Research paper

Nasal conveyance of Methicillin-resistant *Staphylococcus aureus* (MRSA) strains among dental professionals with varying levels of clinical exposure: A comparative study

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ABSTRACT

Objective: Any control program for MRSA requires identifying the whole hospital reservoir. The likelihood of conveyance of MRSA may be higher in dental settings. Hence, the aim was to compare the nasal conveyance MRSA among dental professionals with varying levels of clinical exposure in a tertiary dental hospital in South India.

Methods: A total of 81 volunteers were stratified based on the number of years of clinical exposure. The nasal swabs were subjected to catalase and coagulase tests as well as antibiotic susceptibility test. Chi-Square test was done to compare the different types of organisms isolated from dental professionals with varied levels of clinical exposure.

Results: Out of the total 81 isolates, 62.96% of the isolates were MCONS (Methicillin sensitive coagulase negative staphylococcus), 20.98% of them were MRCONS (Methicillin resistant coagulase negative staphylococcus), 9.87% were MSSA (Methicillin sensitive staphylococcus aureus) and 3.7% were MRSA positive and 2.46% were other gram negative organisms.

Conclusions: MRCONS, MSSA and MRSA were shown to increase with an increase in the clinical exposure years.

1. Introduction

Staphylococcus aureus is a gram-positive coccus and a commensal organism, usually colonizing the skin and nose of healthy people. When turned parasitic, it can instigate an array of infections, ranging from simple, superficial skin infections to life-threatening pneumonia and bloodstream infections such as sepsis and toxic shock syndrome.¹ MRSA entails those *Staphylococcus aureus* organisms, which are resistant to all presently available β -lactam antimicrobial agents, including anti-staphylococcal penicillins (methicillin, oxacillin, nafcillin) and cephalosporins. 'Methicillin Resistant' implies multiple resistance.²

This microorganism is considerably unstable, has wide dissemination and can be transmitted from person to person, causing cross-contamination. Not only skin infections, *S. aureus* can also cause pneumonia, septicemia, osteomyelitis, abscess and other diseases. There is a strong relation between the presence of *S. aureus* and the occurrence of serious infections, such as infective endocarditis.³ MRSA is found in the anterior nares of about 6% of healthcare workers and nearly 3% of

hospital patients admitted. These hospital in-patients who become colonized or infected with MRSA can transpire to be the main reservoir for its spread in this setting.⁴

They are considered a public health problem, especially in large hospitals, since they make the treatment with common antibiotics difficult.^{4,5} Thus, the spread of MRSA strains in hospitals has now become a major issue, worldwide. Studies indicate that MRSA strains now account for 20%–40% of all *S. aureus* isolates in hospitals where these strains are widespread. Patient to patient transmission (generally by means of colonized hands of healthcare workers and also via their clothes or equipments) is considered as the foremost factor in spreading of MRSA in hospital settings.^{5,6}

In dental settings, the likelihood of transmission of MRSA may be higher than among other healthcare settings, because of the close proximity between dentists and patients, and that, the spray and splashes can suspend microorganisms in air.⁷ Recent studies also indicate that the personnel with persistent MRSA nasal colonization may possibly spread the organism via droplet transmission and more readily

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among those with respiratory infections.^{6,8}

Numerous studies have shown an increase in the number of microorganisms during dental clinical procedures, showing contamination from aerosols, especially from high-speed hand pieces and ultrasonic scalers.^{4,9–11} Various surfaces of the dental operator, including air-water syringe and dental chair were contaminated with MRSA, as found in a study conducted by Kurita.¹² Any control program for MRSA thus requires identifying the whole hospital reservoir of resistant strains.⁵ Hence, the aim of this study was to screen for and compare the nasal conveyance of antibiotic resistant pathogens among dental professionals with different years of clinical exposure.

Objectives of the study were: i. Nasal swab collection from anterior nares of dental professionals for isolation of *S. aureus*. ii. Perform Gram staining, Catalase test and Coagulase test for the identification of *S. aureus*. iii. Antibiotic susceptibility testing of these strains using the Kirby–Bauer disc diffusion method.

2. Materials and methods

The present study was conducted among dental professionals with varied levels of clinical exposure at a tertiary oral health care hospital in Southern India which caters to about 200 patients everyday and performs about 2000 major surgeries every year, after approval from Institutional Review board and informed consent from study participants. A total of 81 volunteers who were in good general health were enrolled in the study. Those study participants who had a recent history of hospitalization, those who had taken antibiotics within 30 days, and those undergoing immunosuppressive or chemotherapeutic treatments, were excluded. Study participants with varied levels of clinical exposure were stratified as follows:

- First year undergraduates (with no clinical exposure)
- House surgeons (with 3 years of clinical exposure)
- Post graduates (with 5 years of clinical exposure)
- Teaching faculty (with more than 5 years of clinical exposure)

Each participant were to complete a questionnaire that asked for gender, age, year of schooling, duration of clinical training, and a brief medical history. Nasal swabs were collected by placing a sterile cotton-wool swab in the anterior nares and rotated five times without interruption under even pressure and was withdrawn along the anterior wall of the nose. The swab was then placed in a tube containing 2 ml of Stuart transport medium and subjected to microbiological laboratory procedures.

3. Microbiological laboratory procedures

The nasal swabs that were collected, were cultured on Mc Conkeys agar and Brain Heart Infusion agar plates within 1 h after collection. The swabs were streaked on the agar plates as per the conventional technique. The culture plates were incubated at 37°C for 24–48 h in the incubator. The suspected white opaque and golden yellow colonies were selected and subjected to gram staining. Those isolates, which showed gram positive cocci in clusters were tested for the presence of catalase and coagulase.

Catalase test: With a sterile wooden stick, a little amount of 18 uncontaminated cultures from a general purpose media was transferred onto the surface of a clean glass microscope slide. Three drops of 3% hydrogen peroxide solution were included onto one portion of Gram-positive cocci microbial colony on the slide. Immediate bubbling of gas was indicative of a positive catalase test result for the presence of a *Staphylococcus species*. The non-appearance of bubbles immediately indicated a catalase negative test result (Gram-positive cocci species that are Catalase negative are non *Staphylococcus aureus*).

Coagulase test: After 24 h, characteristic colonies from surface of the medium were transferred to one of the two tubes containing 0.5 mL

of human plasma. Both the tubes were incubated at 37 °C and observed for a period of 4 h. Gelling of plasma, which remains in place even after inverting the tube, was indicative of a positive result.

Antibiotic susceptibility testing: was done using the Kirby–Bauer's disc diffusion method. Saline suspension of the strain was spread over the Mueller–Hinton Agar (MHA) plates after a turbidity matching of 0.5 McFarland standards. Antibiotic discs (HiMedia, Mumbai, India) were placed on these plates after 10 min, with six discs on each. After 24 and 48 h of incubation at 35 °C, the plates were read using the ATCC (American Type Culture and Collection) break-points as reference. The inhibition zones around the discs were measured, based on which, the strain was interpreted as sensitive or resistant.

3.1. Statistical analysis

Data was entered into excel spread sheet and exported to SPSS for analysis. Chi- Square test was done to compare the different types of organisms isolated from dental professionals with varied levels of clinical exposure.

4. Results

4.1. Demographic characteristics of study participants

Mean age and gender wise distribution of participating dental professionals is given in the Table 1. The mean age of dental professionals was 25.2 ± 1.94 and 43.2% were males and 56.8% were females.

We applied gram staining procedure to identify the gram positive cocci. Those isolates, which showed gram positive cocci in clusters were subjected to the catalase test. The catalase positive colonies were then subjected to the coagulase test to distinguish between 'Coagulase positive' and 'Coagulase negative' Staphylococci. It is observed from the Graph 1 that out of the total 81 isolates, 62.96% of the isolates were MSCONS, 20.98% of them were MRCONS, 9.87% were MSSA and 3.7% were MRSA and the remaining 2.46% were other gram negative organisms.

We divided the study participants into 4 groups, based on their clinical exposure. According to dental school's curriculum, students do not partake in clinical practice during the first two years of the program. Only third year onwards, they participate in clinical dental practice. Hence, first year students did not have any clinical experience and house surgeons had three years of clinical experience. Most of the isolated pathogens among first year graduate students and house surgeons were MSCONS. Highly significant differences were found between the groups with respect to isolation of this pathogen, with highest percentage of isolates among first year BDS students (33.3%) and decreasing with increasing years of clinical exposure. Whereas, none of the study participants in both these groups had MRSA strain in their anterior nares. MRSA was isolated from those participants with clinical exposure of 5 years or more. The percentage of other pathogens, that is, MRCONS and MSSA amplified with an increase in the years of clinical exposure and were as high as 35.3% and 37.5% respectively among the teaching faculty (Table 2).

Graph 2 shows antibiotic resistance of the isolated pathogens. It was observed that all the isolated strains were resistant to penicillin, erythromycin and ciprofloxacin. MRCONS, MSSA and MRSA were

Table 1
Mean age group and the gender distribution of the study population.

Characteristics		Number of subjects (n)	Percentage (%)
Total		81	100
Gender	Males	35	43.2
	Females	46	56.8
Mean age (years)		25.2 ± 1.94

Graph 1. Percentage of the pathogenic organisms isolated from the nasal swab culture.

Table 2

Distribution of study subjects based on the positive isolation of selected nasal microorganisms.

Years of clinical exposure	CONS positive n (%)	MRCONS positive n (%)	S.aureus positive n (%)	MRSA positive n (%)
First year BDS	17 (33.3)	2 (11.8)	1 (12.5)	0
House surgeons	15 (29.4)	4 (23.5)	2 (25)	0
Post graduates	12 (23.5)	5 (29.4)	2 (25)	2 (66.7)
Teaching Faculty	7 (13.7)	6 (35.3)	3 (37.5)	1 (33.3)
Chi square value	9.952	1.86	0.60	0.82
Interpretation	0.01 (HS)	0.60 (NS)	0.89 (NS)	0.84 (NS)

susceptible to tetracyclines. All the four strains of pathogens were susceptible to vancomycin.

5. Discussion

Several studies have provided data on higher conveyance rates of MSSA and MRSA among the health-care workers.^{11–13} There could be an increased risk of transmission even in dental set up due to the close proximity between the dentists and patients. Those patients, who may be the asymptomatic colonizers of this dreadful strain, can be a potential reservoir for the transmission of this strain to treating dentists and there may be further transmission of the organism to other patients via improper hand hygiene of dental personnel.

Even though, a nasal colonization of MSSA or MRSA itself does not imply that the carrier has the infectious disease, but, researchers have reported that they might experience self-contamination during low immunity periods and transmission.¹⁴ Therefore the present study was conducted to screen dental professionals with various levels of clinical exposure for nasal conveyance of resistant pathogens. Nasal swab cultures a sample of 81 dental professional volunteers were collected since the anterior nares are known to be the primary colonization site of *S. aureus*.

The highest conveyance of CONS was among the first year BDS students and least among the PGs and teaching faculty. In a study conducted by Aktar N,¹⁵ the majority of the isolates of the nasal swab culture of the health care workers were CONS (73.3%). CONS, occurring as a normal flora of skin and anterior nose are also pathogenic, when the host is compromised. CONS are the most common pathogens in nosocomial blood stream infections and indwelling catheters. Methicillin-resistant Coagulase Negative Staphylococcus (MRCONS) have also been found worldwide.^{15,16}

It is seen from the present study that the MRCONS and MRSA pathogens increased with an increase in the years of clinical exposure, although it did not show statistically significant differences (Table 2). It may be due to the fact that the dental professionals with higher exposure to patients acquired these pathogenic strains. The percentage of isolation among the dental professionals could have increased with the increased prevalence of these pathogens in the community.

All the isolated pathogens were resistant to Penicillins and Erythromycin and most of them were resistant to Ciprofloxacin and Gentamycin which was similar to the results of the study conducted by Shrestha B.¹⁷ It could be the result of inappropriate usage of these antibiotics. Moreover, the resistant bacteria can easily penetrate from the nasopharynx to the oral cavity and next with saliva droplets to the breathing zone air, creating a direct threat to a dentist. None of the isolated pathogens showed resistance to Vancomycin in the present study which was a drug of choice in the treatment of the MRSA infections although recently, strains fully resistant to vancomycin or VRSA (MIC, 64 mg/L) were isolated from 3 patients in the United States.¹⁸ The nasal conveyance was positive for MRSA among 3 of the 81 dental professionals (3.75%).

These dental professionals may thus become “cloud health-care workers” during upper respiratory tract infections with substantially increased dispersal into air. The chances of transmission can be more, if hand hygiene and other infection control procedures are not practiced. Grundmann et al.¹⁹ detected a 15% transmission rate from MRSA-

Graph 2. Antibiotic Resistance pattern of the isolated pathogens.

positive patients to the hands of health care worker after removal of gloves, which is comparable to a rate of 17% from MRSA-colonized patients to the gloves of health-care workers reported by McBryde et al.²⁰ But dental professionals as persistent reservoirs of MRSA in dental institutions are rare, yet important, most frequently acting as vectors and not as the main sources of MRSA transmission.²¹ Since MRSA conveyance is of short duration, we may have missed transient carriers of MRSA in the present study which could have been another drawback of the present study. Hence further studies are required with periodic screening of dental health professionals and collecting swabs from other representative sites as well like the throat and hand to get a better picture of the conveyance rates among the dental professionals.

The pathogenic strains isolated from the dental professionals in our institution were mainly of the following four types: Coagulase negative *Staphylococcus*, Methicillin resistant coagulase negative pathogens, *Staphylococcus aureus* and Methicillin resistant *Staphylococcus aureus*. MRSA isolates were shown to increase with an increase in the years of clinical exposure. Most of the isolated pathogens showed resistance to Penicillin, erythromycin, gentamycin and ciprofloxacin. Few of the isolated *Staphylococcus aureus* and coagulase negative organisms were resistant to methicillin.

6. Conclusions

Screening of the dental health professionals for the conveyance of pathogenic strains should be considered in the routine infection control policy. A periodic unannounced screening of the dental professionals can give an insight into their hand hygiene practice and improve awareness of possible consequences and issues related to transmission. A strict hand hygiene protocol should be followed in every dental institution and clinical set up to reduce the risk of transmission of any pathogens and thus making our dental practice a safe one.

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Declaration of competing interest

All authors report no conflicts of interest relevant to this article.

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