African Journal of Microbiology Research Vol. 5(22), pp. 3716-3719, 16 October, 2011
Available online http://www.academicjournals.org/ajmr
DOI: 10.5897/AJMR11.518
ISSN 1996-0808 ©2011 Academic Journals

*Full Length Research Paper*

# The prevalence of methicillin and vancomycin resistant *Staphylococcus aureus* nasal carriage in large teaching hospital personnel

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Accepted 26 August, 2011

**S*taphylococcus aureus* is one of the most prevalent pathogens in nosocomial infections. We designed this cross-sectional study for evaluation of methicillin and vancomycin resistance in *S. aureus* strains that have been harbouring in the nasal nars of Shohadaie Ashayers hospital personnel (khorramabad, Iran). Samples were obtained by sterile cotton-wool swab moistened with normal saline rotation inside interior nares of 300 personnel and immediately point cultured on a section of mannitol salt agar. Suspected colonies confirmed by biochemical methods. Methicillin and vancomycin resistance of isolated strains was carried out by agar dilution according to recommendations of Clinical and Laboratory Standards Institute. Sixty-four (21.33%) out of 300 samples were nasal carriers for *S. aureus*; 16 (25%) and 4 (6.25%) of the carriers were methicillin resistant *S. aureus* and vancomycin intermediate *S. aureus,* respectively. One (0.33%) of the personnel was carrier for a strain that concurrently was methicillin and vancomycin resistant. No correlation was found between carriage with sex, age, ward and length of occupation, and predisposing diseases. Kind of occupation and level of education were significantly related with carrier state. This is the first report of *S. aureus* resistant strains from lorestan provinces of Iran. Percentage of *S. aureus* carriage in hospital personnel was consistent with other published reliable documents. Isolation of 4 vancomycin intermediate *S. aureus* and 1 vancomycin resistant *S. aureus* strains from studied personnel was the interesting findings of this study. Because of carrying of such resistant strains in hospital personnel and risks of transmission to patients it needs further attention of health officials.**

**Key words:** *S. aureus*, nosocomial infections, methicillin resistant *S. aureus* (MRSA), vancomycin resistant *S. aureus* (VRSA), vancomycin intermediate *S. aureus* (VISA).

## INTRODUCTION

MRSA is reported as a major nosocomial pathogen world wide since the introduction in 1961 (Brown et al., 2005). This strain is causing severe infections in both hospital and community settings (CDC, 1997a; 1997b; Borer et al., 2002; Lee et al., 2006).

After several years of introducing MRSA in the world, health associated diseases and syndromes related to them have been unknown in Iran's hospitals. In many of the hospitals there are not local or national guidelines of MRSA. So there is no suitable tracing or auditing tools for evaluation of resistant strains including MRSA, VRSA, or VISA. Some of the MRSA strains are Panton-Valentin-Leucocidin positive, and reported as a large health care associated outbreaks in Europe and other countries (Borer et al., 2002). MRSA strains colonizing of hospital personnel indicated as more antibiotic resistant and more clonal in origion (Cespedes et al., 2002). Predisposing

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**Abbreviations: MRSA,** Methicillin resistant *S.aureus;* **VRSA,** vancomycin resistant *S.aureus;* **VISA,** vancomycin intermediate *S.aureus;* **CLSI,** clinical and laboratory standards institute; **BHI,** brain heart infusion; **HCWs,** healthcare workers

factors including length of stay, surgery, prior hospitalization, and antibiotic use are main important risk factors of causing MRSA related infections in hospitalized patients (Cook et al., 2006).

On the other hand, recently emergence of VRSA complicated the treatment choices of related infections (CDC, 1997a; 1997b). Although it is not clear that these bacteria transmitted by hospital environments or not, eradication of them with conventional disinfectants nearly impossible (Hardy et al., 2007). Study of nasal carriers of *S. aureus* indicated that genetically and other unknown factors predispose individuals as a host for bacterium (Marjolein et al., 1999). There is no clear epidemiology of methicillin and vancomycin *S. aureus* resistance in Iran, and some published documents indicates existence of VISA, MRSA and even VRSA in our country (Tabbarai et al., 2001; Naderinasab et al., 2003; Saderi and Owlia, 2005; Aligholi et al., 2008). Considering the known risks of resistant Staphylococci that could be spread from personnel to patients and other persons, we designed this study for evaluation of MRSA and VRSA carriage in a university affiliated and the largest teaching hospital personnel of Lorestan province, Iran.

## MATERIALS AND METHODS

Samples were obtained by sterile cotton-wool swab moistened with normal saline rotation inside interior nares of 300 personnel and immediately point cultured on a section of Mannitol Salt Agar (Difco Laboratories, Detroit, Michigan). For economic purpose plates was divided into 8 sections, and each of the sections specified for one of the personnel's with a code number. And results traced with code numbers for convenience. After 48 to 72 h incubation in 37°C presumptive colonies (Gram positive cocci, yellow pigmented, and Catalase positive) selected for further confirmation. Suspected colonies isolated on Sheep Blood Agar and confirmed by conventional methods (Slide Coagulase, DNase, Novobiocin disk).

### Methicillin-resistant *S. aureus* (MRSA) screening

Confirmed strains screened with oxacillin 1 mg disks in Kirby-Bayer method, and zone diameters of ≤ 10 mm considered as candidates of MRSA. According to recommendations of Clinical and Laboratory Standards Institute (CLSI), Mueller-Hinton agar plates supplemented with NaCl (Sigma 4% w/v; 0.68 mol/L) and 6 µg/ml of oxacillin (Sigma) were used for phenotypic confirmation of MRSA (Cespedes et al., 2002). Briefly bacteria cultured in BHI Agar (Difco Laboratories, Detroit, Michigan). 35°C 18 h. 0.5 McFarland dilutions prepared and cultured with sterile swabs in a small section of plates. Growth of one or more than one colony or any light growth considered as a MRSA strain. Culture plates monitored for 72 h after incubation for any growth.

### Vancomycin -resistant *S. aureus* (VRSA) screening

VRSA strains were screened on brain heart infusion (BHI ) agars containing 1, 2, 4, 6, 8, 16, 32 and 64 µg/ml of vancomycin ( Vanko, Jaber Ebn Hayyan pharmaceuticals, Iran) as follows: A swab dipped in the 0.5 McFarland suspensions of isolated *S. aureus* strains, after expressing the excess liquid, plates were inoculated (final concentration=$10^6$ CFU/ml). *E. faecalis* ATCC 29212 and *E. faecalis* ATCC 51299 were used as susceptible and resistant control strains respectively. Based on Clinical and Laboratory Standards Institute recommendations, strains growing < 2, 4 to 8, and ≥16 µg/mL of vancomycin considered as sensitive, intermediate VISA and resistant VRSA to vancomycin, respectively (Tenover et al., 2004; CLSI, 2005; 2005; 2006). In addition of carriage study, each personnel also completed a questionnaire considering age, sex, occupation, level of education, and some general signs including recurrent boils in the face and other sites of body, nasal itching, and headache.

## RESULTS

Sixty-four (21.33%) of 300 studied personnel were nasal carriers for *S. aureus*; (Table 1) 16 (25%) of the carriers were MRSA. 4 (6.25%) of the isolates were VISA. One (.33%) of the personnel of laboratory was carrier for a strain that concurrently resistant against methicillin MRSA and vancomycin VRSA. No correlation was found between carriage, sex and age. 39.58 and 12.5% of low level educated and high level educated (doctors) were carriers for *S. aureus* and MRSA, respectively. Although there are no clinical evidences that *S. aureus* carriage poses the individuals on a minor difficulties, but we found that some of the studied personnel suffering from frequent discomfort in face, head and neck, headache, dryness and itching of nasal canal.

## DISCUSSION

In previous decade's resistant strains of *S. aureus* including MRSA, VRSA and VISA strains reported from all over the world (CDC, 1997a; 1997b; Borer et al., 2002; Brown et al., 2005; Lee et al., 2006). Gaspar et al. (1992) in their study indicated that health care workers could play important role in nosocomial outbreaks of MRSA infections (Gaspar et al., 1992). MRSA may be a management dilemma for healthcare workers (HCWs) with cystic fibrosis (Downey et al., 2005).However some studies indicated that HCWs could play as a source of MRSA outbreaks in patients (Lessing et al., 1996; Borer et al., 2002). On the other hand eradication of such resistant strains by routine hospital methods are both controversial and difficult (Hansen et al., 2007; Wendt et al., 2007). But in rare cases eradication of MRSA reported by expensive methods (Boer et al., 2006). Some studies reported that the emergence of vancomycin resistant subclones of *S. aureus* strains induced in the presence of vancomycin with dependency to concentration of vancomycin (Hiramatsu et al., 1997). In this study authors emphasized that prevalence of MRSA with simultaneously resistance to vancomycin was higher in university hospitals that non- university hospitals or clinics (Hiramatsu et al., 1997). This subject reflects the essentia l role of university hospitals in induction and

**Table 1.** Indicating percentage and number of carriers and isolated resistant strains of S.aureus from interior nares of personnel.

| Total cases | *S. aureus* carriers | MRSA carriers | VISA carriers | VRSA+MRSA carriers |
|---|---|---|---|---|
| 300 | 64 | 16 | 4 | **1** |
| 100% | 21.33% | 5.33% | 1.33% | .33% |

distribution of resistant strains in hospital and even community environments. So carrier state of our teaching hospital personnel evaluated as a risk factor of their role in outbreaks. Because of importance of such strains carrier state of HCWs of Shohadai Ashayer hospital have been evaluated in this study and the existence of VRSA, VISA and MRSA assessed by phenotypic methods. Previous studies indicated that in-house made Muller-Hinton agars containing 5 micro of vancomycin per ml was both sensitive and specific for screening of GISA strains.

It is also cost effective and suitable for large population screenings such as hospital personells. But its confirmation is controversial (Hubert et al., 1999). Sancak et al. (2005) in their investigation of VISA and hetero-VISA among 256 MRSA strains found that confirmation of screened strains on BHI agar supplemented with vancomycin needs further methods such as macro Etest and population analysis. They found no VISA from 256 MRSA strains, but 46(17.97%) of 256 MRSA strains were identified with reduced susceptibility to vancomycin (Sancak et al., 2005). Tiwari and Sen (2006) in their study indicated that out of 783 *S. aureus* two strains have been vancomycin and teicoplanin resistant (one strain with MIC 32 microg/ml and the other strain with MIC 64 microg/ml) and six strains have been vancomycin intermediate. All of the VRSA and VISA strains in this study simultaneously had been grown on BHI vancomycin screen agar (vancomycin 6 microg/ml) and were positive for mecA (Tiwari and Sen, 2006). Kim et al. (2006) in a nationwide surveillance program for VISA and VRSA in Korea indicated that out of 3756 MRSA, 18 (0.5%) had a vancomycin MIC of 4 micro/ml. They reported no strains of VRSA or VISA (Kim et al., 2006). Komijo et al. (2005 Had screened 734 medical staff (356 doctors and 378 nurses) at Yamanashi University Hospital and reported 35 medical staff (12 doctors 3.4% and 23 nurses 6.1%) as MRSA nasal carriers. They proposed MRSA nasal colonization as a risk factor of nasal diseases such as nasal allergy or chronic sinusitis. However there is not enough scientific evidences for confirmation of this hypothesis (Komijo et al., 2005).

Based on the recommendations of CLSI our screening of hospital personnel indicate that one of the laboratory staff have been carried *S. aureus* with vancomycin MIC of ≥16 µg/mL, and 4 of the personnel carried strains with vancomycin MICs of 4 to 8 µg/mL. There is not enough research about resistant strains of *S. aureus* in Iran. *S. aureus* cases with reduced susceptibility to vancomycin have been firstly isolated in Mashhad province of Iran (Naderinasab et al., 2003), after that there has been a report of VRSA from Tehran. In this study out of 139 strains, 5 isolate had a vancomycin MIC of ≥ 128 by agar dilution and E-test methods (Saderi and Owlia, 2005). Tabbarai and colleagues in their study of 1193 nasal swabs of school children (6 to 12 years old) reported that 194 samples (16.3%) were *S. aureus* carriers. Antibiotic resistance pattern of isolates showed that 34.8 and 1.7% of samples were resistant to methicillin and vancomycin, respectively (Tabbarai et al., 2001). Different resistant rates in reported studies in some extent depend on study groups, sampling and antibiogram methods. 4 of 16 MRSA isolates in this study were simultaneously VISA. Because of high prevalency of MRSA and some VISA strains in Shohadai- Ashayer hospital personnel, it has been necessary that, individual and hospital laboratories devise evaluated MIC methods for all of the *S. aureus* isolates from viewpoint of methicillin and vancomycin resistance.

One of the interesting results of this study was the prevalence of some of the general sign and symptoms in nasal carriers of *S. aureus* such as frequent discomfort in face, head and neck, headache, dryness and itching of nasal canal. These results forms a hypothesis in mind that carrier state for *S. aureus* does not a simple colonization, But may be as a predisposing factor for opportunistic infections of carriers in special conditions or even in healthy persons. One of the interpretations of VISA strains existence in personnel of our hospital is that many of the physicians of hospital prescribe vancomycin as an important empirical antibiotic in critical care patients. This point is also explains many of treatment failures related to MRSA infections. As we know one of the best choices for treatment of MRSA infections is vancomycin. Emergence of VISA and VRSA alerts health care professionals for alternative treatments of MRSA infections.

## Conclusion

The present study is the first report of VRSA and VISA from one of the western provinces of Iran. In fact our results needs to be confirmed with molecular typing methods, but phenotypic confirmation of such resistant strains indicate that health care professionals must control the use of antimicrobials including vancomycin as an inducing factor of resistance, considering infection

control guidelines, and develop laboratory identification methods for screening and exact study of resistant strains incidence and prevalence in such settings.

**REFERENCES**


Aligholi M, Emaneini M, Jabalameli F, Dabiri H, Sedaght H (2008). Emergence of High-Level Vancomycin-Resistant *Staphylococcus aureus* in the Imam Khomeini Hospital in Tehran. Med. Princ. Pract., 17: 432-434.

Boer HEl, Elzelingen-Dekker CMV, Rheenen-Verberg CMF, Spanjaard L (2006).Use of Gaseous Ozone for Eradication of Methicillin-Resistant *Staphylococcus aureus* From the Home Environment of a Colonized Hospital Employee. Infect. Control. Hosp. Epidemiol., 27: 1120-1122.

Borer A, Gilad J, Yagupsky P, Peled N, Porat N, Trefler R, Shprecher-Levy H, Riesenberg K, Shipman M, Schlaeffer F (2002).Community-Acquired Methicillin-Resistant *Staphylococcus aureus* in Institutionalized Adults with Developmental Disabilities. Emerg. infect. Dis., 8(9):966-970.

Brown DFJ, Edwards DI, Hawkey PM, Morrison D, Ridgway GL, Towner KJ, Wren MWD (2005). On behalf of the Joint Working Party of the British Guidelines for the laboratory diagnosis and susceptibility testing of methicillin-resistant *Staphylococcus aureus* (MRSA). J. Antimicrob. Chemother., 56: 1000-1018.

CDC (1997a). Reduced susceptibility of *Staphylococcus aureus* to vancomycin—Japan. Morbid. Mortal. Weekly Rep., 46: 624-626.

CDC (1997b). *Staphylococcus aureus* with reduced Susceptibility to vancomycin—United States. Morbid. Mortal. Weekly Rep., 46: 813-815.

Cespedes C, Miller M, Quagliarello B, Vavagiakis P, Klein RS, Lowy FD (2002). Differences between *Staphylococcus aureus* Isolates from Medical and Nonmedical Hospital Personnel. J. Clin. Microbiol., 40: 2594-2597.

CLSI (2005). Performance standards for antimicrobial susceptibility testing. CLSI approved standard M100-S15. Clinical and Laboratory Standards Institute. Wayne, PA. , CLSI.

CLSI (2005). Performance standards for antimicrobial susceptibility testing. CLSI approved standard M100-S15. Wayne, PA.

CLSI (2006). Performance Standards for Antimicrobial Susceptibility Testing. M100-S16. Clinical and Laboratory Standards Institute/NCCLS. Wayne, PA, CLSI.

Cook PP, Catrou P, Gooch M, Holbert D(2006). Effect of reduction in ciprofloxacin use on prevalence of meticillin-resistant *Staphylococcus aureus* rates within individual units of a tertiary care hospital. J. Hospital Infect., 64: 348-351.

Downey DG, Kidd TJ, Coulter C, Bell SC (2005). MRSA eradication in a health care worker with cystic fibrosis; re-emergence or re-infection?. J. Cystic Fibrosis. 4: 205-207.

Gaspar M, Uribe P, Sánchez P, Coello R,Cruzet F (1992). Hospital personnel who are nasal carriers of methicillin-resistant *Staphylococcus aureus*. Usefulness of treatment with mupirocin. Enferm. Infect. Microbiol. Clin., 10: 107-110.

Hansen DPP, Werfel U, Benner D, Brauksiepe A, Popp W (2007). Success of MRSA eradication in hospital routine: depends on compliance. Infect., 35(4):260-264.

Hardy KJ, Gossain S, Henderson N, Drugan C, Oppenheim AB, Gao F, Hawkey PM (2007). Rapid recontamination with MRSA of the environment of an intensive care unit after decontamination with hydrogen peroxide vapor. J of Hospital Infect., 66: 360-368.

Hiramatsu K, Aritaka N, Hanaki H, Kawasaki S, Hosoda Y, Hori S, Fukuchi Y, Kobayashi I (1997). Dissemination in Japanese hospitals of strains of *Staphylococcus aureus* heterogeneously resistant to vancomycin. Lancet 50: 1670-1673.

Hubert SKMJ, Fridkin SK, Gaynes RP, McGowan JEJR, Tenover FC (1999). Glycopeptide-intermediate *Staphylococcus aureus*: evaluation of a novel screening method and results of a survey of selected U.S hospitals.J. Clin. Microbiol., 37(11):3590-3593.

Kim HB, Lee YS, Kim BS, Cha JO, Kwon SU, Lee HJ, Suh JT, Rheem I, Kim JM, Shin BM, Kim MN, Lee K, Lee CS, Kim EC, Oh MD, Choe KW (2006). Prevalence and clinical implications of *Staphylococcus aureus* with a vancomycin MIC of 4 microg/ml in Korea. Microb. Drug. Resist., 12: 33-38.

Komijo A, Eiko Y, Goro T, Zensei M, Tomokazu M, Takashi U, Keisuke M (2005). Prevalence Rate of Nasal Disease among MRSA Nasal Carriers. Japanese J. Rhinol., 44: 127-130.

Lee N, Clive SC, Grace L, Rebecca L, Edman L, Raymond L, Margaret I (2006). Community Case of Methicillin-resistant *Staphylococcus aureus* Infection. Lett. Emerg. infect. Dis., 12:172-74..

Lessing MPA, Jordens JZ, Bowler ICJ(1996).When should healthcare workers be screened for methicillin-resistant *Staphylococcus aureus*? J. Hospit. Infect., 34: 205-210.

Marjolein FQ. VandenBergh EdPF, Yzerman, Alex VB, Hélène AMB, Marly S, Henri AV (1999). Follow-Up of *Staphylococcus aureus* Nasal Carriage after 8 Years: Redefining the Persistent Carrier State. J. Clin. Microbiol., 37: 3133-3140

Naderinasab M, Ghanaat J, Fatehmanesh P (2003). An investigation on the daily increasing resistance of the *Staphylococcus aureus* against vancomycin. The 5th Iranian Congress of Microbiology, Ahwaz.

Saderi H, Owlia P (2005). Vancomycin resistance among clinical isolates of *Staphylococcus aureus*. Arch. Iranian Med., 8: 100-103.

Sancak B, Ercis S, Menemenlioglu D, Colakoglu S, Hasçelik G (2005). Methicillin-resistant *Staphylococcus aureus* heterogeneously resistant to vancomycin in a Turkish university hospital. Antimicrob. Chemother., 56: 519-523.

Tabbarai A, Ghaemi E, Fazeli M, Bakhshandeh N, Behnampour N, Basori M (2001). Prevalence of *Staphylococci aureus* nasal carrier in healthy school students in Gorgan. Gorgan J. Med. Sci., 8(3): 11-16

Tenover FC, Weigel LM, Applebaum PC, McDougal LK, Chaitram J, McAllister S, Clark N, Killgore G, O'Hara CM, Je L, Patel JB, Bozdogan B (2004). Vancomycin-resistant *Staphylococcus aureus* isolated from a patient in Pennsylvania. J. Antimicrob. Chemother., 48: 275-280.

Tiwari H, Sen M (2006). Emergence of vancomycin resistant *Staphylococcus aureus* (VRSA) from a tertiary care hospital from northern part of India. . BMC Infect. Dis., 26: 156.

Wendt C, Schinke S, Württemberger M, Oberdorfer K, Bock-Hensley O, Von Baum H (2007). Value of whole-body washing with chlorhexidine for the eradication of methicillin-resistant *Staphylococcus aureus*: a randomized, placebo-controlled, double-blind clinical trial. Infect. Control. Hosp. Epidemiol., 28: 1036-1043.