

## Prevalence Methicillin-Resistant *Staphylococcus aureus* (MRSA) Nasal Carriage among Healthy Students of Medical Schools in Istanbul (Turkey)

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### Abstract

Nasal carriage of MRSA plays a key role in the epidemiology and pathogenesis of staphylococcal infections. The aim of this study is to examine nasal MRSA carriage among healthy pre-clinical and clinical medical students at School of Medicine in Istanbul (Turkey). We also evaluate the antimicrobial resistance patterns of MRSA strains. In this study, involving 118 medical students, both pre-clinical (n:78) and clinical (n:40) at Bahçeşehir University, School of Medicine was conducted from October 2016 to June 2017. The volunteer students were screened for nasal carriage of *S. aureus* by streaking both anterior nares with sterile moistened cotton swabs. Then all nasal swabs and clinical specimens inoculated onto Chromagar MRSA and blood agar plate media and incubated at 35-37°C for 48 hours. The isolates were identified as *S. aureus* by standard conventional procedures. The antimicrobial susceptibility of the isolates was determined using by the Kirby-Bauer disk diffusion method, according to the EUCAST panels. MRSA carriage among pre-clinical and clinical students were 7.6 % (6/78) and 7.5% (3/40) respectively. There was no statistical significant different between the two groups. Also all MRSA strains isolated from pre-clinical medical students were susceptible to chloramphenicol and tigecycline, whereas numbers of susceptible strain to tetracycline, ciprofloxacin and erythromycin were 5, 4 and 3, respectively. All isolates from clinical medical students also found susceptible to ciprofloxacin, tigecycline and chloramphenicol, only two strains were susceptible to both tetracycline and erythromycin. In conclusion, under the light of these preliminary findings of the study, focused MRSA surveillance to medical students like other health care workers as a potential sources for that pathogen must be in a consideration. There is no statistical significance between both groups, in term of nasal carriage of MRSA and antimicrobial susceptibility patterns. Therefore, this trial would be continued with the same student groups till to their internship period which they will be fully practice in the hospital.

**Keywords:** medical students, MRSA, nasal carriage, antimicrobial susceptibility

### Резюме

Назалното носителство на *Staphylococcus aureus* сред здравето население е глобално явление. Освен това, назалното носителство на MRSA играе ключова роля в епидемиологията и патогенезата на стафилококовите инфекции. Целта на настоящото изследване е да се проучи назалното носителство на MRSA при здрави студенти по пре-клинична и клинична медицина във Факултета по медицина в Истанбул (Турция). Тъй като тези бактерии са мултирезистентни, оценихме също антиминокробната резистентност на щамове MRSA. Изследването, включващо 118 студенти по медицина - пре-клинична (78) и клинична (40), във Факултета по медицина при Bahçeşehir University, е проведено от октомври 2016 до юни 2017 г. Проведен е скрининг със студенти-доброволци за назално носителство на *S. aureus* чрез натривки от ноздрите и стерилни влажни памучни тампони. След това назалните проби и клинични проби се инокулират върху петрита с хромагар за MRSA и кръвен агар и се инкубират при 35-37°C за 48 часа. Изолатите се идентифицират като *S. aureus* чрез стандартни

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процедури - биохимични тестове, морфология след оцветяване по Грам и позитивни тестове за каталаза, коагулаза и ДНКаза. Антимикробната чувствителност на изолатите се определя по диск-дифузионния метод на Кърби-Бауер, с използване на панели антимикробни агенти, съответстващи на препоръките на EUCAST. Носителството на MRSA при студенти от предклинична и клинична медицина, съответно, 7.6 % (6/78) и 7.5% (3/40). Не се установи статистически значима разлика между двете групи. Всички щамове MRSA, изолирани от студентите по предклинична медицина, се оказаха чувствителни към хлорамфеникол и тигециклин, а щамовете, чувствителни към тетрациклин, ципрофлоксацин и еритромицин чувствителни съответно 5, 4 и 3. Всички изолати от студентите по клинична медицина проявяват чувствителност към ципрофлоксацин, тигециклин и хлорамфеникол, докато само два щамове са чувствителни едновременно към тетрациклин и еритромицин. В заключение, в светлината на тези предварителни данни, трябва да се обърне внимание на надзора на MRSA при студентите по медицина и при други работещи в здравеопазването, които са потенциален източник на този патоген. За изясняване на проблема, обаче, е необходимо дългосрочно проследяване на тези студенти по медицина. Няма статистически достоверни разлики между двете групи по отношение на назалното носителство на MRSA и на антибиотиковата чувствителност. Затова ще продължим изследванията с тези групи студенти по време на техните стажове, когато ще работят в болницата на пълно работно време за по-продължителни периоди.

## Introduction

*Staphylococcus aureus* is one of the most common bacterial pathogens and causes various diseases, ranging from localized mild infections to invasive life-threatening diseases (Aung *et al.*, 2017). *S. aureus* is a major resident or transient colonizer of the skin and the mucous membranes, especially the nasal-pharyngeal region and gastrointestinal tract of humans and animals. Transmission of *S. aureus* occurs mainly through person-to-person contact (Kluytmans *et al.*, 1997). Although colonization of multiple body sites occurs, the anterior nares are the most frequent carriage site. *S. aureus* are nasally colonized in a range between 30–50% of healthy persons. *S. aureus* nasal colonization can be an indicator of high risk for subsequent infection (Scarnato *et al.*, 2003; Miller *et al.*, 2012; Bijnen *et al.*, 2015).

MRSA was first identified in England in 1961 after which it emerged worldwide (Jevons, 1961). Since the 1990s, MRSA colonization and infections have been increasingly reported among the general population/community and are referred to as community-acquired MRSA (CA-MRSA) strains (Deresinski, 2005). Therefore, so many trials were carried on community-acquired MRSA (CA-MRSA) and hospital-acquired MRSA (HA-MRSA) isolates about their antimicrobial susceptibilities and ability to toxin production (Munckhof *et al.*, 2009; Bijnen *et al.*, 2015; Aung *et al.*, 2017).

Nasal carriage may be a strong risk factor for some serious infection. Few studies have examined MRSA carriage among medical students. The aim of this study is to examine nasal MRSA carriage among healthy pre-clinical and clinical medical stu-

dents at School of Medicine in Istanbul (Turkey). Since these bacteria have a multiple resistance pattern, thus we also evaluate the antimicrobial resistance patterns of MRSA strains.

## Material and Methods

In this study, involving 118 medical students, both pre-clinical (n:78) and clinical (n:40) at Bahçeşehir University, School of Medicine was conducted from October 2016 to June 2017.

The volunteer students were screened for nasal carriage of *S. aureus* by streaking both anterior nares with sterile moistened cotton swabs to a depth of approximately 1 cm, and rotated 5 times (Scarnato *et al.*, 2003). Nasal swabs were placed in tubes containing sterile normal saline and quickly transported to the medical microbiology laboratory at the Medical School of Bahçeşehir University in Istanbul. Then all nasal swabs were inoculated onto Chromagar MRSA and blood agar plate media and incubated at 35-37°C for 48 hours. The isolates were identified as *S. aureus* by standard procedures, such as gram staining morphology, catalase test, coagulase test, and DNase test positivity (Martineau *et al.*, 1998).

The antimicrobial susceptibility of the isolates was determined using the Kirby-Bauer disk diffusion method, according to the EUCAST panels for the following antimicrobial agents: Chloramphenicol 30 µg, gentamicin 10 µg, erythromycin 15 µg, tetracycline 30 µg, ciprofloxacin 5 µg, tigecycline 15 µg, co-trimoxazole 1.25/23.75 µg, linezolid 10 µg. All intermediate resistance phenotypes were considered as resistant; Resistance to vancomycin

**Table 1.** Antibiotic susceptibility patterns of MRSA strains

Antibiotics	Pre-clinical students		Clinical students	
	S*	R**	S*	R**
Chloramphenicol	6	0	3	0
Gentamicin	4	2	2	1
Erythromycin	3	3	2	1
Tetracycline	5	1	2	1
Ciprofloxacin	4	2	3	0
Tigecycline	6	0	3	0
Linezolid	6	0	3	0
Vancomycin	6	0	3	0
Co-trimaxazole	6	0	3	0

S\*: susceptible, R\*\*: resistant

was also evaluated by E-test Vancomycin strips (Biomérieux, FR), as recommended by the manufacturer. *S. aureus* ATCC 25923 (American Type Culture Collection, Manassas, VA) has been used as a quality control strain for susceptibility testing.

## Results

A total of 118 healthy students were enrolled into the study. Of pre-clinical students were 34 (43.6 %) males and 44 (56.4 %) females. Of clinical students were 16 (%) males and 24 (%) females. The median age (interquartile range) of the pre-clinical and clinical participants were 21 (20–22) years and 22 (21–23) respectively. MRSA carriage among pre-clinical and clinical students were 7.6 % (6/78) and 7.5% (3/40) respectively. There was no statistical significant different between the two groups.

All MRSA strains isolated from both pre-clinical and clinical medical students, were found susceptible to vancomycin, co-trimaxazole and linezolid. Also all MRSA strains isolated from pre-clinical medical students were susceptible to chloramphenicol and tigecycline, whereas numbers of susceptible strain to tetracycline, ciprofloxacin and erythromycin were 5, 4 and 3, respectively. All Isolates from clinical medical students also found susceptible to ciprofloxacin, tigecycline and chloramphenicol, only two strains were susceptible to both tetracycline and erythromycin (Table 1).

## Discussion

*S. aureus* nasal carriage is a global phenomenon among healthy population. Also, nasal carriage of MRSA plays a key role in the epidemiology and

pathogenesis of staphylococcal infections (Yano *et al.*, 2009). *S. aureus* nasal colonization is common in communities worldwide, and the *S. aureus* nasal carriage rate varies by specialized populations (Choi *et al.*, 2006; Chatterjee *et al.*, 2009; Munckhof *et al.*, 2009; Ma *et al.*, 2011). Previous studies revealed low prevalence of nasal MRSA colonization (1.4-9.4%) in Chinese medical students from different regions (Chen *et al.*, 2017). In addition, the analogous report that the detection rate of *S. aureus* at Wenzhou Medical College in Wenzhou was 15.4%, of which 3.0% were MRSA (Du *et al.*, 2011). Besides, the study from Brasil reported that in a medical student community, the detection rate of *S. aureus* and MRSA were 20.6 and 3.4% (Gushiken *et al.*, 2016). However, in a Malaysian report, any nasal MRSA colonization was not found among 209 medical student both pre-clinical and clinical groups (Nordin *et al.*, 2012) Previous reports from Turkey, in medical students nasal MRSA carriage rates were varied in a range of 2.9-9% (Dağı *et al.*, 2015). In this study, MRSA carriage among pre-clinical and clinical students were 7.6 % (6/78) and 7.5% (3/40) respectively. There was no statistical significant difference between two groups.

Increasing antimicrobial resistance has become a global public health problem in recent decades, threatening effective treatment of bacterial infections (Bijnen *et al.*, 2015). Increasing antimicrobial resistance data in the previous century was mainly focussed on hospital settings, the last decade has seen an increase in interest in community-associated resistance (Zetola *et al.*, 2005; Mediavilla *et al.*, 2012; Miller *et al.*, 2012; Bijnen *et al.*, 2015).

Classically, the occurrence of resistant *S. aureus* (including methicillin resistant *S. aureus*, MRSA) was confined to hospitals and long-term-care facilities. In the 21st century however, MRSA infections have also appeared in community-dwelling individuals with no healthcare associated risks such as a recent hospitalization. Most bacterial infections are caused by the patients' own commensal microbiota, which forms a reservoir of bacterial antibiotic resistance genes (Bijnen *et al.*, 2015). Ineffective and inappropriate antibiotic treatment has negative consequences for both the patient and the healthcare system; thus, the infection remains untreated.

The results of this study reveals that, all nasal MRSA isolates from both pre clinical and clinical students were found susceptible to chloramphenicol, tigecycline, co-trimazole, linezolid and vancomycin. Also erythromycin resistance rates were the same in both population as 50%.

The multidrug resistance (defined as resistance to 3 or more antibiotic classes) phenotype is a particular characteristic of MRSA, related to the global presence and spread of multidrug-resistant clones (Sakoulas *et al.*, 2008; Drougka *et al.*, 2014; Stefanaki *et al.*, 2017). Apart from the characteristic resistance to all  $\beta$ -lactams, MRSA also demonstrates resistance to many classes of antibiotics such as macrolide, clindamycin, fluoroquinolones, tetracycline, mupirocin, and co-trimazole (Tenover *et al.*, 2009; McDougal *et al.*, 2010; Stefanaki *et al.*, 2017). Multidrug resistance rates in MRSA strains from European countries, such as Sweden, the Netherlands, Austria, Belgium, Croatia, Spain and France was reported varied between 1.2 to 11% (Bijnen *et al.*, 2015).

In terms of multidrug resistance MRSA besides beta-lactams resistance, we detected only one nasal isolate from pre-clinical student resistance to tetracycline, gentamicin, and ciprofloxacin ; and two nasal isolates from clinical student resistance to erythromycin, clindamycin, and tetracycline.

## Conclusion

Under the light of these preliminary findings of the study focused on MRSA surveillance to medical students, like other health care workers as a potential sources for that pathogen must be in a consideration. However, for clarification of that issue needs a long term survey of these medical students. There is no statistical significance between both groups, in term of nasal carriage of MRSA and antimicrobial susceptibility patterns. Therefore, this trial would be continued with the same student

groups till to their internship period which they will be fully practice in the hospital for longer periods.

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