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Evaluation of nasal carriage of *Staphylococcus aureus* in students of a Dentistry Faculty

Diş Hekimliği Fakültesi öğrencilerinde *Staphylococcus aureus*'un burun taşıyıcılığının değerlendirilmesi

ABSTRACT

Objective: In this study, the carriage of methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-susceptible *Staphylococcus aureus* (MSSA)(S. *aureus*) was investigated in preclinical and clinical student groups at the IYY University Faculty of Dentistry.

Methods: The present study was an in vivo study carried out between December 03 and 21, 2018 at the Dentistry Faculty of İstanbul Yeni Yüzyıl University. A total of 76 preclinical and 76 clinical students provided nasal swab samples for the study. Samples were obtained from the first one-third of the anterior nasal region of both nostrils and examined microbiologically. The study findings were statistically evaluated using a chi-square continuity test with (Yates) correction (P < .05).

Results: S. aureus was detected in 16 of 76 (21.1%) clinical students who treated patients, and 3 of the 76 (3.9%) preclinical students who did not treat patients. No MRSA colonization was found in any group. There was a statistically significant difference in the prevalence of S. aureus between the clinical students who had contact with patients and the preclinical students who did not provide care (P = .003, P < .05).

Conclusion: In this study, the frequency of *S. aureus* findings in the group of clinical students who cared for patients was statistically significant (P = .003, P < .05). This result indicates that infection control protocols should be followed more closely and that, in particular, the clinical students who care for patients should pay more attention to the established rules of hygiene.

Keywords: Dentistry, methicillin-resistant Staphylococcus aureus.

öz

Amaç: Bu çalışmada, Yeni Yüzyıl Üniversitesi Diş Hekimliği Fakültesi' nde, hasta bakmayan 76 preklinik öğrencisi ile, hasta bakan 76 klinik öğrencisinin ön burun bölgesinden alınan sürüntü örneklerinde, metisiline dirençli *Staphylococcus aureus* (MRSA) ve *Staphylococcus aureus* (*S. aureus*) taşıyıcılığı araştırıldı.

Yöntemler: Çalışmamız, burun sürüntü örneği alınmasını kabul eden, hasta bakmayan 76 preklinik öğrencisi ile hasta bakan 76 klinik öğrencisi arasında yapıldı. Araştırmaya katılan öğrencilerin her iki burun deliğinin 1/3 ön burun bölgesinden sürüntü örnekleri alınarak mikrobiyolojik olarak incelendi. Çalışmada elde edilen bulgular, istatistiksel olarak Continuity (Yates) Düzeltmeli Ki-Kare Testi kullanılarak değerlendirildi (*P* < ,05).

Bulgular: Çalışmamızda hasta bakmayan 76 preklinik öğrencisinin 3'ünde S. aureus saptanırken (%3,9), hasta bakan 76 klinik öğrencisinin 16' sında *S. aureus* saptandı (%21,1). MRSA kolonizasyonuna ise hiçbir grupta rastlanmadı. Hasta bakmayan preklinik öğrencileri ile hasta bakan klinik öğrencileri arasında S. aureus görülme oranları açısından istatistiksel olarak anlamlı farklılık saptandı (*P* = ,003; *P* < ,05).

Sonuç: Yaptığımız araştırmada, klinikte hasta bakan öğrenciler grubunda S. aureus oranı istatistiksel olarak anlamlı derecede yüksek bulundu (P = ,003; P < ,05). Bu sonuç, enfeksiyon kontrol protokollerine daha sıkı uyulması ve klinikte hasta bakan öğrencilerin, hijyen kurallarına daha fazla dikkat etmeleri gerektiğini göstermektedir.

Anahtar Kelimeler: Diş hekimliği, methicilline dirençli Staphylococcus aureus

INTRODUCTION

Since there has been a rise in the number of outbreaks of infectious disease in recent years, the behavior and attitudes of health professionals working in the treatment environment have taken on even greater importance. It is crucial that all healthcare workers who may come into contact with blood, blood products, or bodily fluids, and/or who may be exposed to aerosols in their work environment observe certain rules for infection control.¹² As a result of the progressive spread of microorganisms demonstrating resistance to multiple antibiotics, serious problems have been encountered in the treatment of bacterial infections due to microorganisms, which has increased the need for new antibiotics. *Staphylococcus aureus* (*S. aureus*) is one of the most important infectious agents that cause community and hospital-acquired infections throughout the world. Methicillin-resistant *S. aureus* (*MRSA*) infections, particularly in intensive care units, have been reported at increasing rates.³

It has been shown that bacterial aerosols formed during procedures performed by dentists usually contain *streptococci* and *staphylococci* species. However, other infective bacteria have also been detected.¹ Studies have also indicated that the intensity of bacterial aerosols is higher in clinics operating part-time rather than full-time. Natural or air-conditioned ventilation systems have been demonstrated to reduce the concentration of bacterial aerosols in the environment.^{1,3}

It has been reported that during dental treatment, the water systems used in dental units were the source of *Mycobacterium tuberculosis (M. tuberculosis), S. aureus,* and other gram-negative bacteria, and that these bacteria can be easily transferred to oral wounds during treatment procedures.^{1.3,4}

Investigations have revealed that microorganisms can be transferred from one patient to another as a result of working with poorly sterilized instruments or contamination of dynamic hand tools.^{1,2} In a case reported in the literature, MRSA was isolated from the surgical area in the mouth and from the dentist's hand.³

Patients who have positive culture results for MRSA but who do not show the symptoms of diseases caused by this pathogen are considered to have a MRSA colonization.³

If culture-positive bacteria are detected in samples obtained from one or more body regions, such as the nose, then these individuals are assessed as MRSA carriers. Carrier individuals are considered a risk group in the development and spread of infections.⁵

In MRSA carrier individuals, infection can also be spread through contaminated hands and inanimate materials.^{5,6} Among health workers, those with nasal *S. aureus* colonization are an endogenous source of clinical infections. They may also be a source of cross-colonization in the community through the transmission of these bacteria.^{2,6}

Treatment of infections caused by S. aureus has become problematic due to the development of antibiotic-resistant S. aureus (MRSA). $^{2.7}$

Staphylococcal infections can cause a variety of infections, including sepsis, soft tissue or urinary tract infections, and pneumonia. Due to resistance to many antibiotics, such as penicillins, cephalosporins, clindamycin, erythromycin, tetracycline, and aminoglycosides, the treatment options for infections are very limited and the results can be fatal.³

Therefore, the screening of health workers is extremely important for the detection of *S. aureus* and MRSA carriage. The aim of this study was to determine the rate of nasal *S. aureus* and MRSA carriage in clinical students in the dentistry faculty who provided dental care for patients, as well as preclinical students who did not treat patients.

MATERIAL AND METHODS

The present study was an *in vivo* study carried out between December O3 and 21, 2018 at the Dentistry Faculty of İstanbul Yeni Yüzyıl University. Approval from the ethics committee of clinical research of İstanbul Yeni Yüzyıl University was obtained (Date: November 22, 2018, No: 22.11.2018/035). After receiving informed consent forms from all of the students volunteering to participate in the study, the group completed a questionnaire that recorded details of the student's name, age, chronic illness status (diabetes mellitus, heart diseases, asthma, chronic obstructive pulmonary disease) and antibiotic usage.

Students who were selected to participate did not have any systemic disease and had not used antibiotics within the previous 3 days. Then two groups created (preclinical students and clinical students).

The study included 76 clinical students who attended to patients and 76 preclinical students who did not provide care. Nasal swab samples were taken from the first one-third of both nostrils of all of the participants using cotton swabs prepared with sterile saline.

Nasal swab samples were taken and transferred to the laboratory within 30 minutes in the transport medium and incubated for 18 to 24 hours on the appropriate medium. At the end of the incubation period plates were evaluated for beta-hemolysis and the colonies with beta-hemolysis analyzed with different procedures to find out whether the proliferating colonies are *S. aureus* or not. Numbers recorded were then statistically analyzed.

Microbiological Analysis

The nasal swab specimens were incubated for 18 to 24 hours at 37°C on 5% sheep blood agar (Becton Dickinson and Co., Franklin Lakes, NJ, USA) medium within the 30 minutes of sampling. The plates were evaluated following the incubation period (Figure 1). A catalase test was applied to proliferating colonies by inducing beta hemolysis. Colonies with positive test results were confirmed for the presence of *S. aureus* with a rapid diagnostic test, the BBL Staphyloslide Latex Test Kit (Becton Dickinson and Co., Franklin Lakes, NJ, USA) and a coagulase test performed in the tube.

Methicillin susceptibility of the colonies with positive results was investigated and evaluated using the Kirby-Bauer disc diffusion susceptibility method in accordance with the Clinical and Laboratory Standards Institute recommendations.⁸ The inoculum was prepared from the proliferating bacteria, and after adjusting the turbidity to 0.5 McFarland, Mueller-Hinton agar (Becton Dickinson and Co., Franklin Lakes, NJ, USA) plates were inoculated and a cefoxitin disc-30 μ g (Becton Dickinson and Co., Franklin Lakes, NJ, USA) was placed on the culture medium. After 18 to 24 hours of incubation, zone diameter was measured, and the methicillin resistance or susceptibility was determined (Figure 2). A standard strain of *S. aureus* ATCC 29213 was used as a control in all of the tests.

Statistical Analysis

When evaluating the findings obtained in this study, IBM Statistical Package for the Social Sciences for Windows, version 22.0 (IBM SPSS Corp., Armonk, NY, USA) program was used. A chi-square test with Yates (continuity) correction was employed to evaluate qualitative data. Statistical significance was evaluated at P < .05.

RESULTS

S. aureus was detected in 3 of the 76 (3.9%) preclinical students who did not treat patients, and in 16 of 76 (21.1%) of the clinical students who attended to patients. MRSA colonization was not found in any group. There was a statistically significant difference in the prevalence of S. aureus (P = .003, P < .05) (Figure 3).



Figure 1. S. aureus colonies

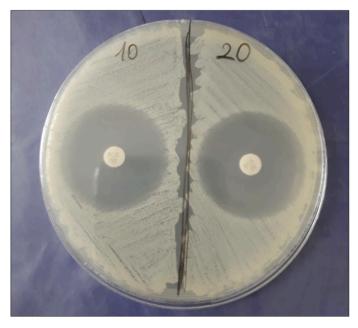


Figure 2. Inhibition zone in a cefoxitin disc diffusion test on Mueller-Hinton agar

The prevalence of *S. aureus* was significantly greater in the clinical students who provided direct dental care for patients (21.1%) compared with the preclinical students who did not look after patients (3.9%) (Table 1).

DISCUSSION

Due to the MRSA epidemic and limited antibacterial treatment options, the importance of MRSA infection among other hospital-acquired infections is increasing. MRSA colonization was found in 45% of the patients hospitalized in intensive care units of European hospitals, and 21% of them had hospital-acquired MRSA.^{2,9,10} *S. aureus* can become resistant to multiple antibiotics, and thus becomes an endemic nosocomial agent, which has led clinicians to search for new solutions, which can be both challenging and expensive.²

It has been reported that investigations performed in US hospitals revealed that methicillin-resistance in *S. aureus* colonies increased from 2.4% in 1975 to 29% in 1991, with a higher resistance level in intensive care units. Between 1990 and 1997, hospital-acquired MRSA incidence rates increased by 260%.²¹¹

Many sources of MRSA infection may be present in the practice of dentistry, including an infected or colonized physician, a dentist's seat, air/water sprays, power buttons of the dental unit, and the light fixture.² The use of personal protective clothing and gloves reduces the likelihood of contact with microorganisms.

Strong aspiration and using antiseptic mouthwashes before a procedure are highly effective in preventing the spread of microorganisms. The antiseptic gargles used should have a long duration of action. Chlorhexidine gargles have been found to be more effective in preventing bacterial aerosols than others. The use of a rubber dam is also a very effective method of preventing contamination originating from dental procedures.¹¹²

Since an infectious agent can be transported through bio aerosols in dentistry clinics, prevention is extremely important. Den-

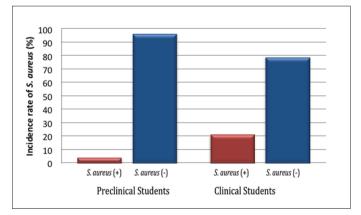


Figure 3. Detection rate of S.aureus among preclinical and clinical students

Table 1. Evaluation of the detection rate of *S.aureus* among preclinical and clinical students.

n (%)	n (%)	γ^2	n
(/~)	n (70)	χ	P
3 (3.9)	16 (21.1)	8.662	.003*
73 (96.1)	60 (78.9)		
		73 (96.1) 60 (78.9)	73 (96.1) 60 (78.9)

 ζ^2 : Chi -square continuity test with Yates correction, *P <

tal plaques, tartars, saliva, blood, used materials, and air/water sprays are known to be potential sources of bacteria and infectious aerosols released during dental procedures.

Microorganisms in an aerosol often adhere to each other or to inanimate surfaces. Aerosols can be inhaled into the lungs or the mucous membranes. Their size is bigger in jumps that are taking place during the process. They can also easily adhere to the surface of the skin, the hair, and clothing.^{12,13}

Various measures can be taken to reduce the number of bacterial aerosols in working areas of dentistry, including hygiene practices. The number of aerosols can also be reduced with the use of air filter systems or ultraviolet radiation.^{1,14}

S. *aureus* can colonize in many places, including the throat, nose, groin, armpits, and perineum. The anterior nasal cavity is the most frequently seen site of MRSA colonization. For this reason, we elected to take swab samples from the frontal area of the nose.

As the number of isolated nasal MRSA strains increases, the likelihood of colonization in other body regions also increases. MRSA strains can survive up to 12 days on abiotic surfaces.^{2,15} Goud et al.¹⁶ examined the prevalence of MRSA in the area surrounding Bengaluru, India, among 1000 people of different age groups and socioeconomic levels. The lowest nasal carriage rate was found to be 9.9% in the group of those aged 20-40 years, and the highest rates was seen among physicians (22%).

In Ethiopia, Shibabow et al.¹⁷ reported on the prevalence of *S. aureus*, and indicated a rate of nasal carriage of 28.8% among 118 health workers, and 44.1% were nasal MRSA carriers. In China, Chen et al.¹⁸ reported that the prevalence of *S. aureus* among 292 people working in the Chinese health sector was 21.6%, and that 4.7% of these cases were nasal MRSA carriers.

In 2017, Hema et al.² reported a prevalence rate of MRSA among 200 graduate students of dentistry and 200 undergraduate students of 24.5% (n=49) and 12.5% (n=25), respectively.

In 2009, Zimmerli et al.¹⁹ evaluated MRSA carriage by taking samples with nose and throat swabs from 500 dentists. *S. aureus* carriage was determined in 210 physicians; however, only 2 indicated methicillin resistance in the nose and throat. In the evaluation of the outcome, it was reported that the incidence of MRSA carriage was low among dentists.

In our study, *S. aureus* was detected in 16 (21.1%) of the 76 clinical students who were treat patients, and in 3 of the 76 (3.9%) preclinical students who did not provide direct dental care. MRSA was not identified in any group. The results showed that even though our hospital has a standard infection control policy and the importance of this topic is constantly emphasized, raising awareness concerning hygiene needs to be further increased and that students should comply with the hand-cleaning and other hygiene rules.

Nasal carriage of MRSA varies between countries and similar investigations. Possible reasons for these differences include variations in infection control policies, methodological differences, differences in the number of samples, and geographical variations.^{18,19} The absence of MRSA in both of our study groups may be due to the number of samples, so we could not compare our results with any research of similar design.

In 2017, Emaneini et al.³ found a high nasal MRSA carriage rate of between 22.7% and 32.8% in a meta-analysis conducted among healthcare workers in Iran between 2000 and 2016. They also linked this condition to the ineffectiveness of infection control policies. Inadequate hand cleaning is significant in the spread of nosocomial pathogens, such as MRSA and vancomycin-resistant enterococci. In Iran, due to a heavy workload, insufficient healthcare personnel, limited infrastructure, negligence of hand cleaning, and a frequent nonuse of masks, aprons, and gloves have been reported as important factors.

In developed countries, treatment with intranasal mupirocin has been proven to reduce the rate of nosocomial MRSA infections due to the fact that hygienic conditions are typically at a top level in hospitals.¹⁹ However, the overuse or irregular use of antibiotics is a common practice in developing countries. For example, cephalosporins and fluoroquinolones are used frequently, and this can lead to the emergence of MRSA in hospitals. In other words, excessive and poor use of antibiotics also plays an important role in the emergence and spread of MRSA.^{20,21}

Nasal MRSA and *S. aureus* carriers among healthcare workers may be the main sources of disease spread. Early and rapid identification of MRSA carriage in critically ill patients where there are MRSA carriers (e.g., intensive care units), screening of healthcare workers in contact with these patients, and other efforts to reduce the spread of MRSA in hospitals have been suggested.^{5,22}

There is a risk that MRSA will infect other patients with a variety of diseases, as well as other healthcare workers. Hospitals must have isolation rooms. Patients infected with MRSA should be kept in separate rooms. Infection control teams must be experienced and well trained. This training should include the basic rules of infection control, information on infections related to the work environment, the infection control policy, precautions to be taken when exposed to infection, and the rules governing sterilization and disinfection. All personnel should receive such training at the beginning of their employment.^{1,23-26}

In summary, poor hand hygiene, the irrational use of antibiotics, and ineffective infection control measures may explain the relatively high nasal carriage of *S. aureus* and MRSA among healthcare workers. Strategies recommended for this purpose include the avoidance of excessive antibiotic use and using utmost care when prescribing antibiotics, an emphasis on hand hygiene, screening for carriage and colonization, and proper observance of environmental cleanup, contact measures, and measures to prevent MRSA contamination.

The risk of the transmission of *S. aureus* and MRSA infection is not just a problem of medical schools, but also rather a general public health problem. It must be a concern for all dental health employees and patients. In the present study, a statistically significant prevalence of *S. aureus* was found among students who treated dental patients in clinics (p=.003, p<.05). This result shows that infection control protocols should be more closely followed and students treating patients in dental clinics should pay more attention to the rules of infection control.

Ethics Committee Approval: Ethics committee approval was received for this study from the ethics committee of İstanbul Yeni Yüzyıl University (Date: November 22, 2018, No: 22.11.2018/035).

Informed Consent: Written consent was obtained from students who participated in this study.

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