AEROSOL CLINICAL STUDY RESULTS

October 1, 2015

Dear Valued Oxyfresh Professional/International Partner:

WE ARE EXCITED TO SHARE EXCITING RESULTS ON A NEW CLINICAL STUDY -

“THE EFFICACY OF A PRE-PROCEDURAL MOUTH RINSE CONTAINING CHLORINE DIOXIDE ON REDUCTION OF VIABLE BACTERIAL COUNT IN DENTAL AEROSOLS DURING ULTRASONIC SCALING: A DOUBLE BLIND, PLACEBO CONTROLLED CLINICAL TRIAL”

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This Clinical study was to assess and compare the clinical and antimicrobial effects of sodium chlorite based toothpaste and mouthrinse in periodontitis patients.

A total 50 generalized chronic periodontitis patient between the ages of 18 and 55 years were enrolled in the study and divided under two categories (A and B). Clinical and microbiological parameters were recorded prior to phase 1 therapy; and subjects were put on conventional oral hygiene regime and sodium chlorite based toothpaste and mouthrinse.

The results of this study showed that there was significant decrease in clinical, and microbiological parameters from baseline to 12 months in both the groups (p < 0.01). The subjects under test group (sodium chlorite based toothpaste and mouthrinse) showed a highly significant reduction to all the parameters as compared to subjects under group B.

Conclusion: sodium chlorite based toothpaste and mouthrinse will be a true alternative for maintaining oral hygiene.

Thank you for choosing Oxyfresh!

Sincerely,

Tom Lunneborg
Vice President of Product Development
POWER RINSE (OXYFRESH): AN EFFECTIVE PRE-PROCEDURAL RINSE
POWER OF CHLORINE DIOXIDE

**40 SUBJECTS**
18 - 55 years old

**GROUP A**
No Pre-Procedure Rinse

**GROUP B**
Pre-Procedure Rinse
10 ml - 1 minute

**Professional Untrasonic Scaling**
(10 minutes - Closed fumigated operatory)

**Microbiological Analysis**
(Aerobic Culture Plate: CFU)

<table>
<thead>
<tr>
<th></th>
<th>P1</th>
<th>P2</th>
<th>P3</th>
<th>P4</th>
<th>P5</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>93 325±3 83</td>
<td>89 35±4 31</td>
<td>89 425±2 84</td>
<td>74 325±4 33</td>
<td>55 85±2 38</td>
</tr>
<tr>
<td>B</td>
<td>13 625±1 61</td>
<td>12 75±1 373</td>
<td>13 175±1 13</td>
<td>10 65±1 63</td>
<td>6 025±1 35</td>
</tr>
</tbody>
</table>

**Percentage Decreased**
(Viable Bacterial Aerosols)

<table>
<thead>
<tr>
<th></th>
<th>P1</th>
<th>P2</th>
<th>P3</th>
<th>P4</th>
<th>P5</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>85.59%</td>
<td>85.73%</td>
<td>85.27%</td>
<td>85.67%</td>
<td>89.21%</td>
</tr>
<tr>
<td>B</td>
<td>Alcohol Free</td>
<td>Stain Free</td>
<td>No Burning Sensation</td>
<td>No Dryness</td>
<td>Safe Agents</td>
</tr>
</tbody>
</table>
Efficacy of preprocedural mouth rinse containing chlorine dioxide in reduction of viable bacterial count in dental aerosols during ultrasonic scaling: A double-blind, placebo-controlled clinical trial

Rajiv Saini
Department of Periodontology, Rural Dental College, Pravara Institute of Medical Sciences, Ahmednagar, Maharashtra, India

ABSTRACT

Background: The risk to dentists, dental assistants, and patients of infectious diseases through aerosols has long been recognized. The aim of this study was to evaluate and compare the efficacy of commercially available preprocedural mouthrinses containing 0.2% chlorhexidine (CHX) gluconate, chlorine dioxide (ClO₂) mouthwash, and water in reducing the levels of viable bacteria in aerosols.

Materials and Methods: This single-center, double-blind, placebo-controlled, three-group parallel-designed study was conducted over a period of 4 months. One hundred twenty patients with chronic periodontitis were divided randomly into three groups (A, B, and C) of 40 patients each to receive the ClO₂ mouthwash, water, and 0.2% CHX gluconate respectively as preprocedural rinse. The aerosol produced by the ultrasonic unit was collected at five standardized locations with respect to the reference point, i.e., the mouth of the patient. The blood agar plates were incubated at 37°C for 48 h, and the total number of colony-forming units (CFUs) was counted and statistically analyzed.

Results: The results showed that CFUs in groups A and C were significantly reduced compared to group B, and \( P < 0.001 \) [analysis of variance (ANOVA)]. CFUs in group C underwent the highest reduction, but statistically there was no significant difference between the mean values of postprocedural CFUs in groups C and A (i.e., \( P > 0.05 \)). The numbers of CFUs were the highest at the patient's chest area and lowest at the patient's front i.e., the 6 o'clock position.

Conclusion: This study proves that a regular preprocedural mouthrinse could significantly eliminate the majority of aerosols generated by the use of an ultrasonic unit, and that ClO₂ mouthrinse was found to be statistically equally effective in reducing the aerosol contamination to 0.2% CHX gluconate.

Key words: Aerosols, chlorhexidine (CHX), chlorine dioxide (ClO₂), mouthrinses, ultrasonic scaling

Introduction

Many routine dental procedures produce aerosol and splatter composed of various combinations of the following: Water; organic particles, such as tissue and tooth dust; and organic fluids, such as blood and saliva. The microbial aerosol peak concentrations in dental treatment rooms were associated more with scaling procedures and to a lesser extent with cavity preparation. Aerosols generated by dentists in their work may contain solid particles and chemicals or gases, as well as bacteria and viruses. Bacterial cells with diameters of approximately 0.2-2.0 \( \mu m \) or viruses with diameters 20-400 nm may be found in aerosols arising from an operative procedure or from subsequently altered splatter. Within a general dental practice, numerous procedures are performed on a daily basis that result in the production of aerosols and splatter. These aerosols may be inhaled into the lungs and reach the alveoli, or they may come in contact with the skin or mucous membranes. Most of the aerosols produced during treatment procedures have diameters \( \leq 5 \mu m \),
and these can cause respiratory or other health problems as they can penetrate into and remain within the lungs. One of the methods of reducing overall bacterial counts produced during dental procedures is preprocedural rinsing with a product containing an antimicrobial agent. Chlorhexidine (CHX) gluconate, a bisbiguanide, is considered to be the most effective antiplaque agent, but it also has some side effects, notably tooth staining, taste alteration, enhanced supragingival calculus formation, and, less commonly, desquamation of the oral mucosa. In an earlier study, rinsing with CHX mouthwash led to a 94.1% reduction in recoverable colony-forming units (CFUs) compared to the nonrinsed control, while the control rinse produced a 33.9% reduction. Studies have stated that 1% povidone/iodine used as a preprocedural mouthrinse has a bactericidal effect on the microorganism, resulting in the reduction of surviving microorganisms for up to 4 h. In another study, it was observed that 0.05% cetylpyridinium chloride was found to be equally effective as CHX in reducing splatter bacteria during ultrasonic scaling.

But with the association of scientifically proven side effects with CHX (0.2%), newer, tissue-friendly mouthrinses with the power of stabilized chlorine dioxide (ClO₂) need to be evaluated. ClO₂ is widely used in various fields for its safe and highly antibacterial activity. This compound has the ability to effectively clean oral tissues on a daily basis without causing harmful side effects.

The aim of this study was to evaluate and compare the efficacy of bacterial aerosol contamination generated by ultrasonic scalers following preprocedural rinse with commercially available ClO₂, 0.2% CHX, and water.

**Materials and Methods**

This clinical trial is registered under Clinical Trials Registry — India (CTRI) no. REF/2014/06/007100. The present study was conducted at the Department of Periodontology, Loni, India after it was approved by the Technical and Ethical Committee of the Pravara Institute of Medical Sciences University, Loni, Ahmednagar, Maharashtra, India. This single-center, double-blind, placebo-controlled, three-group parallel-designed study was conducted over a period of 4 months. The subjects enrolled in this study were selected from the Outpatient Department of Periodontology, Pravara Institute of Medical Sciences, Loni, India. The patients were initially screened for their plaque index (PI) (Silness and Loe) and gingival index (GI) (Loe and Silness) scores, and 120 subjects from both the sexes and with ages ranging 18-55 years, willing to participate in the study, and having a PI score of 2-3 and a GI score of 2-3 were selected for this study after informed consent was taken from them.

In relation to rinse schedules, a double-masked protocol was maintained in this study; the patients were recruited in chronological order by systematic sampling and were randomly allotted to one of the three groups by the examiner. They were then moved to a separate clinical operatory, where they were examined by the examiner for inclusion parameters. The preprocedural rinse was given to the participants, and once the patients performed the rinse, the operator performed scaling. The operator was not involved in any evaluations before or after. The treatment group was concealed from the patient, the operator, and the microbiologist.

Inclusion criteria included:
- a. Having a minimum of 20 permanent teeth,
- b. Not having undergone any dental treatment for the past 3 months,
- c. Moderate to severe gingivitis, i.e., a GI score of 2-3,
- d. Systemically healthy patients.

The exclusion criteria for the study were
- a. The presence of any systemic disease,
- b. The presence of a disease with possible effects on the immune system,
- c. Received antibiotics or nonsteroidal anti-inflammatory drugs (NSAIDs) in the past 9-11 weeks,
- d. Oral prophylaxis with last 3 months,
- e. Pregnant and lactating mothers,
- f. Smokers.

Blood agar plates were used to sample the air during the experimental procedure. Blood agar was chosen because it is a general purpose, nonselective and enriched medium that promotes the growth of microorganisms, such as those sampled from air. Table 1 shows the five standardized locations of the blood agar plates placed

<table>
<thead>
<tr>
<th>Table 1: Standardized distances of plates</th>
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</thead>
<tbody>
<tr>
<td><strong>Plate no.</strong></td>
</tr>
<tr>
<td>------------------------------------------</td>
</tr>
<tr>
<td>Plate 1 (P1)</td>
</tr>
<tr>
<td>Plate 2 (P2)</td>
</tr>
<tr>
<td>Plate 3 (P3)</td>
</tr>
<tr>
<td>Plate 4 (P4)</td>
</tr>
<tr>
<td>Plate 5 (P5)</td>
</tr>
</tbody>
</table>

Reference point: Mouth of the patient
in operatory room for each treatment group and fixed distances of the plates were also maintained with respect to the reference point, i.e., the mouth of the patient [Figure 1].

A closed operatory with the facility to fumigate the room was used for all treatment procedures. Prior to the procedure the surfaces of the operatory were disinfected with ethyl alcohol (70%). Only one subject was treated per day, and the treatment ended on the same day. Prior to the procedure, the ultrasonic unit was switched on and flushed for 2 min, as directed by the manufacturer, in order to get rid of contaminated water due to overnight stagnation in waterlines. 45 min prior to the procedure, a blood agar plate was positioned on the plate 2 spot for a period of 15 min. This was further subjected to microbial assessment in order to check for environmental contamination, if present in the operatory.

One hundred twenty patients who met the minimal criteria for entry were selected. The nature of the procedure and the likely discomforts and risks were fully explained, and informed consent was obtained from each patient. Patients were recruited in chronological order by systematic sampling and were randomly allocated to one of the three following groups: Group A: (ClO$_2$), group B: (water), and group C: (0.2% CHX). Strict asepsis was followed inside the operatory, and the selected subjects entered the operatory wearing headcaps and autoclaved gowns. All activities such as conversation, sneezing, and coughing were strictly prohibited (if any such action occurred incidentally, then that subject was excluded from the study), and the subjects were instructed to refrain from all actions that generate aerosols. The patient was made to sit in a reclined position with his mouth at a standardized height of 3 ft from the floor of the operatory.

Ultrasonic scaling was performed using an EMS ultrasonic scaler. Distilled water was used for all the ultrasonic scaling procedures. Coolant water flow and power settings were adjusted to a medium mode. The amount of water flow from the ultrasonic scaler during 1 min was then measured using a graduated cylinder. Based on these measurements, a water coolant volume of 15 mL per min was used during all the measurements of aerosol contamination. Prior to each trial, the coolant flow of the ultrasonic scaler was adjusted to this volume of water to ensure that coolant volume was consistent for all the trials.

Ultrasonic scaling was done on a randomly selected quadrant (control side) with the ultrasonic scaler for a period of 10 min. Following the 10-min sampling period, blood agar plates were covered and taken off the tray. A gap of 30 min was followed after the scaling procedure so as to allow the aerosols to settle down. After the gap of 30 min, fairly fresh blood agar plates were placed in a similar fixed position with regard to the reference point. The subject was then assigned to 1 min of 10 mL rinse with 0.2% CHX, ClO$_2$, or water. Ultrasonic scaling was again done with the same ultrasonic scaler on the other side (test side) of the same arch for a period of 10 min. Following the 10-min sampling period, blood agar plates were covered and taken off the tray. The blood agar plates were then transported immediately to the microbiology laboratory for microbial assessment. The blood agar plates were incubated at 37°C for 48 h. After the incubation period, the plates were observed for microbial growth. Using a colony counter, the resulting CFUs were counted for each plate. Each colony was assumed to represent a single viable particle in the air, and the microbial concentration was defined as the number of viable particles per cubic foot of air.

The key ingredients of mouthrinses used in the study are illustrated in Table 2.

For statistical analyses, individual measurements were summarized within each individual and then analyzed.
Statistical analysis was performed by applying mean, standard deviation (SD), Student’s unpaired t-test, Probability P, analysis of variance (ANOVA), and Tukey-Kramer multiple-comparison tests.

**Results**

One hundred twenty participants, 40 in each group, were analyzed, and analysis was according to intention to treat, as illustrated in Figure 2; the noncompliance rate for our study was 0 and there was no one who dropped out after randomization. Table 3 displays the age- and sex-wise distribution of the subjects in each group. The mean age of the subjects was 32 years and no significant differences in age or sex were found among groups for any demographic variables. Figure 3 shows the comparison of mean values of the GI and the PI for all three groups. By applying the one-way ANOVA test for repeated measures; the variations among column means were not significantly greater than expected than chance and by applying the Tukey-Kramer multiple-comparison test there was no significant difference between the mean values of GI and PI in Group A, B, and C when compared together, where the value of $F = 1.184$, ($F=\text{variance of the group means / mean of the within group variances}$), $P = 0.3177$, not statistically significant.

The mean and SD values of CFUs for each of the three treatment groups at the five standardized plate locations are summarized in Table 4. This analysis revealed that the 0.2% CHX group showed the maximum reduction of CFUs at all the five plate locations, followed by $\text{ClO}_2$. Compared with the control group (water), both the test groups (0.2% CHX and $\text{ClO}_2$) showed the efficiency of those products in reducing the number of CFUs. The numbers of CFUs were the highest at plate P1 (patient’s chest) and the lowest at plate P5 (6 o’clock position). Comparison of the mean and SD values of CFUs in group A: (ClO$_2$), group B (water), and group C (0.2% CHX) by applying Student’s paired t-test there showed a highly significant difference between the mean values of CFUs values from preprocedural to postprocedural in group A and C where $P < 0.01$, and no significant difference was observed in group B where $P > 0.05$ at all the plates, i.e., P1 to P5.

Comparison of mean postprocedural values of CFUs in group $\text{ClO}_2$ versus water, $\text{ClO}_2$ versus 0.2% CHX, and water versus 0.2% CHX were illustrated in Tables 5-7. On applying the Student’s unpaired t-test, there was found a highly significant difference between the mean values of postprocedural CFUs in group A versus B and group B versus C (i.e., $P < 0.001$), while there was no significant difference between the mean values of postprocedural CFUs in group A versus C (i.e., $P > 0.05$).

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**Table 2: Ingredients of mouthrinse**

<table>
<thead>
<tr>
<th>Mouthrinse</th>
<th>Trade name</th>
<th>Ingredients</th>
</tr>
</thead>
<tbody>
<tr>
<td>ClO$_2$</td>
<td>Power Rinse Oxyfresh®, USA</td>
<td>Deionized water; zinc acetate; sodium citrate; chlorine dioxide concentrate (15% solution); xylitol; sucralose; aloe powder; sodium hydroxide and citric acid. In addition, it is a nonalcoholic preparation, with no dye or color</td>
</tr>
<tr>
<td>CHX</td>
<td>Hexidine ICPA, India</td>
<td>CHX gluconate solution I.P. diluted to CHX gluconate 0.2% in aqueous base</td>
</tr>
</tbody>
</table>

ClO$_2$ = Chlorine dioxide; CHX = Chlorhexidine

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**Table 3: Age- and sex-wise distribution of subjects in groups A, B, and C**

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Group A: ClO$_2$ ($n = 40$)</th>
<th>Group B: Water ($n = 40$)</th>
<th>Group C: 0.2% CHX ($n = 40$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>M</td>
<td>F</td>
<td>M</td>
</tr>
<tr>
<td>&lt;20</td>
<td>2</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>20-30</td>
<td>2</td>
<td>5</td>
<td>3</td>
</tr>
<tr>
<td>30-40</td>
<td>11</td>
<td>11</td>
<td>10</td>
</tr>
<tr>
<td>40-50</td>
<td>4</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>&gt;50</td>
<td>1</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
</tbody>
</table>

Mean±SD: 31.24±11.24, 32.24±10.24, 30.95±11.32, 32.58±12.04, 33.25±14.25, 33.06±11.25

ClO$_2$ = Chlorine dioxide; CHX = Chlorhexidine; SD = Standard deviation; M = Male; F = Female
Discussion

Effective infection control is one of the cornerstones of good dental practice. However, due to many drawbacks, infection control has not been achieved to the greatest level of satisfaction. A marked increase in the airborne organisms was demonstrated in the samples collected from dental clinics where ultrasonic scalers were in use.\(^{[11]}\)

So there remains a risk of transmission of potentially harmful infectious agents to dentists and patients through several vectors including instruments and the air.\(^{[12]}\)

Many dental procedures such as ultrasonic scaling, air polishing, orthodontic debonding, and cavity preparation are known to produce viable bioaerosols. To effectively minimize the formation of bioaerosols, many protective barriers have been suggested, from the use of the mouth mask, to preprocedural rinse, to high-volume evacuators, to high-efficiency particulate air room filters.\(^{[13]}\)

Thus, the present study was designed to compare the efficacy of preprocedural rinse with CHX to be more effective than no rinsing, in reducing aerosols generated during oral prophylaxis using ultrasonic scalers. The results of this study showed that there was a highly significant reduction of bacterial CFU in both group A (ClO\(_2\)) and group C (0.2% CHX); however, there were no significant results in the reduction of bacterial CFU in group C (water), as seen in Figure 4.

In this study, 0.2% CHX pre-procedural rinse significantly reduced CFUs at all the five locations, compared to no preprocedural rinse. This finding is in accordance with data reported by Feres M \textit{et al.}\(^{[9]}\). These results are also consistent with those reported by Muir \textit{et al.},\(^{[14]}\) who found that a preprocedural rinse with CHX to be more effective than no rinsing, in reducing aerosols generated by ultrasonic scaler. The enhanced efficacy of 0.2% CHX in reducing the CFUs could be because of the reason

\[\text{CHX} + \text{bioaerosols} \rightarrow \text{inactive CFUs} + \text{biologically inert particulate material}\]

Hence, the enhanced efficacy of 0.2% CHX pre-procedural rinse significantly reduced CFUs at all the five locations, compared to no preprocedural rinse with CHX. The enhanced efficacy of 0.2% CHX in reducing bacterial CFUs compared to water and 0.2% CHX, is shown in Table 4.

<table>
<thead>
<tr>
<th>Location</th>
<th>ClO(_2) Mean ± SD</th>
<th>Water Mean ± SD</th>
<th>0.2% CHX Mean ± SD</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>P1</td>
<td>13.625±1.61</td>
<td>9.63±2.84</td>
<td>11.12±2.10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>P2</td>
<td>12.75±1.373</td>
<td>90.37±2.72</td>
<td>10.78±1.69</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>P3</td>
<td>13.175±1.13</td>
<td>88.56±3.36</td>
<td>8.78±1.10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>P4</td>
<td>6.025±1.35</td>
<td>54.35±3.13</td>
<td>3.90±1.18</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

\(P = \text{Plate locations (P1-P5)}; \text{SD} = \text{Standard deviation}; \text{HS} = \text{Highly significant}\)

Table 5: Comparison of mean and SD values of CFUs in ClO\(_2\) vs water

\(P = \text{Plate locations (P1-P5)}; \text{SD} = \text{Standard deviation}; \text{HS} = \text{Highly significant}\)

Table 6: Comparison of mean and SD values of CFUs in ClO\(_2\) vs 0.2% CHX

\(P = \text{Plate locations (P1-P5)}; \text{SD} = \text{Standard deviation}; \text{NS} = \text{Not statistically significant}\)

Table 7: Comparison of mean and SD values of CFUs in water vs 0.2% CHX

\(P = \text{Plate locations (P1-P5)}; \text{SD} = \text{Standard deviation}; \text{HS} = \text{Highly significant}\)

\[\text{CHX} + \text{bioaerosols} \rightarrow \text{inactive CFUs} + \text{biologically inert particulate material}\]
that CHX starts its antimicrobial action at the point of generation of aerosols and also at the time of onset of formation of aerosol. The antiplaque activity of CHX appears to be due to the retention of the drug in oral tissues and its subsequent slow release in an active form.

Similarly, when ClO$_2$ was used as a preprocedural rinse, fewer CFUs were developed than without preprocedural rinse. The enhanced efficacy of ClO$_2$ in reducing the CFUs could be because ClO$_2$ may act as a strong component in the obliteration of the microbiota via oxygenation and neutralization of toxins produced by the bacteria in the oral cavity. The stabilized ClO$_2$-based products also destroy the volatile sulfide compounds (VSCs), which further reduces the triggering of gingival inflammation. In vitro studies demonstrated stabilized ClO$_2$-based oral rinse microbicidal activity against various oral pathogens.$^{[15-19]}$ These studies showed that ClO$_2$-based oral rinse kills oral bacteria associated with the development and/or progression of oral diseases up to 99% in 10 s, and that the oral rinse is less toxic than CHX to human gingival cells in vitro.

In this study, it was also observed that on applying Student’s unpaired $t$-test, there was no significant difference between the mean values of postprocedural CFU in ClO$_2$ and 0.2% CHX (i.e., $P > 0.05$) as seen in Table 5. Thus, by using any of the preprocedural mouthrinses, both will be equally potent in reducing the bacterial aerosols during ultrasonic scaling.

There was a lot of research support, and newer, higher versions of medicated combinations are available in the market, compared to the traditional follow-up of alcohol-based mouthwash. The product with the key ingredient of sodium chlorite (i.e., liberates ClO$_2$) has been confirmed to have equal potential in terms of reducing aerosol contamination with minimal side effects and more tissue compatibility.

The highest bacterial counts were detected on the plate 1, positioned at the patient’s chest. These findings agree with those of Bentley et al.$^{[20]}$ who observed that the larger salivary droplets generated during dental procedures settle rapidly from the air with heavy contamination on the patient’s chest. Next-higher counts were found on the plate 2, positioned toward the operator, followed by plate 3, positioned toward the assistant’s side. In addition, moderate bacterial contamination was found on plates 4 and 5 respectively.

This study demonstrated that a sufficient amount of aerosol and spatter from the patient is ejected far enough to come in contact with the dental personnel performing the treatment. Though the results underlined the need for mouth rinsing before dental procedure, ironically, few dentists put it into practice as a regular regimen in their dental practice. One of the barriers to such implementation is the strong bitter taste, brown discoloration of teeth, dryness, and burning sensation associated with the traditionally used alcohol-based mouthrinses. In such situations, ClO$_2$-based mouthrinses would be a genuine alternative, as these products showed equal efficiency in reducing the aerosol contamination compared to the alcohol-based products. But the advantage and edge over the traditional alcohol-based mouth rinse is that the ClO$_2$-based mouthrinses are more tissue-friendly, with no side effects such as burning sensations, dryness, taste alterations, or staining.$^{[10]}$

The limitations of this study should be considered in the interpretations of the results. The CFUs that were counted here are the values that represent the bacteria capable of growing on blood agar plates. No attempt has been made to identify the bacteria: Either pathogen or nonpathogen. However, viruses, fungi, and specific bacteria require specialized media that were not cultured in this study. Future studies are needed to investigate the viable pathogenic microorganisms generated during the use of ultrasonic scaling devices.

**Conclusions**

Aerosol production during ultrasonic scaling is very hazardous to the patient, the operator, and the public at large. Hence, preprocedural rinsing should be made a regular practice in all dental setups, along with high-
vacuum evacuation and other barrier techniques.[21] ClO₂ mouthrinse was found to be statistically equally effective in reducing the aerosol contamination produced by ultrasonic scaling to, though slightly less potent than, 0.2% CHX. However, considering the disadvantages of alcohol based mouthrinse (strong bitter taste, brown discoloration of teeth, dryness, and burning sensation), ClO₂-based mouthrinses can serve as effective and safe agents for aerosol control during professional ultrasonic scaling in a dental setup.

References


Source of Support: Nil, Conflict of Interest: None declared.
Chlorine dioxide: An ideal preprocedural mouthrinse in dental set-up

Rajiv Saini
Department of Periodontology, Pravara Institute of Medical Sciences, Loni, Ahmednagar, Maharashtra, India

ABSTRACT

Background: Aerosols generated during ultrasonic scaling is a potential risk factor for cross-contamination in dental settings. The aim of this study is to evaluate and compare the efficacy of commercially available chlorine dioxide as preprocedural mouthrinses in reducing the level of viable bacteria in aerosols.

Materials and Methods: This single-center clinical double-blinded study was conducted over a period of 4 months. A total of 80 patients were divided randomly into two groups (A and B) of 40 patients each to receive the chlorine dioxide mouthwash and water as preprocedural rinse. The aerosol produced by the ultrasonic unit was collected at five standardized location with respect to the reference point, that is, the mouth of the patient. The blood agar plates were incubated at 37°C for 48 h, and total number of colony-forming units (CFUs) was counted and statistically analyzed.

Results: The results showed that CFUs in test group A were significantly reduced compared with control group B, \( P < 0.001 \) (analysis of variance). The numbers of CFUs were highest in the patient chest area and lowest at the patient front, that is, 6 o’clock position.

Conclusion: This study proves that a regular preprocedural mouthrinse with chlorine dioxide could significantly reduce aerosols generated during professional oral prophylaxis.

Key words
Aerosols, chlorine dioxide, mouthrinses, ultrasonic scaling

INTRODUCTION

The oral cavity is a reservoir for a large number of microorganisms including bacteria and viruses. This ecological niche can be a pool for opportunistic and pathogenic microorganisms that can pose a risk for cross-contamination and infection and may even cause systemic infections. This is of particular importance in the case of routine dental practice, as the risk of exposure to microorganisms in the oral cavity is increased due to the open and invasive nature of the procedures. There are a number of possible means by which transmission of viral and bacterial pathogens can occur in the dental practice. The patient’s own saliva and blood are major vectors of cross-transmission. Blood-borne contamination can occur by exposure to the infectious material through the nonintact skin and mucosal lesions.\(^\text{[1]}\) The use of an antimicrobial mouthrinse by the patient before dental procedures is based on a similar principle of reducing the number of oral microorganisms. This reduction also reduces the number of microorganisms that may escape a patient’s mouth during dental care through aerosols, spatter, or direct contact. Aerosols are of great concern since they can remain suspended in the air for a great length of time. Hygienists utilizing prophy cups and ultrasonic scalers need to focus on limiting splatter and aerosols as well as lowering the amount of bacteria.\(^\text{[2,3]}\) These aerosols may be inhaled into the lungs to reach the alveoli or may come in contact with the skin or mucous membranes. Most of the aerosols produced during treatment procedures have a diameter of 5 \( \mu \text{m} \) or less, and these can cause respiratory or other health problems because they can penetrate into, and remain within the lungs.\(^\text{[4,5]}\) Chlorhexidine gluconate, a bisbiguanide, is considered to be the most effective anti-plaque agent,\(^\text{[6]}\) but it also has some side-effects, notably tooth staining, taste alteration, enhanced supragingival calculus formation and less commonly desquamation of the oral mucosa.\(^\text{[7]}\) Hence, in this clinical study an attempt has been made to evaluate the efficacy of preprocedural rinse of chlorine dioxide based mouthrinse (Oxyfresh\(^\text{®}\) Power
Rinse) in reducing the microbial content of the aerosol in dental office.

MATERIALS AND METHODS

Study population
Totally, 80 systemically healthy individuals, age ranged 18–55 years were selected for participation in the study as illustrated in Table 1. Inclusion criteria was: Dentition with ≥20 teeth (minimum of five teeth per quadrant), with plaque index (PI) (Silness and Loe) and gingival index (Loe and Silness) scores between 2 and 3 were selected in the study. Patients with other oral lesions, wearing any fixed or removable prosthesis, and with any past history of systemic illness or allergy to components of mouth rinse were excluded from the study. The selected subjects were further instructed not to mouthrinse on the day of appointment. All subjects were explained the purpose of the study and informed consent was obtained from them. The study was approved by the Institutional Ethical Committee.

Study design
This was a clinical double-blinded interventional study; the preprocedural rinse was given to participants, and once the patients performed the rinse, the same operator performed scaling. The operator was not involved in any evaluations before or after. The treatment group was concealed from the patient, operator, and microbiologist. Study populations were randomly assigned into two groups who underwent prophylaxis after preprocedural rinsing for 1 min before scaling was performed, that is, test group (A) - Chlorine dioxide mouthrinse and control group (B) - Sterile water. The key ingredients of the chlorine dioxide mouthrinse used in the study is deionized water; zinc acetate; sodium citrate; chlorine dioxide concentrate (15% solution); xylitol; sucralose; aloe powder; sodium hydroxide and citric acid. In addition, it is nonalcoholic preparation, with no dye and color. To avoid aerosol contamination, the operating area was fumigated on the day before the treatment. Only one patient/day was treated on alternate days with ultrasound scaling. Before ultrasonic scaling, agar plates were placed on five standardized positions in context to a reference point, that is, patient’s mouth as illustrated in Table 2.

Clinical protocol
Oral prophylaxis was done on a randomly selected quadrant (control side) with the ultrasonic scaler for a period of 10 min. After the gap of 30 min, fair fresh blood agar plates were kept on the similar fixed position from the reference point as shown in Figure 1 (culture plate locations). The subjects were instructed to rinse with 10 ml mouthrinse (control and test) for a period of 1 min. Oral prophylaxis was again done with the same ultrasonic scaler on the other side (test side) of the same arch for a period of 10 min. Coolant water flow and power setting were adjusted on a medium mode. The amount of water flow from the ultrasonic scaler during 1 min was then measured using a graduated cylinder. Based on these measurements, a water coolant volume of 15 ml/min was used during all the measurements of aerosol contamination. Following the 10 min sampling
period, blood agar plates were covered and taken off the tray. All agar plates were sent for microbiological analysis to the microbiological laboratory for the colony-forming unit (CFU) count on the same day of ultrasonic scaling procedure.

RESULTS

By applying Student’s unpaired t-test there was no significant difference between mean values of index (gingival index) and PI in both the groups (test and control) as illustrated in Table 3; that confirmed that all the subjects involved in both the groups in this study were equally affected with gingival inflammation. By applying Student’s paired t-test there was a highly significant difference between mean values of CFUs values at all the plates from pre to post in test group A (chlorine dioxide) where value of P < 0.01; while no significant difference observed in control group B (sterile water) where value of P > 0.05 as shown in Table 4. By applying Student’s unpaired t-test there was a highly significant difference between mean values of post-CFU in groups A and B in all the plates as shown in Table 5.

DISCUSSION

Aerosol and splatter are a concern in dentistry because of their potential effects on the health of the immune-compromised patients and on dental personnel. There are also regulations by the Occupational Safety and Health Administration about aerosol contamination abolition as a part of standards for indoor air quality. One of the reports indicated that the ultrasonic scaler is the greatest producer of contaminated aerosol and splatter. Use of an antiseptic mouthwash by the patient prior to ultrasonic scaling has also been shown to be effective in reducing bacterial aerosols during treatment. When chlorine dioxide was used as a preprocedural rinse, fewer CFUs were developed than without preprocedural rinse. The enhanced efficacy of chlorine dioxide in reducing the CFUs could be because of the reason that sodium chlorite (stabilized chlorine dioxide) may acts as a strong component to obliterate the microbiota via oxygenation and neutralization of toxins. The stabilized chlorine dioxide based products also destroy the volatile sulfide compounds, which further reduce the triggering of gingival inflammation. Chlorine dioxide also plays a vital role in damaging the cell membrane of the bacteria. The percentage changes for value of CFU from pre to post were 85.59% in plate 1, 85.73% in plate 2, in 85.27% plate 3, 85.67% in plate 4 and 89.21% in plate 5, respectively. These results confirmed that the preprocedural rinse with chlorine dioxide based mouth rinse was competent enough to reduce the viable bacterial count in aerosol during ultrasonic scaling in the dental operatory. The highest bacterial counts were detected on the plate 1 positioned at the patient’s chest as illustrated in Figure 2 (colony formation in culture plate for groups A and B). These findings agrees with that of Bentley and Nancy who observed that the larger salivary droplets generated during dental procedures settle rapidly from the air with heavy contamination on patient’s chest. Next higher counts were found on the plates 2, positioned towards operator followed by plate 3, positioned towards the assistant side. Furthermore, a moderate bacterial contamination was found on plates 4 and 5 respectively. Compliance to the preprocedural is the main hurdle, and most of the conventional mouthrinse are alcohol based that leads to burning sensations, dryness, taste alterations and staining. Chlorine dioxide based mouth rinse would be a true alternative in reducing the aerosol contamination with the advantage over the traditional alcohol based mouth rinse as they are more

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**Table 3: Comparison of mean and SD values of GI and PI**

<table>
<thead>
<tr>
<th>Clinical parameters</th>
<th>Mean±SD (n=40)</th>
<th>Student’s unpaired t-test value and significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A: Chlorine dioxide</td>
<td>Group B: Sterile water</td>
</tr>
<tr>
<td>GI</td>
<td>2.757±0.277</td>
<td>2.7425±0.227</td>
</tr>
<tr>
<td>PI</td>
<td>2.675±0.225</td>
<td>2.681±0.2345</td>
</tr>
</tbody>
</table>

GI – Gingival index; PI – Plaque index; SD – Standard deviation

**Table 4: Comparison of mean and SD values of CFUs from pre to post**

<table>
<thead>
<tr>
<th>Culture plates</th>
<th>Mean±SD (n=40)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A: Chlorine dioxide</td>
<td>Group B: Sterile water</td>
</tr>
<tr>
<td>Plate 1 pre</td>
<td>93.32±53.83</td>
<td>92.50±3.01</td>
</tr>
<tr>
<td>Plate 1 post</td>
<td>13.62±1.61</td>
<td>90.62±1.84</td>
</tr>
<tr>
<td>Plate 2 pre</td>
<td>89.35±4.31</td>
<td>92.32±3.45</td>
</tr>
<tr>
<td>Plate 2 post</td>
<td>12.75±1.37</td>
<td>90.37±2.72</td>
</tr>
<tr>
<td>Plate 3 pre</td>
<td>89.42±5.28</td>
<td>90.63±3.06</td>
</tr>
<tr>
<td>Plate 3 post</td>
<td>13.75±1.13</td>
<td>88.56±3.36</td>
</tr>
<tr>
<td>Plate 4 pre</td>
<td>74.32±54.33</td>
<td>74.36±3.93</td>
</tr>
<tr>
<td>Plate 4 post</td>
<td>10.65±1.63</td>
<td>71.54±3.30</td>
</tr>
<tr>
<td>Plate 5 pre</td>
<td>55.85±2.38</td>
<td>56.27±2.95</td>
</tr>
<tr>
<td>Plate 5 post</td>
<td>6.02±5±13.35</td>
<td>54.35±3.13</td>
</tr>
</tbody>
</table>

SD – Standard deviation; CFUs – Colony forming units

**Table 5: Comparison of mean and SD values of CFUs from post to post**

<table>
<thead>
<tr>
<th>Plates</th>
<th>Mean±SD (n=40)</th>
<th>Student’s unpaired t-test and P with significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A: Chlorine dioxide</td>
<td>Group B: Sterile water</td>
</tr>
<tr>
<td>Plate 1</td>
<td>13.62±1.61</td>
<td>90.62±1.84</td>
</tr>
<tr>
<td>Plate 2</td>
<td>12.75±1.37</td>
<td>90.37±2.72</td>
</tr>
<tr>
<td>Plate 3</td>
<td>13.75±1.13</td>
<td>88.56±3.36</td>
</tr>
<tr>
<td>Plate 4</td>
<td>10.65±1.63</td>
<td>71.54±3.30</td>
</tr>
<tr>
<td>Plate 5</td>
<td>6.02±5±13.35</td>
<td>54.35±3.13</td>
</tr>
</tbody>
</table>

SD – Standard deviation; CFUs – Colony forming units
tissue friendly with no side-effects and good compliance among the patients.

**CONCLUSION**

Preprocedural rinse used by patients before a dental procedure are anticipated to reduce the number of pathogens released by a patient in the form of aerosols or spatter that subsequently can contaminate equipment, operatory surfaces, and dental health care personnel. Though aerosol production cannot be totally eradicated with infection control procedures, the hazards of these aerosols can be minimized by preprocedural rinsing. The results of this study confirmed that Prerinsing with chlorine dioxide based mouthrinse (Oxyfresh® Power Rinse) was effective in reducing the aerosol contamination. More longitudinal multi centric studies with larger subjects will be planned to precisely analyze and compare the effectiveness of the chlorine dioxide bases mouth rinses with alcohol based mouthrinses.

**REFERENCES**


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