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Original Article

Bacterial contamination of air and surfaces during dental procedures—An experimental pilot study using *Staphylococcus aureus*

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Abstract

Objective: The oral cavity contains numerous microorganisms, including antimicrobial-resistant bacteria. These microorganisms can be transmitted via respiratory particles from patients to healthcare providers and vice versa during dental care. We evaluated the spread of *Staphylococcus aureus* during standardized dental procedures using different scaling devices and rinsing solutions.

Methods: During systematic therapy for dental biofilm removal (guided biofilm therapy), using an airflow or ultrasound device to a model simulation head. *Staphylococcus aureus* suspension was injected into the mouth of the model to mimic saliva. Different suction devices (conventional saliva ejector or a prototype) and rising solutions (water or chlorhexidine) were used. To assess contamination with *S. aureus*, an air-sampling device was placed near the oral cavity and samples of surface areas were collected.

Results: *S. aureus* was only detected by air sampling when the conventional saliva ejector with airflow was used. No growth was observed during treatments with the ultrasonic piezo instrument or the prototype suction device. Notably, a rinsing solution of chlorhexidine digluconate decreased the bacterial load compared to water. Surface contamination was rarely detected (1 of 120 samples).

Conclusions: Although our findings indicate potential airborne bacterial transmission during routine prophylactic procedures, specific treatment options during biofilm removal appear to reduce air contamination. These options include ultrasonic piezo devices or the prototype suction device. The use of chlorhexidine reduced the CFU counts of *S. aureus* detected by air sampling. Surface contamination during dental procedures was a rare occurrence.

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During the coronavirus disease 2019 (COVID-19) pandemic, oral healthcare workers were considered high risk for infection due to their close proximity to patients and the use of aerosol-generating procedures, particularly periodontal treatment with ultrasonic scalers and airflow devices.[1] Respiratory particles, such as aerosols, droplets, or splatters, can transfer viruses but also other microorganisms like bacteria.[2] Given that the oral cavity contains >1,000 taxa, oral healthcare providers are continually exposed to a multitude of bacteria, some of which are antimicrobial-resistant pathogens. Importantly, *Staphylococcus aureus*, which can lead to several infectious disease syndromes, such as skin and soft-tissue infection or endocarditis, is frequently detected in the nose and throat and this can be the source of transmission and dissemination to other body sites.[3–10]

To mitigate the spread of microorganisms via respiratory particles, numerous infection control and prevention measures, including personal protective equipment and cleaning and disinfection protocols, have been established.[11,12] In addition to personal protective equipment, diminishing aerosol or droplet generation during dental procedures should decrease transmission risk.

We evaluated the dissemination of *S. aureus* during standardized dental procedures, and focused specifically on the influence of various scaling devices and rinsing solutions. Our findings provide valuable insights into potential infection prevention and control strategies. We hypothesized that different combinations of scaling and suction devices, as well as the choice of antiseptic solutions, would affect the dissemination of *S. aureus*. To test this hypothesis, we designed an experimental study simulating droplet and aerosol generation during dental biofilm management (ie, guided biofilm therapy or GBT). GBT is a conceptual stepwise cleaning approach

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that combines modern technologies to achieve efficient and comprehensive dental biofilm removal in routine clinical dentistry incorporating disclosing agents, airflow devices and ultrasonic scalers.[13] The combinations of cleaning and suction apparatuses and the selection of rinsing solutions varied throughout the research phase.

Methods

This research was conducted over nine experimental days at the Center for Dental Medicine, University of Zurich, Switzerland. Each day comprised three runs.

Setting

A dental model simulation head was placed on the patient chair in a designated consultation room (Fig. 1A). The room was equipped with air conditioning and a window. The biofilm-targeted therapy was performed according to Vouros et al[13] and was completed after ~10 minutes. Each day, 3 treatment courses were performed (ie, 3 runs). All designated sampling surfaces were cleaned with ethanol (80% v/v) between runs after samples were taken. Each day, a different combination of suction, handheld device, and rinsing solution was randomly applied (Table 1).

Dental treatment

The model simulation head, which simulated varying stages of periodontal disease, underwent GBT[13] through either an ultrasonic piezo instrument or an erythritol air-polishing device (AIRFLOW-One with AIRFLOW-1 PLUS powder, EMS, Switzerland). Two distinct suction devices, a conventional saliva ejector or a prototype provided by EMS Switzerland (Fig. 1C), were used alongside either a chlorhexidine digluconate 0.1% (CHX, BacterX, EMS, Switzerland) containing rinsing solution or water. Compared to the conventional saliva ejector, the prototype suction collects splatters in a more efficient way because of two soft flanges. The device is similar to the commercially available product GBT Flowcontrol (ref FV-112, EMS Elector Medical System SA, Nyon, Switzerland). The rinsing solution was used during the entire treatment period. Following each run, the oral cavity of the model was disinfected with ethanol (94% v/v). The window of the room was open during experiments. No treatment was performed for 20 min after each procedure and the window was left open.

Staphylococcus aureus suspension used to simulate contaminated saliva

An overnight liquid culture of *Staphylococcus aureus* (Cowan I) was diluted in phosphate-buffered saline (PBS) on the morning of each treatment day to create sampling stock. Prior to each run, the solution was diluted in PBS to achieve a concentration of 10^5 CFU/mL. Simultaneously, the resuspension was streaked onto agar to assess the bacterial count for each run. To mimic saliva, this suspension was steadily rinsed into the mouth of the model simulation head with a constant flow of 4 mL/min.

Air and surface sampling

An air sampling device [MAS-100 NT, MBV AG, Switzerland; flow rate, 100 L/min; Columbia agar + 5% sheep blood (COS) agar plates] was placed on a chair in 1 m distance to the dental model simulation head during treatments (Fig. 1 A and B). In addition, the following control experiments were performed on an additional day: (1) air sampler next to air conditioner; (2) air sampler 1 m from oral cavity but without treatment; and (3) air sampler 1 m from oral cavity with treatment (settings of experimental day 1, using PBS without *S. aureus*). COS plates were incubated for 24±2 hours at 36±2°C.

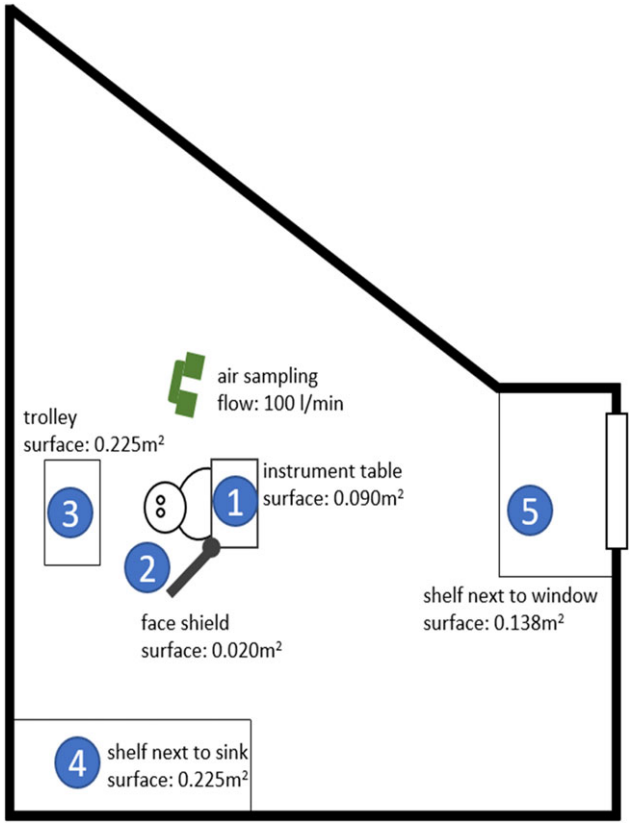

Figure 1. Schematic representation of the consultation room and sampling locations. Blue circles, surface sampling locations; green, air sampler/air sampling location.

Surface contamination was assessed by sampling 5 distinct room areas using gauze wipes (Mesoft 5×5 cm, Mölnlycke Health Care AB, Göteborg, Sweden). The wipes, stored in a sterile tube with 0.9% sodium chloride, were swiped over the defined areas in a zigzag pattern and were then returned to the NaCl solution. Wipes were shaken for 30 minutes at 250 bpm and were sonicated for 5 minutes at 44 Hz. Subsequently, the solution was plated on COS agar and was then incubated for 24±2 hours at 36±2°C.

Microbiology analysis

If growth was evident on the COS plates, species verification was carried out using MALDI Biotyper Sirius (Bruker Daltonics GmbH, Germany). If colonies were morphologically identical, only 1 colony was evaluated.

Statistical analysis

Medians and interquartile ranges (IQRs) for overall growth during the air sampling were calculated using R software (R Foundation for Statistical Computing, Vienna, Austria). The Wilcoxon rank-sum test was performed to compare CFU counts between the rinsing solutions (chlorhexidine and water).

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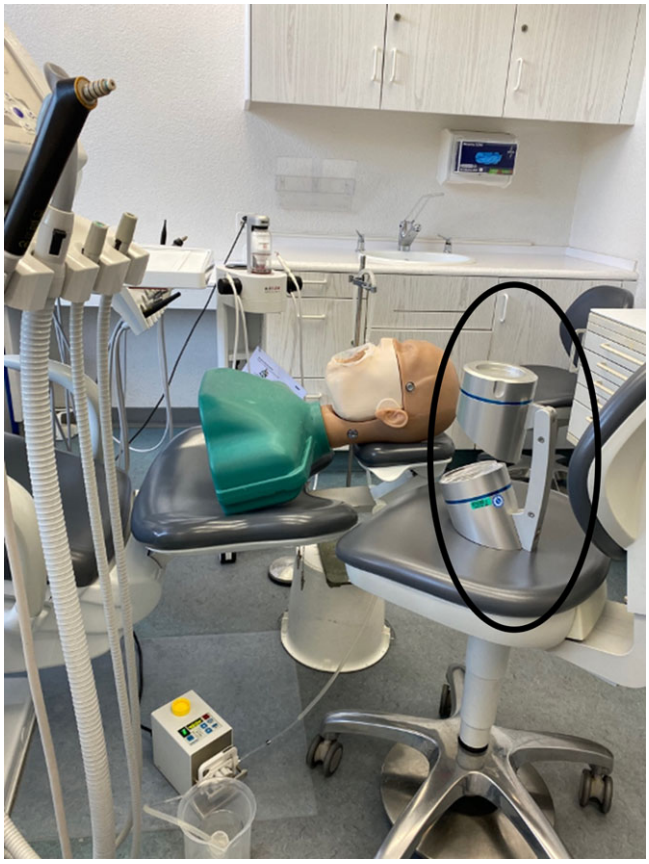

Figure 2. Model simulation head with air sampler (black circle).

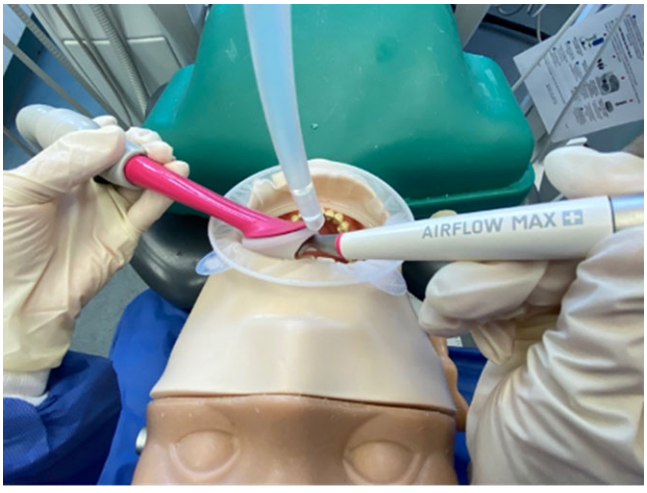

Figure 3. Prototype suction device (pink) and airflow device (white).

Results

Setting

The different treatment settings are shown in Table 1. The same settings were applied on experimental days 1 and 9. The mean inoculum ranged between 3.73×10^4 and 5.16×10^5 CFU/mL.

Air sampling

In total, 27 air samples were collected. Growth was detected in all of them. *S. aureus* was identified in 7 runs (25.9%) across 3 different experimental days (days 1, 2, and 9) (Table 1). On experimental days 1 and 9, all 3 runs tested positive (day 1, 13 CFU/3 runs; day 9, 63 CFU/3 runs). On experimental day 2, 1 run (1 CFU/3 runs) tested positive. On all 3 days, the saliva ejector was used in combination with the airflow.

In total, 31 CFU grew on average on each experimental day. On days using water, higher CFU counts were detected compared to CHX: median, 31 CFU (IQR, 14.00–58.50) versus 23 CFU (IQR, 9.75–31.00) ($P = .24$).

All control experiments showed no growth of *S. aureus*.

Surface samples

Except for day 1, 15 samples were collected each experimental day, for a total of 120 samples. Surface areas varied between 0.02 m^2 and 0.225 m^2 (Fig. 1A). *S. aureus* growth was detected in 1 sample (an investigator's face shield) on experimental day 4 (ie, saliva ejector + ultrasonic piezo instrument + CHX).

Discussion

Our findings demonstrate the presence of *S. aureus* in the air around the oral healthcare providers and the patient. Furthermore, we observed that certain treatment settings, such as the use of an ultrasonic piezo instrument or a prototype suction device, can potentially mitigate a transmission risk. Conversely, surface contamination was a rare occurrence and was primarily detected in the immediate vicinity of the oral cavity.

The issue of bacterial or viral spread through droplets or aerosols became a focal point during the coronavirus disease 2019 (COVID-19) pandemic.[1] Rautemaa et al[7] reported significant bacterial contamination at various sites during the operation of high-speed dental instruments. Consistent with other studies,[14] our air sampling revealed a diverse microbial population, but only a fraction (25.9% of the runs) confirmed the presence of *S. aureus*. This finding suggests that only this proportion of positive samples represented a true reflection of bacterial dissemination from the oral cavity during treatment. Prior research indicated that treatment tools and methods can affect oral microorganisms, such as decreasing bacterial counts in biofilm when ultrasonication is applied.[15] Interestingly, 1 combination of treatment settings (airflow and saliva ejector) accounted for all episodes of the air contamination. In addition to demonstrating the potential for bacterial transmission during dental procedures, these findings highlight the necessity for optimized device combinations.

To minimize bias by improving investigator's skills over time, we repeated the experiment with the same settings on the first and the last days. We detected *S. aureus* in all runs, indicating that these findings are truly due to the treatment settings. This observation supports that the combination of an ultrasonic piezo instrument or prototype suction device with airflow does not result in detectable aerial spread of *S. aureus*. Compared to water, the application of CHX, a compound included in a protection protocol for dental personnel,[16] resulted in a reduction of *S. aureus* in CFU counts when airflow and a saliva ejector were used. The reduced median overall growth for CHX and water (23 CFU vs 31 CFU) was not statistically significant ($P = .24$).

Surface contamination with *S. aureus* within the consultation room was rare. Although other studies have identified

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Table 1. Experimental Schedule and Results

Day	1	2	3	4	5	6	7	8	9
Suction Device	Saliva Ejector				Prototype Suction Device				Saliva Ejector
Biofilm removal device	Airflow		Ultrasonic		Airflow		Ultrasonic		Airflow
Rinsing solution	H_2O	CHX	H_2O	CHX	H_2O	CHX	H_2O	CHX	H_2O
Runs	3	3	3	3	3	3	3	3	3
Staphylococcus aureus mean inoculum (CFU/mL)	2.12×10^5	1.08×10^5	1.04×10^5	5.02×10^5	5.73×10^4	5.16×10^5	9.24×10^4	6.53×10^4	3.73×10^4
S. aureus detection									
Air sampling, total CFU	13	1	0	0	0	0	0	0	63
Run 1 (CFU)	6	0	0	0	0	0	0	0	4
Run 2 (CFU)	1	0	0	0	0	0	0	0	57
Run 3 (CFU)	6	1	0	0	0	0	0	0	2
Surfaces	N/A	No	No	Yes	No	No	No	No	No
Location	N/A	N/A	N/A	Face shield	N/A	N/A	N/A	N/A	N/A

Note. H_2O: water; CHX: chlorhexidine; N/A: not applicable; CFU: colony-forming unit; CFU/mL: CFU per milliliter.

contaminated surfaces in dental settings,[17–19] our investigation of potential patient-to-surroundings bacterial transmission did not mirror these findings; we observed only 1 positive sample of 120 samples collected. The low contamination rate was indeed surprising. Unlike many studies with agar plates at varying distances, we used gauze wipes (adapted from Oie et al[20]) to sample surface areas. Bahador et al[19] showed a positive correlation between treatment duration and the level of bacterial contamination, suggesting that our exposure time (~10 minutes) may have been too brief. Although this is a valid consideration, the total duration from the first run to the final surface sampling exceeded 1 hour. Furthermore, we cannot exclude residual effects of the disinfectant. Nevertheless, surface disinfection after every run is simulating real-life infection control measures. Previous studies have shown a decrease in contamination during scaling therapy as the distance from the oral cavity increases[21] and that the highest contamination levels are detected nearest to the oral cavity.[22] The fact that the face shield was contaminated in only 1 of the 24 runs was surprising but emphasizes the importance of personal protective equipment.

In addition to its clinical significance and its potential presence in the nose and throat, *S. aureus* has been detected in surface samples in other studies.[8–10,19] Therefore, we believe that *S. aureus* is a good indicator for spread of any pathogens during dental procedures. The chosen inoculum reflects CFU counts in saliva and dental plaques, and therefore also contributes to the real-life scenario of this experimental study.[23]

Our study had several limitations. We did not assess different bacterial concentrations, exposure times, or air sampling locations. All treatment procedures were performed by the same investigator. Although no evidence suggests that the negative results after day 2 were due to improved skills, this remains a possibility. It is uncertain whether the detected numbers of *S. aureus* colonies in the air and on the face shield actually would result in clinical transmission. Furthermore, we evaluated only the potential effect of contaminated saliva; we did not evaluate whether other colonized materials such as dental plaque could increase the bacterial load in the air or on surrounding surfaces. Future trials should incorporate this aspect.

In conclusion, our findings indicate that bacterial microorganisms can be transmitted into the air during scaling therapy if certain treatment settings (eg, using airflow and a saliva ejector) are employed. However, adjusting these settings to incorporate the use of an ultrasonic instrument or a prototype suction device resulted in no detection of *S. aureus* in the air. In our experimental setup, surface contamination was exceptionally rare (1 of 120 samples) and was only observed in the immediate vicinity of the oral cavity.

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Competing interests. P.W.S. received travel grants from Pfizer and Gilead, speaker's honorary from Pfizer, and fees for advisory board activity from Pfizer and Gilead outside the submitted work. P.R.S. received speaker's honorary and other (non-)financial research support from E.M.S. All other authors declare no conflicts of interest.

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