FREQUENCY OF METHICILLIN-RESISTANT STAPHYLOCOCCUS AUREUS (MRSA) IN HEALTHY NASAL CARRIERS IN POMORAVLJE DISTRICT

Ljiljana Petrović Jeremić

S. aureus is a frequent cause of infections both in hospital setting and outside it. The growing resistance of this pathogen to beta-lactam and other antibiotics complicates the treatment. The aim in this study was to investigate the frequency of nasal carriage of S. aureus among healthy population who can be the source of infection, as well as a share of MRSA colonisation.

We analyzed 5,776 throat and nose swab samples taken from adult healthy population; the swabs were analyzed in microbiology labs of the Public Health Institute Ćuprija. The isolates were identified by standard procedures based on clumping factor production and using manitol-salt agar with novobiocin disk. Methicillin resistance was determined by disk-diffusion with a 30 mcg cefoxitin disk.

The prevalence of nasal MRSA colonisation was 3,63%; 4,29% of subjects were nasal carriers of S. Aureus. The values obtained in this paper are in keeping with data obtained in other European countries. The prevalence of MRSA among carriers is low, and the treatment to complete absence of germs and implementing measures to prevent the spread of infection are therefore urgently needed. Acta Medica Medianae 2010;49(1):33-36.

Key words: staphylococcus, nasal carriage, MRSA

Introduction

In the recent years, worldwide, the data on distribution and spread of MRSA (methicillin-resistant Staphylococcus aureus) both in hospital setting and outside it have been collected, which poses a threat to the whole human community. Based on the data of the Center for Disease Control (Centers for Disease Control and Prevention-CDC), it has been estimated that the number of MRSA infections has doubled in the last 10 years, and that the number of deaths in the United States owing to these infections is higher than the number of deaths from AIDS (1).

Among the MRSA strains of hospital and community origin, besides similarities, there are many differences (2): epidemiological, clinical, according to the virulence factors and frequency of occurrence (3,4). With regard to hospital isolates, CA-MRSA (community-asssociated MRSA) isolates are still significantly less distributed (up to the nineties of the last century MRSA was found only among hospital patients and hospital staff), although there are predictions that the difference will decrease in the future (5). CA-MRSA isolates are more virulent, spread quickly and can cause more severe infections than HA-MRSA (hospital-acquired MRSA) because they possess genes that encode toxins and other virulence factors (PVL-genes leukocidin, enterotoxin) (2). However, they are sensitive to the majority of antibiotics (quinolones, sulfonamides, lincozamides, tetracyclines, fusidin acid), and CA-MRSA infection (infection of the skin and soft tissue, osteomyelitis, necrotizing pneumonia) are more easily treated.

Bearing in mind that in our country there are very few studies on the frequency of staphylococcal carrier state in healthy individuals who may be a significant source of infections, it would be interesting to analyzed the distribution of healthy carriers and the proportion of MRSA in healthy population in the district of Pomoravlje.

Aim

In our country, as well as the worldwide, there are very few studies about the staphylococcal nasal carriage status among the healthy individuals who can be a source of infection (carriers who are employed in the food production and distribution industry). The purpose of this study was to investigate the distribution and share of MRSA carriage state in the healthy population in the Pomoravlje district which consists of 6 municipalities: Jagodina, Cuprija, Paracin, Rekovac, Despotovac and Svilajnac, and the set goals were the following:
1. Collection and identification of Staphylococcus aureus isolates from throat and nasal swabs from healthy individuals capable to work.  
2. Testing sensitivity of isolated staphylococcus to methicillin.

## Material and methods

The data used for the present study are based on the examination of 5.766 samples (throat and nasal swabs), collected from healthy, employed individuals aged 16-60 years who, because of the nature of work, were subjected to health surveillance (pursuant to the Law on the protection of population from infectious diseases -Official Gazette of the Republic Serbian 125/04).

In the microbiological laboratory of the Public Health Institute Cuprija, all throat and nasal swab specimens were inoculated on blood agar (Torlak, Belgrade). The plates were incubated overnight at 35º C in aerobic conditions, and examined the next day for colonies suggestive of S.aureus. They were tested by coagulase test tube with rabbits plasma (Torlak, Belgrade) using tablets of Novobiocyn (ROSCO, Danemark). Testing sensitivity of staphylococci to methicillin was performed by disk diffusion method in accordance with the requirements of Clinical and Laboratory Standard Institute (CLSI): suspension of bacterial culture density 0.5 McFarland was inoculated on Mueller Hinton agar ( Torlak, Belgrade), and after 10 minutes the following antibiogram tablets were placed: penicillin (10 ij), cefoxitin (30 mcg), gentamicyn, chloramphenicol, and sulfometoxasol-trimetoprim (Torlak, Belgrade). After 24-hour incubation at 35º C, the zones around the tablets were interpreted in accordance with standards of CLSI. Penicillin sensitive S.aureus isolates were investigated on the production of beta-lactamases by nitrocefin test (bio-Meriux): Nitrocefin disk soaked in distilled water becomes violet for 30 seconds, if penicillin-sensitive staphylococcus is applied.

### Results

From 5.766 examined samples, in 248 (4.29%) nose swabs Staphylococcus aureus was isolated. Methicillin resistance was shown in 9 (3.63%) isolates, (Table 1).

#### Table 1. Distribution of staphylococcal MRSA carrier state and the share of MRSA in employees subjected to sanitary inspection

<table>
<thead>
<tr>
<th>Number of samples</th>
<th>S. aureus number (%)</th>
<th>MRSA number (%)</th>
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</thead>
<tbody>
<tr>
<td>5.776</td>
<td>248 (4.29)</td>
<td>9 (3.63)</td>
</tr>
</tbody>
</table>

Except to beta-lactams antibiotics, we tested susceptibility of S.aureus to gentamycin, chloramphenicol and sulfometoxasol-trimetoprim: staphylococci from nasal throats were the most sensitive to gentamycin - 83,8%, to chloramphenicol – 78,2%, and to SXT - 67,7%, (Table 2).

#### Table 2. Susceptibility of S.aureus to gentamycin, chloramphenicol, SXT

<table>
<thead>
<tr>
<th>Isolates of S.aureus (sensitive)</th>
<th>gentamycin number (%)</th>
<th>gentamycin number (%)</th>
<th>gentamycin number (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>208</td>
<td>83,8</td>
<td>194</td>
</tr>
</tbody>
</table>

There were 8 (3,22%) S.aureus isolates sensitive to penicillin, and 5 (2,01%) nitrocefin negative staphylococcus isolates (Graph 1).

#### Graph 1. The frequency of penicillin sensitive S.aureus determined by nitrocefin test

### Discussion

S.aureus is the leading cause of community infections, and one of the common causes of hospital bacteriemia. The problem of S.aureus resistance to antimicrobial drugs is very challenging, because it involves the resistance to all beta-lactam antibiotics that are now in clinical use. This mechanism of resistance is associated with double increase of treatment costs and increasing mortality. In the beginning, MRSA used to occur only in large hospitals, but nowadays, it is more and more frequent cause of colonization and infection in smaller hospitals as well as community environment. Patients who were colonized in the hospital after discharge may colonize and infect patients in community. In contrast, the patients infected and colonized in community can introduce MRSA into hospital (6). From all over the world arrive daily reports on CA-MRSA strains as causes of hospital bacteremia and other infections. They are characterized, except for resistance to all penicillin and cephalosporin antibiotics , as well carbapenemes, by good sensitivity to «older» groups of antibiotics: aminoglycosides, sulfonamides, lincosamides (7).

Investigation of staphylococcal carrier state was conducted in a small number of countries. The data differ both by the frequency of carrier state and the representation of the MRSA carrier state. CA-MRSA prevalence in the European countries is generally low (1,5-9%) (3,8-10), and data were obtained by examining the nasal carrier state among healthy population (children, soldiers, healthy adults who in the last 12 months had no contact with the hospital centers).

In the U.S.A, among healthy population, the incidence of MRSA is around 2% (11,12). In our country, the frequency of MRSA in healthy carriers is around 4% (1), and that is in keeping
with the data from the reference literature. According to antibiotics from other groups, for example, aminoglycosides, quinolones, sulfonamides, community MRSA isolates showed good sensitivity (13-15), which is evident from this paper. However, most foreign authors recommended rifampicyn and mupirocin for the eradication of staphylococcal nasal carrier state (12,16). It is well known that long ago penicillin lost the primacy in the treatment of staphylococcal infections. The ability to create the inducible enzymes - penicilinase, which dissolves beta lactam ring of penicillin, spread quickly among the members of the genus Staphylococcus, and today the percentage of penicillin-sensitive staphylococcus is less than 5% (Graph 1).

The risk of transmission of MRSA from the carriers to healthy population outside hospital setting is low. However, the people whose hospital stay was long, and who were treated with antibiotics, then patients on dialysis and surgical patients and, in general, the immuno-suppressed are susceptible to infection. Nasal carriers of S. aureus, particularly those associated with the production and distribution of food, must be away from work and treated until decolonisation. However, after a local application of antimicrobial drugs, MRSA can persist or colonize some extranasal sites, or can during the treatment develop resistance to previously sensitive agent. Therefore, it is safest to take simple but effective measures to prevent the spread of MRSA, from personal hygiene (particularly hands) and hygiene of working places.

**Conclusion**

The data presented in this study show that in the healthy, employed population in the district of Pomoravije the percentage of nasal carrier state is 4.29%, of which 3,63% of staphylococci are resistant to methicillin. According to these values, we are not different from the rest of our country as well as other European countries. The prevalence of MRSA among carriers is still low, but with a tendency to increase; therefore, the treatment to decolonosation and implementation of measures to prevent the spread of infection are necessary.

**References**

УЧЕСТАЛОСТ МЕТИЦИЛИН-РЕЗИСТЕНТНИХ СТАФИЛОКОКСА УПОРЕДИ СТАФИЛОКОКСА У ЗДРАВИХ КЛИКОША У ПОМОРАВСКОМ ОКРУГУ

Лилiana Петровић Јеремић

Стапфилококс ауреус је чест узрок инфекција у болници, али и ван нje. Нjегова rастућа rezистенциja на бетa-лактамске и дrуге антибиотике оtežava leчeњe, a учесталост стафилококног kликоштa kod здравих ljudи, koji могу бити izvor zaraze, kao i udeo MRSA među njima bili су cilj ovog rada.

Ispitano je 5776 briseva grla i nosa poreklom od zdravih odraslih ljudi u mikro-biološkoj laboratoriji Завода за javno здрављe (ZZZJ) Ćuprija. Izolati су идентификовани testom koagulaze u epruveti i pomoću manitol slanog agara sa tabletom novobiocina. Detekcija meticilin rezistentног Staphylococcus aureusa (MRSA) vršena ја disk-difuzionom metodom sa tabletom cefoksitina (30 mcg).

Prevalенca назалне MRSA колонизације iznosila je 3,63%, a 4,29% ispitиваниh bili су nosиoci S. aureusa.


Ključне reči: стафилокок, назално kликоштво, MRSA