

Evaluation of bacterial aerosol contamination during dental procedures

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Abstract

Introduction: Bio-aerosols are airborne particles that contain bacteria, viruses and fungi, or originate from living organisms, which possess potential for transmitting infections in healthcare settings including dental clinics due to their ability to lodge in the smaller air passages of the lungs. Many dental procedures that use mechanical instrumentation have the potential to create contaminated aerosols and splatter. These aerosols represent an infection hazard to the dental team and other patients due to their gross contamination with microorganisms and body fluids.

Objective: The purpose of the present study was to evaluate the bacterial aerosol contamination during dental procedures in the dental operatory.

Materials and Methods: 60 patients with mild to moderate gingivitis attending outpatient department of Periodontics were included after obtaining consent. Settle plate method employing 10% sheep blood agar plates was used to collect the aerosols at three standardized locations. The sampled blood agar plates were transported immediately to the Department of Microbiology for:

1. Identification of bacterial colonies as per standard procedures.
2. Counting the number of colonies formed on blood agar plates using colony counter unit.

Results: Alpha hemolytic *Streptococci* (81.67%) were the predominant bacteria isolated followed by *Micrococcus* (38.89%). *Staphylococcus aureus* was isolated in 30% among which 10 isolates (18.5%) were methicillin resistant. Plates placed on the patient's chest area showed highest mean colony count of 105.3 CFU/plate, plates on dental tray showed 68.38 CFU/plate and plates placed 6 inches away from patient's mouth showed 40.12 CFU/plate.

Conclusion: To minimise the risk posed by dental aerosols it is imperative to implement control measures aiming to reduce the generation and exposure to microbial aerosols in dental clinics.

Keywords: Bacterial aerosols, Dental procedures, Dental clinic, Oral healthcare professionals, Colony forming unit.

Introduction

Bio-aerosols are airborne particles containing living organisms such as bacteria, viruses, fungi and their metabolites. The particles in a bio-aerosol range from 0.3 to 100 µm in diameter, in which respirable size fraction of 1 to 10 µm is of primary concern due to their ability to penetrate and lodge in the smaller air passages of the lungs. Thus they possess great potential for transmitting infections.¹ The presence of airborne bacteria in hospital environment is of increasing concern due to their potential role in causing hospital-acquired infections (HAI). The possible routes of human exposure to airborne microbes include inhalation, ingestion and dermal contact, with inhalation being the predominant route.^{1,2}

Oral health care professionals have increased chances of exposure to a wide range of microorganisms present in saliva and other body fluids during dental treatment.³ Dental clinics where various oral procedures are done can generate bio-aerosols and splatters. Common dental procedures which can generate aerosols containing microorganisms include sonic & ultrasonic scalers, turbine hand pieces and air syringes.⁴ In addition to patient's saliva, nasal and throat secretions, dental plaque, blood, tooth tissues and materials used in the dental treatment may all serve as sources of aerosols.⁵ The aerosols thus generated are particles small enough to remain airborne for extended periods of time before they settle on environmental surfaces.⁶ Further the

risk of exposure to microorganisms is high due to the open and invasive nature of the procedures done in oral cavity.⁷

The situation is further complicated due to the ecologically distinct nature of the oral cavity. The oral microflora comprises of both aerobic and anaerobic bacteria, including gram positive bacteria like *Streptococci*, *Lactobacillus*, *Actinomyces*, *Bifidobacterium* and gram negative bacteria like *Fusobacterium*, *Porphyromonas*, *gingivalis*, *Prevotella intermedia*, *Aggregatibacter*. Highly papillated surface of tongue and specialised host cell types of palate act as a reservoir for bacteria. Non-shedding surface of teeth also supports large masses of distinct micro flora. This unique nature of the oral cavity serves as an ideal medium for bacterial growth.⁸ The air in the dental space can thus be contaminated with such bacteria. Therefore the microbiological quality of air contained in the dental clinic space is important as it is inhaled by both the dentist and the patient, serving as a potential threat to the health of both.

Hence the present study was conducted in order to evaluate the bacterial aerosol contamination of dental clinic space during dental procedures.

Materials and methods

A hospital-based prospective study was conducted at Sri Siddhartha Dental College and Sri Siddhartha Medical College and Hospital, Tumkur over 4 months duration. 60 patients attending outpatient department of Periodontology, Sri Siddhartha Dental College were included in the study.

Patients were in the age group ranging between 21 to 55 years including both the genders. Ethical clearance was obtained prior to the study and written informed consent was obtained from the participating patients.

Inclusion Criteria: Systemically healthy patients with mild to moderate gingivitis.

Exclusion Criteria:

1. Patients with history of antibiotic therapy in the last 6 months.
2. Patients with history of using any chemotherapeutic mouth rinses and oral irrigation during the past 6 months.
3. Patients with any history of systemic diseases like renal or hepatic disease, blood dyscrasias and immunosuppression.

Sample collection: A simple method of gravitation or settle plate method was used for sample collection. Petri-plate containing agar medium was exposed face upwards to the atmosphere to collect particles that settle by gravity. Blood agar plate (90 mm diameter) was employed in this study to collect the aerosol sample during procedures as it is a general purpose, enriched and non-selective medium which can promote the growth of microorganisms sampled from air. An aerobic bacterium carried by inert particles deposits on to the surface of the agar and grows as a colony, it is counted as Colony forming unit (CFU)

Three 10% sheep blood agar plates were placed at three standardized locations –

1. Chest of the patient
2. On the tray of dental chair
3. 6 inches away from patient's mouth.

Preparation of the Operatory: All the procedures were done in a closed operatory. Each day at the end of all the procedures the room was fumigated, kept closed and was left unused for duration of 15 hours. In order to know the baseline atmospheric microbial contamination levels, a petri dish (90 mm diameter) containing 10% sheep blood agar was kept exposed in the middle of the room for 30 minutes before the start of each day's work. Staff avoided activities that can generate aerosols like conversation, coughing or sneezing. The plates were sent to microbiology laboratory and were incubated aerobically at 37°C for 48 hours.

Microbiological evaluation: All the blood agar plates were immediately transported to microbiology laboratory. After incubating aerobically at 37°C for 48 hours, those blood agar plates showing growth were studied further for-

1. Identification of the predominant and representative bacterial colonies by standard procedures.
2. To perform antimicrobial susceptibility testing - The Kirby Bauer's disc diffusion method laid by CLSI guidelines was used to test the antimicrobial susceptibilities of organisms isolated.
3. The isolates were tested against amoxicillin, gentamicin, ceftazidime, ceftazidime/clavulanic acid, cefoxitin, piperacillin/tazobactam, imipenem and cotrimoxazole.

4. Colony counter unit was used to count the number of colonies formed on blood agar plates and were expressed as colony-forming units per plate (CFU/plate).

Statistical analysis: Data was analysed using statistical software SPSS version 16. The cut-off level for statistical significance was taken at 0.05. Paired t-test was performed to compare mean CFU/plate at different locations.

Results

60 patients in the age group 21-55 years were included in the study, out of which 30 were males and 30 were females with mild to moderate gingivitis who were otherwise systemically healthy. Poor oral hygiene was commonly noted in both the genders. 18 males (60%) and 23 females (76.6%) had the habit of betel nut chewing. 22 males (73.33%) gave history of smoking.

Bacterial growth was observed in all 180 blood agar plates. Maximum number of colonies was seen on the plates positioned on patient's chest area. This was followed by the plate on the tray of dental chair. Few colonies were formed at plates placed 6 inches away from the operating area. All the plates showed mixed growth of bacteria predominantly gram positive cocci and aerobic spore forming bacilli.

Alpha hemolytic *Streptococci* (81.67%) were the predominant bacteria isolated followed by *Micrococcus* (38.89%). *Streptococci* were found to be sensitive to all the antibiotics. Six isolates (9.5%) of *Staphylococcus epidermidis* were found to be methicillin resistant. *Staphylococcus aureus* was isolated in 30%, among which 10 isolates (18.5%) were methicillin resistant.

Plates also showed the growth of gram negative bacilli. *Escherichia coli* were observed in 13.33% plates. Among 24 isolates of *Escherichia coli*, 5(20.8%) were found to extended spectrum beta-lactamase producers. *Pseudomonas aeruginosa* was observed in 6.67% plates and was found to be sensitive to imipenem (100%) and piperacillin/tazobactam (100%).

Plates placed on the patient's chest area showed mean colony count of 105.3 CFU/ plate, plates on dental tray showed 68.38 CFU/plate and plates placed 6 inches away from patient's mouth showed 40.12 CFU/plate as depicted in the table below.

Table 1: Percentage of bacterial isolates

Bacterium	No of isolates %
<i>Alpha hemolytic Streptococcus</i>	81.67%
<i>Micrococcus</i>	38.89%
<i>Staphylococcus epidermidis</i>	35%
<i>Staphylococcus aureus</i>	30%
<i>Aerobic spore forming bacillus</i>	20%
<i>Escherichia coli</i>	13.33%
<i>Pseudomonas aeruginosa</i>	6.67%

Table 2: Mean colony forming units/agar plate (CFU/plate) according to the location of the blood agar plate

Variable	Baseline (before treatment)	During treatment		
		Chest of the patient	On the tray of dental chair	6 inches away from patient’s mouth
Mean CFU/plate	6.37	105.3	68.38	40.12
Standard deviation	1.551	14.56	7.26	6.56

Graph: Showing comparison of mean CFU/ plate values at different locations

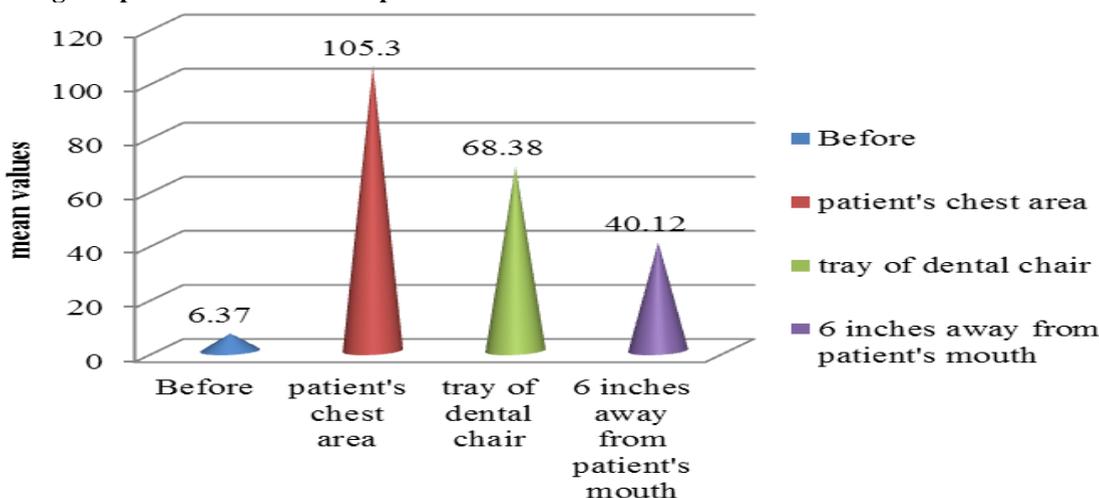


Table 3: Paired samples test values (sig – significant)

Pair	T-value	P-value
Patient’s Chest area & before	51.9	0.000(sig)
Tray of dental chair & before	63.23	0.000(sig)
6 inches away from patient’s mouth & before	37.51	0.000(sig)
Patient’s chest area and tray of dental chair	19.75	0.000(sig)
Patient’s chest area and 6 inches away from patient’s mouth	37.27	0.000(sig)
Tray of dental chair and 6 inches away from patient’s mouth	40.89	0.000(sig)

Discussion

Dental procedures making use of high-speed instruments have the potential to aerosolize saliva and thus are capable of producing viable aerosols. These aerosols can remain suspended in air for long time periods before being inhaled by dental staff and other patients.^{9,10} Evaluation of the quality of air can be performed by microbiological sampling and particle counting. Official standards for air control are based primarily on the measurement of CFU/m.³ Even though active air samplers provide the number of CFU per cubic metre (CFU/m³) of air, they have their own disadvantages like being expensive, noisy, fallout of microorganisms is not evaluated and different samples give different results.¹¹ Passive air sampling using settle plates are cheaper alternatives which measure the microbial fallout rather than air-suspended microbes.^{11,12}

In the present study, settle plate method using blood agar plates was employed. It was observed that maximum number of colonies was seen on the plates positioned on patient’s chest area, followed by the plates placed on the tray of dental chair. Plates placed at 6 inches away from patient’s mouth showed least CFU/plate. This is similar to studies conducted by Acharya S et al.⁹ and Rao RM et al.¹⁰

Plates placed on the patient’s chest area showed mean colony count of 105.3 CFU/ plate, plates on dental tray showed 68.38 CFU/plate followed by plates placed 6 inches away from patient’s mouth with 40.12 CFU/plate. This correlates with studies conducted by Acharya S et al.⁹ and Shivakumar KM et al.¹³ This could be attributed to the nature of larger salivary droplets which settle down rapidly on patient’s chest area. Therefore it can be noted that both the dentist and the patient get exposed to high amounts of bacteria in the form of aerosols.

It was observed that the microbial contamination generated during dental treatment was significantly higher than what was seen at the beginning of the day before the treatment started (baseline 6.37 CFU/ plate). This is found to be in agreement with the results reported by various studies conducted by Shivakumar KM et al.¹³, Jimson S et al.¹⁴ and Barlean L et al.¹⁵

In the present study gram positive cocci were isolated predominantly. Alpha hemolytic *Streptococci* were isolated in 81.67% plates. A study conducted by Monteiro PM et al.⁶ also showed predominance of gram positive cocci. They reported that *Micrococcus* sp. (99.9%), *Streptococcus* sp. (99.9%), *Staphylococcus epidermidis* (84.8%) and

Staphylococcus sp. (99.9%) were isolated. Study conducted by Jimson S et al.¹⁴ reported that *Streptococci* were isolated in 100% followed by Coagulase negative Staphylococci and *Staphylococcus aureus*.

Staphylococcus species which were considered as transient colonizers of the oral cavity are now gaining importance due to the emerging antimicrobial resistance. In our study 18.5% of *Staphylococcus aureus* and 9.5% of *Staphylococcus epidermidis* were found to be methicillin resistant. A study conducted by Ramalingam AJ et al.¹⁶ reported 12.5% of *Staphylococcus aureus* from oral cavity lesions were methicillin resistant. However no methicillin resistant CONS were isolated in their study. There is an increase in the number of patients with oro-nasal colonisation of methicillin resistant Staphylococcus aureus (MRSA). The importance of oral carriage of MRSA lies in the fact that it can serve as a reservoir for cross-infection to the dental staff and to other patients.¹⁷ Thus patients undergoing dental procedures may get colonized with antibiotic resistant bacteria if hygiene practices are insufficient and will continue to be an increasing problem if adequate precautions to control the spread of these organisms are not in place.

In the present study *Escherichia coli* and *Pseudomonas aeruginosa* were isolated in 13.33% and 6.67% samples respectively. A study conducted by Prashanth et al.¹⁸ observed that the predominant organisms identified were Pseudomonas, Proteus, gram positive cocci and aerobic spore forming bacilli. In healthy individuals oral colonization by Enterobacteriaceae or Pseudomonas spp. is usually transient.⁷ Therefore presence of Pseudomonas aeruginosa has generally been attributed to the contaminated Dental unit water lines (DUWL). This is because the water pipelines provide a favourable moist environment for microbial proliferation and biofilm formation leading to contamination with high densities of gram negative microorganisms like *Pseudomonas aeruginosa* and *Legionella species*.^{7,19} Hence the microflora from the DUWL and the patient's oral cavity together generated during various dental procedures in the form of aerosols combines with the surrounding air leading to a change in the original composition of the environment. Eventually this becomes a source of infection for both the dentist as well as the patients.

These aerosols can also contaminate the nearby instruments on the instrument trays as demonstrated by the bacterial growth observed on the plates placed on the dental tray in the present study. This can further act as a source of infection to the patients.

The present study demonstrates the nature of aerosols generated during dental procedures and the extent to which they may spread as all three sites sampled in the room showed significant contamination. This indicates that dental procedures carry a potential risk of hospital infection for the oral healthcare professionals. But the present study mainly focuses on the aerobic bacterial isolates that are capable of growth on blood agar. Fungi, Mycobacteria and strict anaerobes that require special media or growth conditions

are therefore not counted. Thus it is a partial picture of the airborne contamination that actually occurs during dental procedures. In spite of this limitation, the study provides an insight into the amount of airborne material that can be generated during various dental procedures.

There is a growing concern regarding the potential role of airborne bacteria as a source of hospital-acquired infections (HAI). 10–20% of endemic nosocomial infections can be attributed to airborne bacteria.² In hospital settings exposure to aerosols contaminated with microorganisms capable of disease transmission can lead to various diseases like mild flu, pneumonia, streptococcal and staphylococcal infections, viral infection, conjunctivitis, tuberculosis and severe acute respiratory syndrome (SARS).²⁰

Conclusion

Therefore it is imperative to implement control measures to reduce exposure to microbial aerosols in dental clinics. But due to the inherent nature of the procedures performed, it is difficult to completely eliminate the risk posed by dental aerosols. However it is possible to minimize the risk by following relatively simple precautions like personal barrier protection and pre-procedural mouth rinses. Dental health care personnel should use personal protective measures like face shields, surgical masks, and gowns. Periodic disinfection of dental equipment is highly recommended. Ventilation and air-conditioning system in good working order can reduce aerosols. Procedure rooms should be periodically disinfected by fumigation. Pre-procedural use of mouth rinses have also been shown to be effective in reducing the aerosol contamination. Further bio-aerosol monitoring is recommended in order to track and control hospital associated infections and as well as for the purpose of surveillance for infection control.

Conflicts of Interest: None.

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