infection among institutions, results potentially may not be universally applicable. Second, external validation was not performed in this study. In the future, it will be necessary to perform an external validation analysis or to examine whether preemptive infection control for patients with high-risk factors actually works effectively.

CONCLUSION

A change in practice necessitates an emphasis on prevention. It is an alarming signal for transmission of methicillin resistant *Staphylococcus aureus* from personnel to patients. Staff members will pick up and propagate MRSA from one patient to another in the absence of effective hand hygiene. It is also possible that the environment serves as an inanimate reservoir because MRSA can survive desiccation and has been

demonstrated on a variety of surfaces and medical equipment in hospitals. Therefore, more emphasis should be given on healthcare worker education, enhanced surveillance, patient cohorting and enhanced environmental cleaning. Throat swab culture showed the highest positivity to MRSA in nursing staff 82% (41/50) as well as in patients 78% (39/50). Wearing mask and proper hand washing should be compulsory for nursing staff at least in intensive care units to prevent the transmission of infection to patients. The risk may be enhanced by hygiene failures in the ICU environment, which would allow persistence of resilient pathogens. An improvement in the time to diagnose patients along with isolation room resources would enhance efforts to minimize ICU transmission of MRSA.

REFERENCES

[1] Jevons MP. Methicillin-resistant *staphylococcus*. *Bio Med Jour.* 1961; 1: 124-25.

[2] Vincent JL, Bihari DJ, Suter PM, Bruining HA, White J, Nicolas-Chanoin MH, et al. The prevalence of nosocomial infection in intensive care units in Europe: results of the European Prevalence of Infection in Intensive Care (EPIC) study. *J Amer Med Assoc.* 1995 Aug 23-30; 274(8): 639-44.

[3] Calfee DP, Durbin LJ, Germanson DM Toney, Smith EB, Farr BM. Spread of methicillin-resistant *Staphylococcus aureus* (MRSA) among household contacts of individuals with nosocomially acquired MRSA. *Infect Control Hosp Epidemiol.* 2003; 24: 422-26.

[4] Fridkin SK. Increasing prevalence of antimicrobial resistance in intensive care units. *Crit Care Med.* 2001; 29 (Suppl 4): N64-N68.

[5] Archibald L, Phillips L, Monnet D, McGowan JE Jr, Tenover F, Gaynes R. Antimicrobial resistance in isolates from inpatients and outpatients in the United States: increasing importance of the intensive care unit. *Clin Infect Dis.* 1997; 24: 211-15.

[6] Kohlenberg A, Schwab F, Meyer E, Behnke M, Geffers C, Gastmeier P. Regional trends in multidrug-resistant infections in German intensive care units: a real-time model for epidemiological monitoring and analysis. *J Hosp Infect.* 2009; 73: 239-45.

[7] Ibelings MMS, Bruining HA. Methicillin-resistant *Staphylococcus aureus*: acquisition and risk of death in patients in the intensive care unit. *Eur J Surg.* 1998; 164: 411-18.

[8] Law MR, Gill ON. Hospital acquired infection with methicillin-resistant and methicillin sensitive Staphylococci. *Epidemiol Infect.* 1988; 101: 623-29.

[9] Pujol M, Pena C, Pallares R. Risk factors for nosocomial bacteremia due to methicillin-resistant *Staphylococcus aureus*. *Eur J Clin Microbiol Infect Dis.* 1994; 13: 96-102.

[10] Merrer J, Santoli F, Vecchi C. Colonization pressure and risk of acquisition of methicillin resistant *Staphylococcus aureus* in a medical intensive care unit. *Infect Control Hosp Epidemiol.* 2000; 21: 718-23.

[11] Collee JG, Fraser AG, Marmion BP, Simmons A. 2006. Tests for the identification of Bacteria. Mackey and McCartney practical Medical Microbiology, New Delhi, India: Elsevier, p.131-149.

[12] Murray PR, Baron EJ, Jorgenson JH et al. 2003. Special phenotypic methods for detecting antibacterial resistance. Manual of Clinical Microbiology, Washington DC: ASM Press, p.1179.

[13] Thornsberry C, McDougal LK. Successful use of broth microdilution in susceptibility tests for methicillin-resistant (heteroresistant) Staphylococci. *J Clin Microbiol.* 1983; 18: 1084-091.

[14] The Clinical and Laboratory Standards Institute (CLSI) (2008) Performance standards for antimicrobial susceptibility testing: eighteenth informational Supplement, M100-S18 CLSI. Wayne, Pa.

[15] Uemura E, Kakinohana S, Higa N, Toma C, Nakasone N. Comparative Characterization of S.aureus Isolates from Throats & Nose of healthy Volunteers. *J Infect dis.* 2004; 57: 21-24.

[16] Verma S, Joshi S, Chitinis V, Hemwani N, Chitinis D. Growing problem of MRSA- Indian Scenario. *Indian J Med Sci.* 2000; 54(12): 535-40.

[17] Sharma S, Mall A. The prevalence, antibiogram and characterisationof methicillin resistant *Staphylococcus aureus* among the patients from the Doon Valley hospitals. *African J Microbiol Res.* 2011; 5(21): 3446-451.

[18] Kim T, Oh PI, Simor AE. The economic impact of methicillin resistant *Staphylococcus aureus* in Canadian hospitals. *Infect Control Hosp Epidemiol.* 2001; 22(2): 99–104.

[19] Majumdar D, Barua A, Paul B. Nasal Carriage of Methicillin Resistant Staphylococci in Healthy Population of East Sikkim. *Indian J Community Med.* 2009; 34(4): 364-65.

[20] Joel WB, Bernadette B. Community Acquired Methicillin Resistant *Staphylococcus aureus*: Prevalence and Risk Factors. *Athl Train.* 2006; 41(3): 337–40.

[21] Thea C, Carol L, Moore, Perri MB, Donabedian SM, Masch W, et al. Molecular epidemiology of MRSA Blood Stream in Urban Detroit. *J Clin Microbiol.* July 2008; 46(7): 2345-52.

[22] Mahmood K, Tahir T, Jameel T, Zlauddin A, Aslam HF. Incidence of MRSA causing Nosocomial Infection in a tertiary care hospital. *Annals.* 2010; 16(2): 91-96.

[23] Zer1 Y, Karaoglan I, Mustafa N, Balcı1 I, Karagoz ID, Ozaslan M, et al. Investigation of nasal colonization of health care workers by Methicillin Resistant *Staphylococcus aureus* with using new generation real-time PCR assay: Discussing of risks. *African J Biotech.* 2009; 8 (20): 5542-546.

[24] Mohd A, Khan MA, Sharma S. Current trend of antimicrobial susceptibility pattern of methicillin resistant *S. aureus* isolated from tertiary care hospital Delhi NCR region India. *IJRAP.* 2011; 2(4): 1339- 342.

[25] Hafiz S, Hafiz AN, Ali L, Chughtai AS, Memon B, Ahmed A, et al. Methicillin resistant *Staphylococcus aureus*: a multicenter study. *J Pak Med Assoc.* 2002; 52(7): 312-15.

[26] Tyagi A, Kapil A Singh P. Incidence of MRSA in Pus Samples at a Tertiary Care Hospital, AIIMS, New Delhi. *JIACM.* 2008; 9(1): 33-35.

[27] Klevens RM, Morrison MA, Nadle J. Invasive Methicillin Resistant *Staphylococcus aureus* infections in the United States. *J Amer Med Assoc.* 2007; 298: 1763–771.

AUTHOR(S):

1. Dr. Rajesh Bareja
2. Dr. Kanu Goel
3. Dr. Shinu Pottathil
4. Dr. Vinod Kumar Narang
5. Dr. Varsha Ashok Singh
6. Dr. Prem Singh Grover

PARTICULARS OF CONTRIBUTORS:

1. Assistant Professor, Department of Microbiology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.
2. Assistant Professor, Department of Pathology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.
3. Assistant Professor, Department of Microbiology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.
4. Professor, Department of Microbiology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.
5. Professor, Department of Microbiology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.
6. Professor, Department of Microbiology, M.M.I.M.S.R, Mullana, Ambala, Haryana – 133207, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:

Dr. Rajesh Bareja,
Assistant Professor, Department of Microbiology, Maharishi Markandeshwar Institute of Medical Sciences and Research, Mullana, Ambala, Haryana – 133207, India.
Email: rajeshbareja@gmail.com
Ph: +91-9034882188

FINANCIAL OR OTHER COMPETING INTERESTS: None.

Date of Publishing: **Dec 31, 2013**