

SCREENING FOR METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS AND DETECTION OF *mecA* GENE BY PCR AMONG THE HEALTH CARE WORKERS IN A TERTIARY CARE HOSPITAL

Dheepa N¹, Rajesh K R.², Seetha K S.³

ABSTRACT

Background : Methicillin Resistant *Staphylococcus aureus* (MRSA) is recognized worldwide as a major nosocomial pathogen in hospitals. MRSA strains may spread readily in hospitals from colonised or infected persons. Screening for MRSA carriers among this population is necessary for nosocomial infection control.

AIMS & OBJECTIVES : The aim of the study was to determine the prevalence of MRSA and detection of *mecA* gene among the Health Care Workers (HCWs). The objective of the study was to screen HCWs for *Staphylococcus aureus* carriage and detection of *mecA* gene.

MATERIALS & METHODS : A total of 100 HCWs were screened for *Staphylococcus aureus* carriage. Using pre-moistened sterile cotton swabs, specimens collected from the anterior nares, palms and web spaces were cultured & identified by conventional methods for *Staphylococcus aureus*. The antimicrobial susceptibility test was performed. *Staphylococcus aureus* isolates were screened for methicillin resistance by Oxacillin (1 µg) disc diffusion, Oxacillin screen agar (6 µg/ml) and Cefoxitin (30 µg) disc diffusion & confirmed by detection of *mecA* gene by PCR.

RESULTS : Out of the 100 HCWs, 23 % were found to be *Staphylococcus aureus* carriers. The overall MRSA carrier rate was 10 %. Out of 10 MRSA carriers, 7 were from nasal and 3 were from web spaces . All 10 MRSA were detected by Oxacillin screen agar and Cefoxitin disc diffusion as against 8 MRSA by Oxacillin disc diffusion method. Male HCWs harboured statistically significant MRSA. All 10 MRSA isolates were positive for *mecA* gene by PCR.

CONCLUSION : Our study concludes that MRSA was prevalent in 10 % of HCW's and anterior nares was the commonest site. Male HCWs were predominant carriers of MRSA.. Cefoxitin disc diffusion method can be used as an alternative to the technically demanding PCR.

Keywords : Methicillin Resistant *Staphylococcus aureus* (MRSA), Health Care Workers (HCW's).

INTRODUCTION

Methicillin is an antibiotic derived from Penicillin, which has been used as a drug clinically since 1960¹. Indiscriminate use of methicillin has led to resistance in *Staphylococcus aureus*^{2,3}. Methicillin Resistant *Staphylococcus aureus* (MRSA) is recognized worldwide as a major nosocomial pathogen⁴. MRSA is due to expression of the *mecA* gene. Presence of abnormal PBP-2a which has low affinity to beta-lactam antibiotics, is responsible for the resultant resistance to Staphylococci^{1,5}. MRSA strains may spread readily in hospitals from colonised or infected persons. Colonised employees are generally asymptomatic, although they are a potential reservoir of infections acquired by patients.^{6,7} Although, MRSA strains have not been shown to be more virulent than *Staphylococcus aureus*,^{6,8} very high mortality rates have been reported from several centres.^{6,9} Screening for MRSA carriers among this population is necessary for nosocomial infection control⁷. The most effective preventive tool is rapid confirmation of MRSA existence, followed by efficient execution by infection control measures. The aim of the study was to determine the prevalence of MRSA and detection of *mecA* gene among the Health Care Workers (HCWs). The objective of the study was to screen HCWs for MRSA carriage by Oxacillin disc diffusion (1 µg), Oxacillin screen agar (6 µg / ml), Cefoxitin (30 µg) disc diffusion method and *mecA* gene detection by PCR technique.

MATERIALS & METHODS

A total of 100 HCWs were screened for *Staphylococcus aureus* carriage. Using premoistened sterile cotton swabs, specimens were collected from the anterior

¹Post Graduate student, ²Professor, ³Professor & Head, Dept. of Microbiology
Vinayaka Mission's Kirupananda Variyar Medical College, Salem.

Cefoxitin disc diffusion sensitivity reported by K.B Anand et al²⁰ and by Annie Felten et al²¹ shows 96.8 %, 100 % and 96.4 %, 100 % respectively. In our study, both Oxacillin screen agar and Cefoxitin disc diffusion methods showed 100 % sensitivity (Table.1). However, Oxacillin screen agar generally does not detect borderline resistant strains, especially when these strains have heterogeneous resistance nature.²² The conventional culture technique for MRSA diagnosis is common and inexpensive. However, such techniques are time consuming, taking 48 - 72 hrs and standardization is difficult¹. Since 1980, molecular techniques have been performed in MRSA diagnosis, these techniques display the highest level of sensitivity and allow simultaneous detection of *Staphylococcus aureus* and the *mecA* gene²³. Also, because molecular techniques can utilize collected samples directly, the time required is also much shorter than for standard diagnostic techniques²⁴. Similar to previous studies, our study also detected *mecA* gene from all MRSA isolates. Results of Cefoxitin disc diffusion test is in concordance with the PCR for *mecA* gene and thus the Cefoxitin disc diffusion method is very suitable for detection of MRSA and the test can be an alternative to PCR for detection of MRSA in resource constraint settings. Prevention of MRSA infection merits discussion as once introduced in a hospital, MRSA are very difficult to eradicate. Whether the eradication of MRSA carrier state will lead to a decreased rate of MRSA infection is yet to be documented. Local therapy with Mupirocin ointment has been shown to eliminate MRSA nasal colonisation in both patients and hospital personnel, but recolonisation often occurs after therapy is discontinued. It is possible that long term intermittent therapy with Mupirocin may be more effective in suppressing or eradicating MRSA colonisation.⁸

CONCLUSION

Our study concludes that MRSA was prevalent in 10 % of HCWs and anterior nares is the commonest site for MRSA carriage. Male HCWs (13.78%) were predominant carriers of MRSA. Cefoxitin disc diffusion shows 100 % sensitivity in comparison to *mecA* gene detection by PCR. Hence, Cefoxitin disc diffusion method can be used as an

alternative to the technically demanding PCR. Since nasal carriage of MRSA in HCWs is asymptomatic, screening is extremely advantageous to prevent transmission and occurrence of nosocomial MRSA infections.

REFERENCES

1. Yasemin Zer , Iikay karaoglan , Mustafa Namiduru , Iclal Balci , Isik Didem Karagoz , Mehmet Ozaslan , Halil Ibrahim , and Aynur Suner. Investigation of nasal colonization of health care workers by methicillin – resistant *Staphylococcus aureus* with using new generation real-time PCR assay : Discussion of risks African journal of Biotechnology.2009; vol.8 (20): pp 5542-5546, 19 October.
2. S Rallapalli , S Verghese , R S Verma . Validation of multiplex PCR strategy for simultaneous detection and identification of workers by methicillin – resistant *Staphylococcus aureus* Indian Journal of Medical Microbiology. 2008;vol .26 (4): pp 361-364.
3. S Vidhani , P L Mehndiratta , M D Mathur Study of methicillin – resistant *Staphylococcus aureus* (MRSA) isolates from high risk patients. Indian Journal of Medical Microbiology 2001;vol.19 (2): pp 13-16.
4. Carol A Kurbis , MD CCFP FRCPC and John L Wylie, Ph D. Community – based cluster of methicillin – resistant *Staphylococcus aureus* in Manitoba Can J Infect Dis. 2001; vol. 12 (30): pp 149-152.
5. Mathews AA , Thomas M Appalaraju B , Jayalakshmi J. Evaluation and comparison of tests to detect methicillin – resistant *Staphylococcus aureus*. Indian J Pathol Microbiol 2010;vol.53 (1) : pp 79–82.
6. R Goyal , S Das , M Mathur Colonization of methicillin – resistant *Staphylococcus aureus* among health care workers in a tertiary care hospital of Delhi. Indan J . Med. Scie 2002; vol .56 : pp 321-324.
7. S Mathanraj , S Sujatha , K Sivasnageetha , S C Parija . Screening for Methicillin – Resistance in *Staphylococcus aureus* carriers among patients and health care workers of a tertiary care hospital in south india. Indian Journal of Medical Microbiology 2009; vol .27 (1): pp 62-64.
8. Peacock Je Jr, Moorman DR, Wenzek RP *et al*. Methicillin Resistant *Staphylococcus aureus* : microbiologic characteristics , antimicrobial susceptibilities and assessment of virulence of an epidemic strain. J. Infect dis 1981; 144: pp 575-582.
9. Locksley RM, Cohen ML, Quinn TC, et al Multiple Antibiotic resistant *Staphylococcus aureus*: introduction, transmission and evolution of nosocomial infection. Ann Int Med. 1982; vol.97: pp 317-324

10. JB Sarma, GU Ahmed. Characterisation of methicillin – resistant *Staphylococcus aureus* strains and risk factors for acquisition in a teaching hospital in northeast india. . Indian Journal of Medical Microbiology 2010; vol.28(2): pp 127-129.
11. M S Shenoy , G K Bhat , A Kishore , M K Hassan. Significance of MRSA strains in community associated skin and soft tissue infections. Indian Journal of Medical Microbiology 2010; vol.28(2): pp 152-154.
12. Clinical and laboratory standards institute. Performance standards for antimicrobial disk diffusion tests. Approved standards. 9th ed. CLSI document M2-M9. Wayne Pa: CLSI; 2006.
13. Broen DF, Edwards D I, Hawkey PM , Morrison D, Ridgway GL, Towner K J , et al . Guidelines for the laboratory diagnosis and susceptibility testing of methicillin – resistant *Staphylococcus aureus* (MRSA). Antimicrobial Agents And Chemotherapy 2005; vol.56: pp 1000-1018.
14. Clinical and laboratory standards institute (CLSI) M100-S22 January 2012, vol.32, No. 3, Page No. 81
15. D.Jegadeeshkumar^{1*}, V. Saritha¹, K. Moorthy², B.T. Suresh kumar Prevalence, antibiotic resistance and RAPD analysis of food isolates of *Salmonella* species International Journal of Biological Technology 2010; vol. 1(3): pp50-55
16. N. S. Abu Hujie and F.A.Sharif. Detection of methicillin – resistant *Staphylococcus aureus* in nosocomial infections in Gaza strip . African journal of microbiology research 2008; vol 2: pp 235-241
17. Hare Krishna tiwari, Darsan sapkota , Malaya Ranjan Sen . High prevalence of multidrug – resistant MRSA in a tertiary care hospital of northern india . Infection and Drug Resistance, 2008; pp 57-61.
18. Olowe O. A., Eniola K.I.T., Olowe R.A., Olayemi A.B. Antimicrobial susceptibility and Beta-lactamase detection of MRSA in Osogbo. SW Nigeria. Nature and sciences 2007; vol.5(3).
19. Muhammad Saeed Anwar, Ghazala Jaffery, Khalil, Muhammad Tayyib, Shahid Raza Bokhari. Staphylococcus Aureus and MRSA nasal carriage in general population J Coll Physicians Surg Pak . 2004; vol.14:pp 661-664.
20. K B Anand , P Agrawal , S Kumar , K Kapila. Comparision of Cefoxitin Disc Diffusion Test , Oxacillin Screen Agar , And PCR For mecA Gene For Detection of MRSA. Indian Journal of Medical Microbiology 2009; vol.27(1): pp 27-29.
21. Annie Felten , Bernadette Grandry , Philippe Henri Lagrange, and Isabelle Casin Evaution of three techniques for detection of low – level methicillin – resistant *Staphylococcus aureus* (MRSA) : a Disk diffusion method with cefoxitin and Moxalactam , the Vitek 2 system , and the MRSA – Screen Latex Agglutination test . Journal of Clin. Microbiol 2002; vol .40 (8) : pp 2766-2771.
22. Jana M. Swenson, Fred C. Tenover . Results of Disk Diffusion Testing with Cefoxitin Correlate with Presence of mecA in *Staphylococcus* spp. Journal of Clin. Microbiol 2005; vol.43(8): pp 3818-3823.
23. Holferder m, Eigner U, Turnwald AM, witte w, weizenegger M,Fahr A. Direct detection of methicillin – resistant *Staphylococcus aureus* in clinical specimens by a nucleic acid-based hybridisation assay. Clin. Microbiol.Infect 2006; vol.12: pp 1163-1167.
24. Jones D, Speck M, Daschner FD, Grundmann H. Apid PCR based identification of methicillin – resistant *Staphylococcus aureus* from screening swabs. Journal of Clin. Microbiol 2002; vol.40: pp1821-1823.