Methicillin Resistant *Staphylococcus Aureus* in Ventilator Associated Pneumonia in Toxicological Intensive Care Unit

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ABSTRACT

Background: Methicillin resistant *Staphylococus Aureus* (MRSA) is a cause of nosocomial infections at intensive care unit (ICU), which imposes a high mortality and morbidity on the health care systems.
The objective of this study was to evaluate the role of MRSA in patients with clinically suspected ventilator associated pneumonia (VAP) in toxicological ICU admitted patients.

Methods: This cross-sectional study was performed over a period of six months from August 2009 to February 2010. A total of 84 patients with clinically suspected VAP were selected from all 381 ICU admitted patients under mechanical ventilation for more than 48 hours. MRSA Screen Agar was used to detect resistance in *Staph aureus* specimens. MRSA was determined as the main outcome.

Results: MRSA was the cause in 54% of *Staph aureus* infected VAPs. Although MRSA infection was not significantly associated with age, gender, cause of poisoning, chronic disease, paraclinical findings, length of hospital stay, and antibiotic prescription ($P>0.05$ for all comparisons), it was reported higher in those who expired than those who survived (66.7% vs. 31.9%, $P<0.012$).

Conclusion: In the main referral toxicological ICU in Tehran, in more than 1 of 3 clinically suspected VAP cases, MRSA was seen which was associated with the poorer outcome, higher inpatient mortality.

Keywords: Intensive Care Unit**,** Methicillin Resistance, Pneumonia, *Staphylococus Aureus*, Ventilator.

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INTRODUCTION

During the past decade, changes of antimicrobial agents' resistance patterns and epidemiology of this have emerged [1, 2]. One of the most important examples is methicillin resistant *Staphylococus Aureus* (MRSA) which is any strain of *Staphylococcus Aureus* bacteria that is resistant to beta-lactamase antibiotic and their derivatives [3].

MRSA is a multi-drug resistant pathogen which poses significant burdens in terms of both community-acquired and nosocomial infections [4]. For community-acquired infection, high risk groups are the homeless, those who play close-contact sports, military personnel, men who have sex with men and injecting drug users [5]. MRSA develops from skin and soft tissues to systemic infections such as septicemia and pneumonia [6].

Hospital acquired MRSA infection can happen when one strain is transmitted to other patients or through close contacts of infected persons or transferring from hospital settings. Previous antibiotic coverage is another reason of spreading MRSA [2, 7]. Spread of many antimicrobial-resistant pathogens in ICU situation is because of its unique nature [2].

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Nosocomial pneumonia includes hospital-acquired pneumonia, ventilator-associated pneumonia (VAP), and healthcare associated pneumonia [8]. VAP is a pulmonary infection that occurs after at least 48 hours of mechanical ventilation in the intensive care unit (ICU) with *Pseudomonas aeruginosa, Staphylococcus aureus,* and *Enterobacter* as the most common causes. Nevertheless, VAP distribution varies in different countries and hospitals [9, 10]. MRSA is an increasing reason of VAP worldwide. The prevalence of MRSA in VAP due to *Staph aureus* is 50% [7, 10, 11] and VAP mortality rate related to MRSA is high. Incidence of MRSA infection in ICU patients in some European countries is as high as 30 % [12].

The aim of this investigation was to determine the role of MRSA in clinically suspected cases of VAP in ICU of Loghman Hakim Hospital, a tertiary referral hospital in Tehran, Iran. Such information benefits effective antibiotic therapy in these patients.

MATERIALS AND METHODS

Design and setting

This prospective study was performed over a period of six months from August 2009 to February 2010 in the Toxicological Intensive Care Unit at the Loghman Hakim Hospital Poisoning Center- the unique referral care center of poisoning in Tehran, Iran. This center serves nearly 20,000 poisoned patients each year. Daily turnover of the inpatients in this center is 80-100 patients.

Participants and sampling

Patients were enrolled in study based on inclusion criteria, such as at least 48 hours of being on mechanical ventilation with clinically diagnosis of suspected VAP. Patients with AIDS, lung cancer, chronic obstructive pulmonary disease and patients who received antibiotics before 24 hours of admission were excluded from the study.

Clinical diagnosis of VAP was done in the presence of persistent or progressive radiographical infiltration and at least two of the following criteria: 1) temperature higher than 38° C or lower than 35 ° C, 2) leukocyte count higher than 10000/μL or lower than 4000/μL, 3) presence of new purulent respiratory secretion or any changes in sputum, 4) positive blood cultures or pleural effusion cultures, 5) detection of rales or dullness on chest examination, and 6) at least 10% decrease in arterial PO_2 [13].

Measures and measurements

Data were collected by an infectious disease specialist and a well trained ICU nurse. Age, sex, mental status by Glasgow coma scale, and type of poisoning were collected as baseline. For each case, endotracheal aspirate samples were performed on the basis of the standard procedure.

Specimen collection and microbial sampling

All of the patients underwent non-protected endotracheal aspiration (NPEA) by a 12 F suction catheter gently guided through the endotracheal tube for a 24cm length followed by injection of 2-5 ml saline into endotracheal tube and aspiration. Then the specimens were immediately sent to laboratory for microbiological processing. The diagnostic threshold for non-protected tracheal aspiration (NPTA) was 10^5cfu/mL.

All of the specimens were mechanically liquefied and homogenized by mixing with vortex for 1 minute and then centrifuged for 10 minutes. All of the samples were gram-stained for assessment of the type of putative bacteria and evaluation of intracellularity. Specimens were, then, plotted on 5% sheep blood agar, Manitol salt agar, and EMB agar. The plates were incubated over night at 37ºC. After preliminary characterization of the isolated bacteria by gram stain and colony morphology, species identification was done. Then MRSA screen agar was used to detect resistance in *Staph aureus* specimens. The detailed methodology of the latter was as follows:

1) Suspension of well-isolated colonies of organisms from an 18-24 plate culture into a tube of Trypticas soy broth or normal saline and adjustment of turbidity to a 0.5 McFarland standard, 2) Spot inoculation of the above suspension to screen agar, 3) Incubation at 35 ºC, and 4) Examination after

24h; plates were quality controlled by *Staphylococcus aureus* ATCC standards.

Ethical considerations

The study protocol with code number of 87-01-113-6156 was approved in 2009 by the research ethics committee of Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Statistical analysis

The statistical package for social sciences (SPSS version 16) was used to perform statistical analysis. Data were analyzed by Chi-square, Fisher's Exact Test, and independent samples t-test where appropriate. The alpha level of significance was set at 0.05.

RESULTS

Of 84 patients with suspected VAP, 62 (73.8%) were males. The mean age of the patients was 40.6 ± 16.4 with the age range of 16-80 years. Overall, 15 patients (17.9%) had a history of chronic diseases. Most patients were in deep coma (n=59, 70.2%).

The reasons for ICU admission were poisoning by antidepressant tablets in 55 patients (65.5%), opioids in 28 cases (33.3%), and organ phosphorus toxins in 1 case (1.2%). WBC counts in 64 patients (76.2%) were reported 12000/µL to 25000/µL and in 2 patients were up to 25000/µL. Left shift was detected in 83 cases.

Protocol of antimicrobials prescription for patients, length of stay at hospital, and inpatient mortality can be seen in Table 1.

Fifty nine (58.7%) tracheal cultures were positive for *Staphylococcus aureus*, followed by *Pseudomonas aeruginosa* (Table 2). MRSA was seen in 32 cases (38.1%) of the suspected VAPs.

Although MRSA infection was not significantly associated with age, gender, cause of poisoning, chronic disease, paraclinical findings, length of hospital stay, and antibiotic prescription (P>0.05 for all comparisons), it was reported higher in those who expired than those who survived (66.7% vs. 31.9%, P=0.012)

Table 1. Healthcare delivery to patients with suspected VAP in Loghman Hakim Hospital Toxicological Intensive Care Unit.

Antibiotic	VAP patients No.(%)
Meropenem+Vancomycin	20 (23.8%)
Meropenem+Vancomycin+amikacin	61 (72.6%)
Ciprofloxacin + Vancomycin	1 (1.2%)
Ceftriaxone+ Clindamycin	2 (2.3%)
Length of ICU stay	
1	8 (9.5)
2	28 (33.3)
3	27 (32.1)
4	14 (16.7)
5	3 (3.6)
6	1 (1.2)
7	1 (1.2)
8	2 (2.4)
Outcome	
Died	15 (17.8)
Survived	69 (82.2)

Table 2. List of isolated microorganisms detected in VAP.

Microorganism	No. (%)
Staphylococcus aureus	59 (70.2)
Pseudomonas aeruginosa	3 (3.6)
Klebsiella pneumoniae	2 (2.4)
Enterobacter	1 (1.2)
Acinetobacter	2 (2.4)
Other gram-negative bacilli	3 (3.6)
Sterptococcus pneumoniae	1 (1.2)
Candida albicans	2 (2.4)
Staph epidermidis	1 (1.2)
No growth	10 (11.9)

DISCUSSION

In patients with *Staph aureus* infected VAP, the cause wass MRSA in 54%, which was associated with higher inpatient mortality. This supports the results of other studies [10]. According to a recent study on microbial etiology of definitive VAP with positive cultures in ICU of Loghman Hakim Hospital, *Staphylococcus aureus* was the most common organism in about 60%; however, the rate of MRSA was not determined [13]. According to another recent study in this hospital, MRSA was reported to be

responsible for about 90% of staphylococcal infections at ICU [14]. National Nosocomial Infections Surveillance (NNIC) system reports a higher prevalence of antibiotic-resistant strains such as MRSA in ICU patients than in other patient populations [15]. In another study, *Staphylococcus aureus* and *Klebsiella pneumonia* were found as the most common causes of aspiration pneumonia [16].

In different regions, the frequency of MRSA from total infections has been reported to be in a wide range, from less than 1% in Netherlands to more than 65% in Japan [7,11].

Resistant agents may cause ineffectual therapy of VAP patients which increase mortality and morbidity rate, length of stay, and cost [9, 12, 15, 17]. In the present study, MRSA increased the mortality, but not the length of stay.

MRSA is considered as an important pathogen in patients with VAP in ICU settings and this is because of the difficulties with treatment and infection controls [1, 7, 12, 15].

Incidence of VAP depends on various factors, most notably the host and duration of mechanical ventilation, and it varies between 9% and 70% (average: 20% to 25%). *Staphylococcus aureus* is, however, among organisms which can increase its occurrence in ICU [18, 19]. NNIS system reports that mean rates of VAP is 5.4 per 1,000 mechanical ventilator days [20]. Therefore, information about the resistance patterns and microbial epidemiology in each ICU is of importance for appropriate selection of antibiotics [12, 21, 22].

This study had its own limitations, from low sample size to lack of genetic study of MRSA and no assessment of sensitivity to other antibiotics. However, it was conducted in a unique referral center in the country with a high patient turn over, and this increased the importance of the results.

CONCLUSION

In the main referral toxicological ICU in Tehran, MRSA was the pathogen in more than 1 of 3 clinically suspected cases with VAP and it was associated with the outcome which is higher inpatient mortality.

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