Title

Reduction of cultivable bacteria in aerosols generated by ultrasonic scaling by use of a chlorine dioxide mouthrinse as the lavage

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REDUCTION OF CULTIVABLE BACTERIA IN AEROSOLS GENERATED BY ULTRASONIC SCALING BY USE OF A CHLORINE DIOXIDE MOUTHRINSE AS THE LAVAGE.

by

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THESIS

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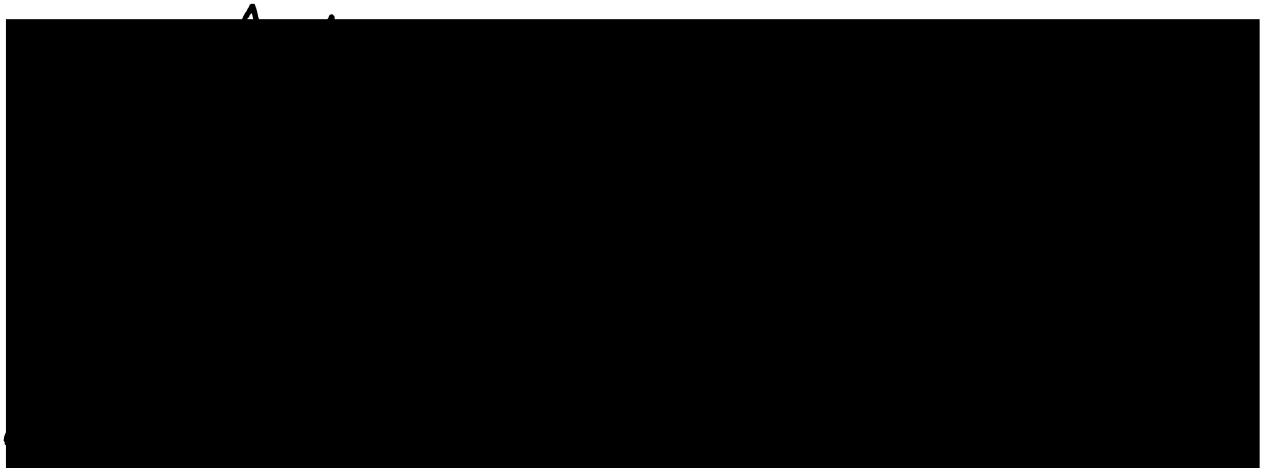

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ABSTRACT

Objective: The purpose of this double-blind, placebo-controlled study was to determine if use of a phosphate-buffered stabilized 0.1% chlorine dioxide mouthrinse when used as a lavage during ultrasonic scaling would reduce the number of colony-forming units of bacteria in the aerosols generated. Chemical mouthrinses and toothbrushing prior to dental procedures reduce the number of bacteria in the mouth and thereby reduce the bacterial load of the aerosols produced. While some mouthrinse used pre-procedurally may remain in the oral cavity during a subsequent dental procedure, it will soon become highly diluted by tap water sprays of ultrasonic scalers. Our hypothesis is that using a phosphate buffered-stabilized 0.1% chlorine dioxide mouthrinse itself as a lavage instead of tap water will incorporate more of the active agent into the aerosol and splatter particles where a greater and prolonged effect is anticipated. Aerosols of droplets of 0.5 to 5μm and its content may be inhaled and penetrate to the lung alveoli. Destruction of the microorganisms in the aerosol should reduce the risk of airborne infections.
Methods: 50 healthy subjects in the periodontal clinic were seated in a separate operatory for dental recall ultrasonic scaling procedures, and the air was sampled for cultivable microorganisms before, during, and after the procedure. The student dentist and patients were blinded to the use of ordinary tap water or phosphate-buffered stabilized chlorine dioxide mouthrinse as the ultrasonic lavage. Assignment to test (26) or control (24) group was by table of random numbers. Air samples were collected by a vacuum device onto trypticase soy agar (TSA) culture plates, incubated for 48 hours, and colony-forming units (CFU) counted at 11x magnification by two separate blinded examiners. The aerosols were assessed by means of counts of CFU on the TSA plates incubated after sampling 1,000L of air in a dental operatory before, during and 20 minutes after the ultrasonic scaling.
Results: The CFU means of pre-procedure samples were 32.5 (±20.2) and 32.9 (±38.2) and the groups did not differ. The mean count during ultrasonic scaling with water was increased five-fold to 166.0 (±196.6) CFU (P=0.001). The count during chlorine dioxide lavage was 67.1 (±98.8) CFU, about two-fold greater than pre-operatively. The test counts were a 60% reduction from that of controls (P=0.03). Post-scaling counts were close to pre-operative levels in both groups. No adverse reactions to the chlorine dioxide mouthrinse were observed in the test group.
Conclusions: The use of a stabilized chlorine dioxide mouthrinse as the lavage during ultrasonic scaling provides a statistically significant reduction of the cultivable bacteria in aerosols generated by the procedures.

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INTRODUCTION

In the past 20 years there have been documented, confirmed cases of transmission by an airborne route of tuberculosis,[1] the measles virus,[2] and most recently the SARS virus.[3] In an environment such as the dental office where aerosols, splatter, and fomites are produced from an oral cavity source laden with bacteria, viruses, subgingival fluid, and blood, this should be of considerable concern to the dental professionals and their surrounding staff. Aerosol and splatter contaminants are produced by numerous sources and procedures; high-speed handpieces, air-water syringes, air-polishing units, coughing patients, and sonic and ultrasonic scalers. It has been demonstrated that dental procedures incorporating the use of water sprays or rotary instruments generate aerosols with significantly greater numbers of bacteria than those produced by nondental oral activities, including coughing and sneezing.[4] Prospero et al. showed that a large number of microbes reach healthcare workers' faces during restorative and oral hygiene work, suggesting the same for any blood-borne pathogens present in the patient's mouth.[5] By monitoring the breathing zones of primary and assistant surgeons, it has also been shown that the mucous membranes lining the upper respiratory tract and alveolar macrophages in the gas-exchange region are likely to be exposed to aerosolized blood in an operating room.[6]

Fotos et al. showed that significant levels of *Legionella*-specific IgG and IgM antibodies were found in 20% and 16%, respectively, in 270 dental clinical personnel. This compares with 8% and 10%, respectively, for a randomly selected non-clinical group

($P<0.005$).[7] Comparing individuals' "years spent in the clinic environment" with the incidence of significant antibody levels strongly suggests that the risk of *Legionella* infection increases proportionately with increased clinic exposure time. Using petri dishes positioned around patients with pulmonary tuberculosis, Belting et al. showed by culture that tubercle bacilli are thrown into the air by a dental highspeed handpiece.[8] Using paper points as a means to collect gingival crevicular fluid samples, and the PCR technique, HIV-1 DNA has been isolated in the gingival crevicular fluid of AIDS patients.[9] Mononuclear cells present in gingival crevicular fluid and harboring proviral HIV-1 DNA could represent a potential source of HIV-1 in the presence or absence of local bleeding.

Splatter particles from the patient's mouth travel in a ballistic trajectory, landing on the face and clothing of the operator, assistant, and nearby surfaces.[10] The droplets of splatter are >50 μm diameter and may be visible. Splatter from the patient's mouth can be generated by breathing, speaking, sneezing, coughing, tooth brushing, gargling, high-speed drilling, polishing with a rubber cup or Robinson bristle brush, air-water syringe spray, and ultrasonic scaler.[10] In contrast, aerosol particles, nominally < 50 μm, may drift in the air for some time. They do not settle readily on open agar plates, so they are sampled by vacuum devices that draw them into liquid culture medium or onto agar plates. Aerosols generated by dentists in their work may contain particles and harmful chemicals or gases, as well as bacteria and viruses.[11]

Sources of aerosols in the dental environment include high-speed handpieces, air-water syringes, sonic and ultrasonic scalers, and air-polishing devices.

HIGH-SPEED HANDPIECES

Aerosols generated by high-speed handpieces have been sampled by an air sampler onto an agar plate and have been shown to contain tooth debris, microorganisms, water droplets, and saliva.[12] About 85% of the generated aerosol particles are <3.5μm, and 99% are <5μm.[13] Given that particles <5μm can penetrate to the alveoli of the lungs and initiate infection,[14] and that in a 15-min sample over 700 colony-forming units (CFU) <3.5μm can be detected,[12] the aerosols produced by high-speed handpieces should be a large concern to practitioners.

ULTRASONIC SCALERS

Saliva, blood, and water in the mouth can be aerosolized by the ultrasonic tip, contaminating large areas in an operatory.[15] Several studies show that the ultrasonic scaler produces more airborne contamination than any other instrument in dentistry.[7,16,17] In a laboratory trial, ultrasonic scalers spread splatters of fluorescein dye up to 25 inches away from a dental model.[18] It has also been found that ultrasonic scalers generate aerosols with bacteria peaking at over 300 CFU/cu. ft. of dental operatory volume.[10] Cultures from aerosol samples produced by ultrasonic scalers have yielded *Streptococcus viridans, Lactobacilli, Actinomyces, Staphylococcus albus, Pseudomonas aeruginosa,* and *Streptococcus pyogenes.*[19] Open culture plates 2 feet away from the patient's mouth during ultrasonic scaling were contaminated with alpha-streptococci.[20] Using a slit-type

air sampler for collection of aerosols, CFU counts on blood agar rose 3000% above baseline levels during ultrasonic scaling even after all patients brushed and used antiseptic mouthwash before tests, and were still 230% higher 35 min after scaling.[21]

Sonic scalers and air-polishing devices have also been shown to raise aerosols. Using an air sampler and blood agar plates, it was shown that sonic scalers generate aerosols comparable to those generated by ultrasonic devices.[22] Using blood agar plates positioned around an operatory, air-polishing devices were shown to produce significantly more CFU than control or rubber cup polishing even with the use of a high-volume evacuator (HVE).[23]

SAFEGUARDS AGAINST AEROSOLS

Currently there are several safeguard measures to protect dental practitioners, their staff, and the patients from the transmission of infection through the airborne route. Face masks, chemical mouthrinses, and high-volume evacuation are just a few. Though prudent and recommended, many of these safeguards may provide a false sense of security for the patient and operator.

Facemasks appear to be the first line of defense against aerosol infection but their effectiveness must be questioned. Insufficient filtering must be taken into consideration. Using an apparatus for testing aerosol filtration media for efficiency, a conventional cotton mask was found to between 43.01-93.6% efficient while a glass fiber mat combined with non-woven fabric mask was found to be between 98.1-99.4% efficient.[24]

Though masks have been designed to filter particles from 1-3 microns, many viruses, bacteria, and spores fall well below this range.[6] Fluid resistance also affects mask efficiency. Moist or wet aerosols combined with the clinician's breath saturate masks, allowing microbes and particles to wick through.[25] A face-mask should be changed every 20 minutes in a humid environment but this is rarely done.[26] Ill-fitting masks also diminish the value of the mask. A poorly fitting mask will end up being worn "out of position," such as under the nose.

High-volume evacuation is another line of defense that is used by practitioners to prevent the spread of infection through aerosols and splatter. Testing with an ultrasonic scaler, Harrel et al. reported a 93% reduction of aerosols with the use of a high-volume evacuator compared to a control.[27] Though significant, the study was performed on a Dentoform model and not in the mouth. Similar results were reported when an air-polishing device was used with and without the use of a high-volume evacuator.[10] Various devices that attach the high-volume evacuator directly to the ultrasonic scaler have been developed and have shown some promise. Nonetheless, it is extremely difficult for a clinician working without an assistant to utilize a high-volume evacuator while simultaneously using an ultrasonic scaler. Many operators continue to practice without the use of a high-volume evacuator. Dental hygienists almost never have assistants and thus are forced to practice without the use of a high-volume evacuator.

Chemical mouthrinses are often used prior to dental procedures in an attempt to reduce the number of bacteria in the patient's mouth and thus the bacteria in aerosols produced

by the procedure. Veksler et al. demonstrated that preoperative rinsing with 0.12% chlorhexidine gluconate diminished the quantity of aerobic and facultative flora of the saliva by culturing saliva samples taken before and after rinsing with 15ml of 0.12% chlorhexidine (CHX) or water.[28] Muir et al. demonstrated that preoperative rinsing with 0.2% chlorhexidine gluconate significantly reduced the number of organisms isolated on culture plates during ultrasonic scaling therapy.[29] It has also been reported that rinsing with an alcoholic mouthwash solution of essential oils significantly reduced the CFU produced by aerosols generated from ultrasonic scaling compared to a control alcohol rinse.[30] A 94% reduction in recoverable bacteria in the aerosol compared to the non-rinse control was reported. In one comparison of rinses before use of an air-polishing device, 0.12% chlorhexidine was significantly better at reducing splatter CFU than essential oils or water.[31] Larato[32] et al. and Knighton[33] reported that despite mouthrinses and tooth brushing before dental procedures, aerosols generated by high-speed handpieces were up to 2200% higher than preoperatively, and were still 250% higher 30 min later. Wyler et al. reported that preoperative brushing was no better than rinsing with water, and of four commercially available rinses tested, a quarternary ammonia compound achieved the best aerosol reduction (85%) during use of high-speed handpieces.[34]

Though evidence shows that pre-procedural chemical mouthrinses may reduce the number of bacteria in aerosols, it is obvious that a limited amount of the chemical agent will be incorporated into the aerosols given the limited supply of the chemical agent remaining the in mouth following a rinse. One would not expect that pre-procedure rinses would penetrate the dental biofilm (plaque) which the ultrasonic scaler would

aerosolize. Nor would rinses affect the gingival crevicular fluid or the blood droplets raised by the scaler. By incorporating the chemical rinse into the coolant or lavage, an abundant supply of the chemical agent could be present throughout the procedure. A laboratory experiment of aerosols of simulated saliva generated on an anatomic model by a high-speed handpiece tested the effect of antimicrobial solutions run through the handpiece spray as a coolant during the drilling. Of eight agents tried, those better than a water control for CFU reduction were 0.001% Merthiolate, Nitromersol, 0.5% Povidone-iodine, 5.0% sodium chloride, 5% lithium chloride, and 3% hydrogen peroxide.[35] Incorporating a chemical rinse into the lavage of an ultrasonic scaler should also provide an abundant supply of the chemical agent into the aerosol.

THE PROJECT

Given that no procedure has proven to be sufficiently effective in reducing microbial contaminants in aerosols, it makes sense to continue to study various ways to achieve this goal. In this study, we evaluated the use of a phosphate-buffered stabilized 0.1% chlorine dioxide (ClO_2) mouthrinse as a lavage during ultrasonic scaling and compared the CFU in aerosols generated to that generated with customary tap water lavage. ClO_2 has been used as a disinfectant in treatment of community water systems. It has more oxidizing power than chlorine[36] and rapidly kills common water and sewage pathogens,[37,38] and it kills spores,[39] yeasts,[40] and viruses.[12,41,42] It is generally tasteless and odorless. The null hypothesis to be tested was that the antimicobial lavage would not reduce the number of colony-forming units (CFU) of bacteria in aerosols to a greater extent than water lavage.

MATERIALS AND METHODS

Fifty adult patients were recruited from the patient population pool at the UCSF postgraduate periodontal clinic. All patients were in good general health. Patients had all been or were currently being treated for moderate to advanced periodontitis. All patients required scaling with root planing or a recall/maintenance procedure involving the use of an ultrasonic scaler. Patients were excluded if they required prophylactic antibiotics or had been taking antibiotics within the last 3 months, if they had used a medicated mouthrinse within the previous 4 hours, or were taking any other investigational drug. All subjects agreed to participation and signed a written consent form prior to involvement in the study. Subjects were not compensated in any way. The study was approved by the UCSF Committee on Human Research. Using a table of random numbers, the subjects were assigned to either the control or test group. Even numbers were assigned to the control group while the odd numbers were assigned to the test group. Twenty-six subjects were included in the test group and 24 in the control group. Ultrasonic scaling was performed by nine different periodontal residents throughout the course of the study.

The study was conducted in an enclosed operatory (11'x12'x8' or 1080 cu.ft.), which had central forced air circulation of 11,380 cu.ft. of air per hour as measured at the air supply diffuser, or 10 room air changes per hour, and positive pressure with respect to the main clinic. The clinician and assistant wore clean gowns with neck and waist tied and elastic arm cuffs, disposable face masks, disposable head caps with elastic bands, and eye protection. The patients were positioned in the reclining dental operatory chair with the

clinician and assistant at either side. Room temperature and humidity were recorded at the beginning of each sample. Plaque index[43] was recorded on each subject prior to treatment. 1,000L of air was then sampled using an airborne microbial monitoring unit (M Air T Air Tester, Millipore Corp., Bedford, Mass.) in the enclosed room over a time period of 7 minutes with the patient, assistant, and operator present but sitting quietly. The sampler was placed behind the operatory chair on a counter at a distance of 24" from the field of work (see figure 1.) in order to shield it from splatter. Following the commencement of the ultrasonic scaling of calculus and plaque deposits, a second sample was taken in the same manner as the first sample. The ultrasonic scaling was continuous during the entire sampling time. During the ultrasonic scaling, a saliva evacuator was utilized. A high-speed evacuator was not used. A third sample was taken 20 minutes following the completion of the procedure in the enclosed room with only the investigator present. Any complaint or mention of taste or odor from the chlorine dioxide was noted. Aerosol/related symptoms of cough, nasal irritation, running eyes, itchy skin, and headaches were also observed for.

The airborne microbial monitoring unit was a portable, battery-powered unit that pulled air samples through a sterile sieve consisting of 1,000 micro-perforations to minimize colony overlapping and reduce the potential for desiccation of the medium (see figure 2.). The particle diameter cutoff size was 3.5 μm. The sieve was changed between each air sample (3 sieves per subject) and sterilized between each use. The air sample particles were forced to impact a sterile plate of trypticase-soy agar (TSA) having ruled crosslines on the bottom for ease of counting cultures. The cassette surface rapidly rehydrated after

impaction thereby renewing the water activity and reducing further effects of drying on organisms present. Water loss after sampling was 4.2% and the agar volume was 34 mL. The device sampled air at a rate of 140L/min for the first 500L, and 180L/min for the second 500L averaging 1,000L/7 minutes.

The procedure performed on the patient consisted of using an ultrasonic scaler to remove calculus and plaque deposits. The ultrasonic scaling device was attached to a dispensing system that supplied the lavage for the ultrasonic scaler. An ultrasonic scaler (Cavitron Bobcat, Dentsply, York, PA) with a 25 kHz Swivel™ #10 Universal Ultrasonic Insert (Hu-Friedy, Chicago, IL) attached to a Dentsply Dual-Select Dispensing System reservoir was used (see figure 3.). Tap water (control) or a phosphate-buffered stabilized 0.1% chlorine dioxide mouthrinse (test, CloSYSII, Rowpar Pharmaceuticals, Scottsdale, Ariz.) was placed into the dispensing system before commencement of the scaling. The chlorine dioxide mouthrinse was not diluted. The operator, assistant, and patient were blind to the choice of either tap water or mouthrinse lavage.

Following sampling, the TSA culture plates were aseptically removed from the sampling device and covered. The samples were placed in an aerobic incubator at 37° for 48 hours. The colonies were counted twice independently by two separate investigators under 10.5 times magnification and reported as CFU/1000 L (see figure 4). To check for accuracy, counts were repeated without reference to first count. Colony counts of original and repeat counts of the two investigators were averaged for statistical analysis. Treatment differences were determined by analysis of mean and standard deviation of counts and

considered significant at a=0.05 or less. Levene's test for equality of variances was followed by paired *t* tests within treatment groups and unpaired *t* tests between groups. Spearman Rank correlations between the intraoperative CFUs and humidity, temperature and mean plaque index score were completed.

RESULTS

Using a table of random numbers, 24 subjects were assigned to the control group and 26 to the test group. No occurrences of discomfort or complications were caused by the use of the chlorine dioxide rinse as an ultrasonic lavage in any person. No subject complained of taste or odor from the chlorine dioxide.

During ultrasonic scaling, the mean temperature in the operatory was 23.3° C (range 22.1 to 27.5° C), and the mean humidity was 50% (range 34% to 67%). The mean total colony count before ultrasonic scaling was not significantly different between the control group (32.9±38.2 CFU) and the test group (32.5±20.2 CFU) (see table 1.). During ultrasonic scaling, the total colony count increased significantly (P=0.001) by 5-fold over baseline to 166.0±196.6 CFU in the control tap water group. In the chlorine dioxide test group, the total colony count increased to 67.1±98.8 CFU, a 2-fold increase that was not significantly greater than the baseline level (see figure 5.). Twenty minutes after the scaling was completed, the aerosol counts had returned to near preoperative baseline levels for the test and control groups. Those counts did not differ significantly from preoperative levels, nor from each other.

Spearman Rank correlations between the intraoperative total colony count and humidity were very low and not statistically significant. The mean plaque index score in the test group (0.54±0.29) was similar to that in the control group (0.48±0.27) and was not significantly associated with intraoperative CFUs when correlated using Spearman Rank.

The colonies were not identified. The majority were smooth-surfaced and dome-shaped, ranging from translucent gray to opaque whitish or yellow. Gram-stained smears showed primarily Gram-positive cocci in pairs, tetrads, chains, or clusters. Thirteen air samples exhibited mold colonies on the agar ranging from 1-3 per plate. There was no pattern to their appearance or relation to test or control, and they are considered incidental background contamination of the air in the dental clinic.

DISCUSSION

The use of chlorine dioxide as a lavage during ultrasonic scaling appears to be a safe and nonirritating method to reduce total counts of bacteria in aerosols. Over 40 persons (test group, operators, and investigators) were exposed with no adverse effects. Not a single subject mentioned nor noticed a difference in taste or odor with the chlorine dioxide. Aerosol-related symptoms of persistent cough, nasal irritation, running eyes, itchy skin, and headaches have been reported to be significantly greater ($P<0.025$) in dental hygienists than in a control group of clerical workers.[16] None of the volunteer student dentists nor patients reported any such problems in this study.

Variables amongst the subjects included room temperature and humidity, and subject plaque levels. These variables were recorded and correlated with intraoperative CFUs.

They were not associated with intraoperative CFUs and did not have a significant impact on the results. The same ultrasonic scaler, ultrasonic insert, lavage dispensing unit, operatory room, and aerosol sampler were used for every subject to reduce variability.

The study indicated a statistically significant 60% reduction (P=0.03) in CFU during the use of chlorine dioxide as a lavage. Although the standard deviations in CFUs was large, the 60% reduction in CFUs during use of chlorine dioxide lavage was statistically significant. The variance could not be accounted for by supragingival plaques scores, the temperature or humidity. In an environment where an assistant is rarely available to operate a high-volume evacuator during ultrasonic scaling for the average dental hygienist, a chlorine dioxide lavage can provide relevant protection. Face masks, pre-procedural rinses, high-volume evacuators, and high-efficiency particulate air room filters all have disadvantages and are not completely fail safe.[17] Pre-operative rinses have been reported to decrease counts of bacteria in saliva and thereby the counts in aerosols[44], but such rinses would not be expected to penetrate the biofilms of dental plaque nor have much effect on the subgingival microorganisms which would be dispersed by ultrasonic scaling. A comparison between chlorine dioxide, an essential oil, or a chlorhexidine gluconate as a mouthwash or as an ultrasonic lavage has not been done. High-volume evacuators are not commonly used in a single operator environment. Face masks are commonly worn improperly and not completely effective against aerosols. Because there is no complete safeguard available against contamination of aerosols, chlorine dioxide as a lavage can provide an easy method to help protect dental practitioners and their staff.

The colonies in the samples were not identified, and the TSA agar used would not be expected to grow out all bacteria present or to identify known pathogens. Also, this study did not provide any information concerning the effects of chlorine dioxide lavage on blood contamination of aerosols or on endotoxin reduction. The data does show a significant reduction in CFUs. Although most of the organisms were probably Gram-positive aerobic cocci, we assume that the reduction would apply across all species that might be present. The reduction of bacteria that could not be grown on the TSA agar cannot be confirmed with the data from this study.

Aerosols generated by high-speed handpieces and air-water syringes were not studied. Future studies may investigate the use of chlorine dioxide as a coolant in high-speed handpieces and in air-water syringes.

CONCLUSIONS

Aerosols generated during ultrasonic scaling create considerable amounts of microorganisms and generate droplets small enough to penetrate to the alveoli of the lungs, posing a risk of nosocomial infection for all persons in the dental clinic. Face masks, preprocedural rinses, high-volume evacuators, and air filters provide some protection but are far from fool-proof. The use of chlorine dioxide as the lavage during ultrasonic scaling safely and significantly reduced the exposure by 60% and thus represents an additional infection control method available to dentists and dental hygienists.

Table 1. Average colony forming unit measurements before, during, and 20 minutes after ultrasonic scaling.

Total Colony Counts/1000 Liters of Air (CFU)

TIME OF MEASUREMENT	CHLORINE DIOXIDE LAVAGE (n=26)	TAP WATER LAVAGE (n=24)
BEFORE SCALING	32.5 ± 20.2(Std. Dev.)	32.9 ± 38.2
DURING SCALING	67.1 ± 98.8*	166.0 ± 196.6†
20 MINUTES AFTER SCALING	34.7 ± 28.8	27.5 ± 30.1

*P= 0.03 vs. tap water lavage.
†P=0.001 vs. values before and after scaling.
The values before and after scaling were not significantly different between or within groups.

Table 2. Colony Forming Unit (CFU) counts for control group (tap water as lavage).

Sample #	Preoperative CFUs	Intraoperative CFUs	Postoperative CFUs	Temp °C	Humidity
1	10	88.75	9.25	22.4	67
2	188.5	723.25	147.25	22.6	67
5	33.5	110.5	28.5	22.7	59
6	100.25	606.25	44.25	23	37
7	20	245.5	51.75	23.4	56
8	38.75	51.25	4	25.1	34
10	12	105	5	23.2	44
13	18.25	12.75	45	23.1	59
15	41.75	33	32.5	22.7	35
18	34.75	8	8	22.5	48
21	16	5.5	17.25	23	50
22	24	136.5	7.5	23.6	46
23	19.75	111.75	8	22.9	50
24	18.25	218.5	21.5	23.6	44
28	9	30.25	11.75	23	46
29	15.5	35.25	16.25	27.5	35
31	30	387.75	32	22.7	54
32	11.25	442.75	14.75	23	54
36	12	19.25	33.5	22.1	50
37	19.75	105.25	16	22.1	60
42	43	336.75	62.25	24.7	50
43	24.5	34.25	25	22.7	50
46	38	36	11.5	22.7	55
48	10.25	100.25	6	22.1	58
Mean	**32.88**	**166.01**	**27.45**	**23.18**	**50.33**
Std. Dev.	**38.24**	**196.64**	**30.14**	**1.17**	**9.27**
Variance	**1462.21**	**38666.20**	**908.34**	**1.36**	**85.97**
Max	**188.50**	**723.25**	**147.25**	**27.50**	**67.00**
Min	**9.00**	**5.50**	**4.00**	**22.10**	**34.00**

Table 3. Colony Forming Unit (CFU) counts for test group (chlorine dioxide as lavage).

Sample #	Preoperative CFUs	Intraoperative CFUs	Postoperative CFUs	Temp °C	Humidity
3	85.5	62.5	49	22.7	63
4	30.75	346.25	153.5	23.5	64
9	28.25	325	38.25	23.5	54
11	9.5	27.5	37	24	51
12	77.25	68.25	37.25	22.8	50
14	79	149.5	22.75	22.8	39
16	10.5	17.75	11	22.6	42
17	42.5	22.25	14.5	22.4	44
19	27	6	37.75	21.8	41
20	43	27.75	12	22.4	45
25	29.25	284.5	39.75	22.4	50
26	17	27.25	27.25	22.6	41
27	37.25	30.5	35.25	22.5	52
30	16.5	64.25	36.25	22.9	47
33	26.5	10.5	23.5	22.2	53
34	30	30.25	62	22.7	56
35	30	17.25	37.25	21.8	54
38	18.5	109.25	18.25	22.3	60
39	28.75	26.25	71.5	22.1	62
40	16.25	14.25	10.75	22	60
41	18.75	3	9	22.8	65
44	44	15	30.75	23.1	44
45	13	15.25	8.25	22.3	60
47	28	9.5	32.5	22	59
49	37.25	17	20.5	23.2	52
50	19.75	17	26.25	22.3	50
Mean	**32.46**	**67.07**	**34.69**	**22.60**	**52.23**
Std. Dev.	**20.21**	**98.77**	**28.84**	**0.54**	**7.81**
Variance	**7864.27**	**2720.67**	**831.70**	**0.29**	**60.98**
Max	**85.50**	**346.25**	**153.50**	**24.00**	**65.00**
Min	**9.50**	**3.00**	**8.25**	**21.80**	**39.00**

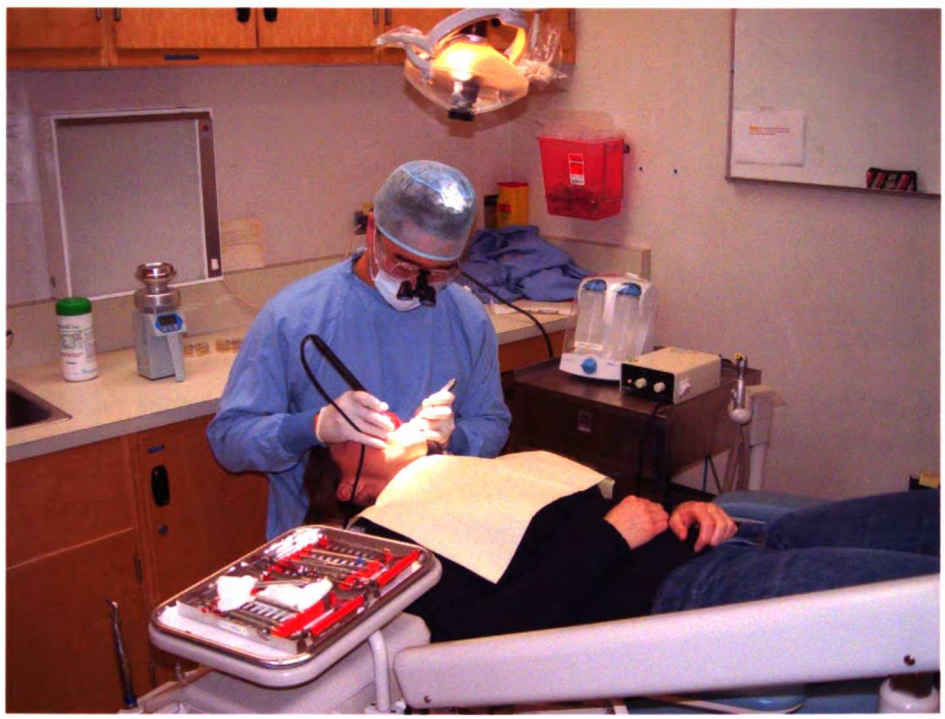


Figure 1. Intraoperative sampling during ultrasonic scaling. The air sampler is placed on the counter 24 inches behind the operator so that it is shielded from splatter.

Figure 2. The air sampler, consisting of a rechargeable battery, timer, turbine, housing for culture plates, and removable sieve through which air is circulated to the culture plate.

Figure 3. The ultrasonic scaler and the reservoir for providing mouthrinse or water as the lavage.

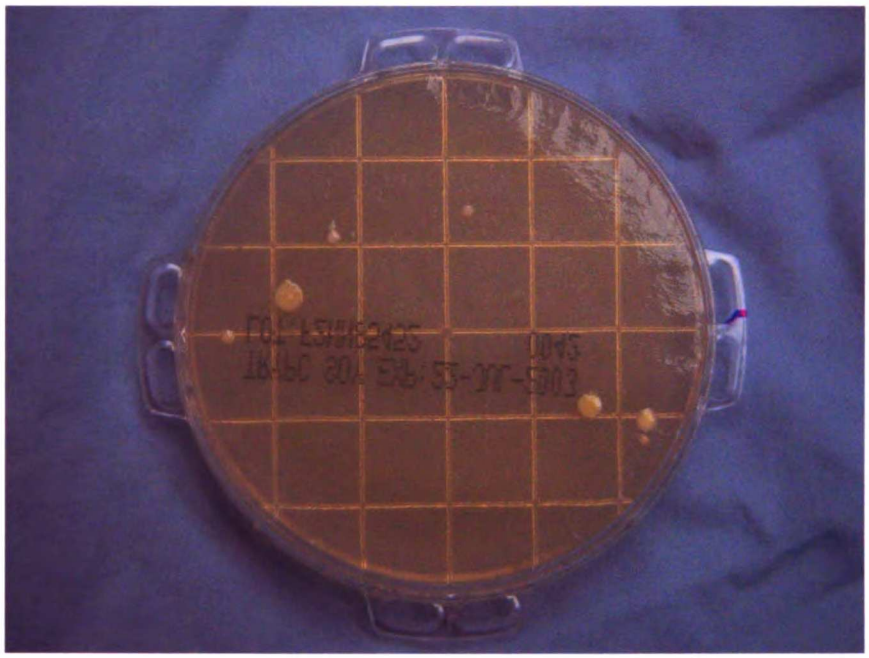
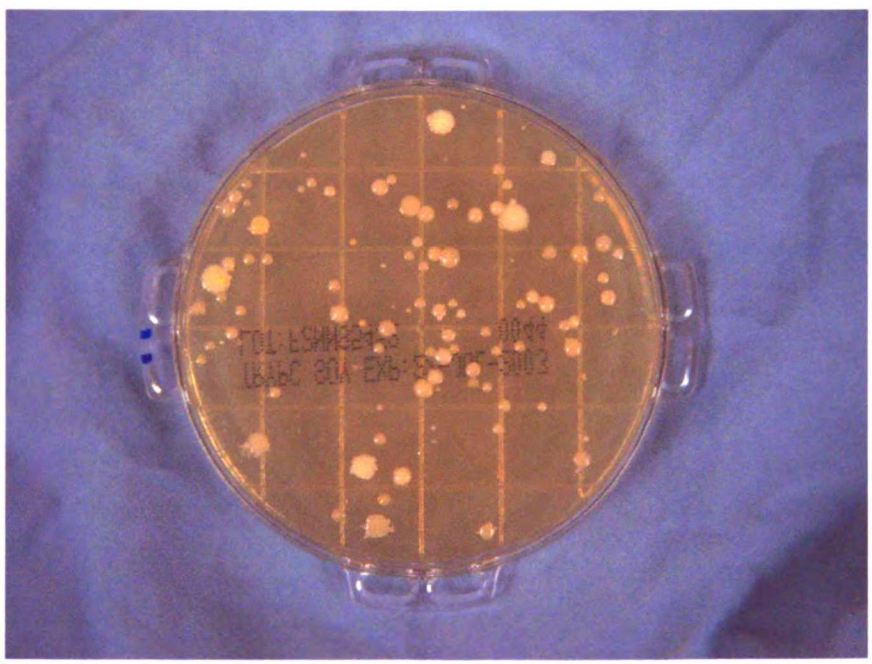


Figure 4. Culture plates with lugs that seat within the air sampler chamber. The culture medium is TSA, cultured aerobically for 48 hours at 37 C. Above, a preoperative sample; below, an intraoperative sample. Counts were made at 10.5X magnification.



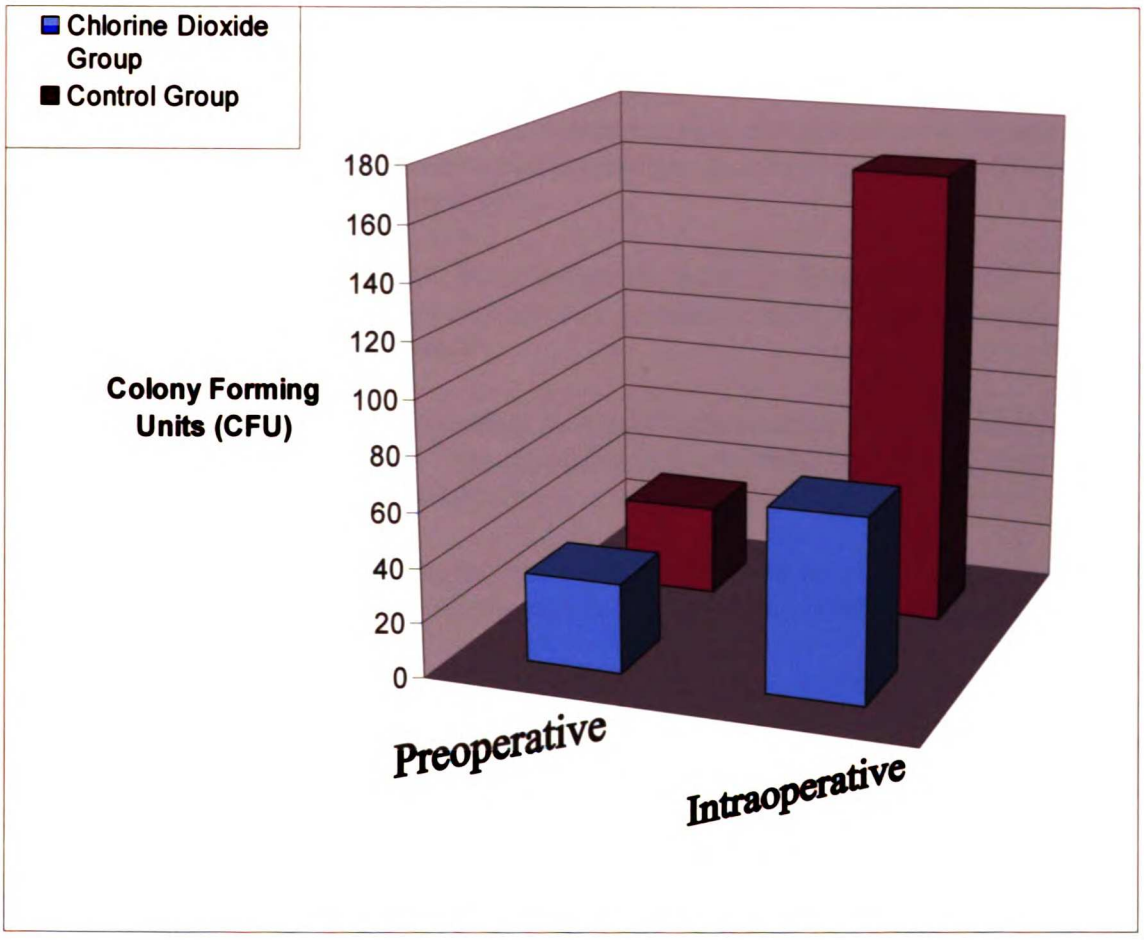


Figure 5. Graph of colony forming units for the chlorine dioxide group and the control group.

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