

Research Article

Comparative Evaluation of Efficacy of Chlorhexidine and Herbal Mouthwash as a Preprocedural Rinse in Reducing Dental Aerosols: A Microbiological Study

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Objective. The risk to dentists, 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 mouth rinses containing 0.2% chlorhexidine (CHX) gluconate, Befresh™ herbal mouthwash, and water in reducing the levels of viable bacteria in aerosols. **Materials & Methods.** This was a single-center, double-masked, placebo-controlled, randomized, three-group parallel design study. 30 patients (10 patients in each group) were recruited in the study. Patient rinsed mouth with 10 ml of CHX, 10 ml Befresh™, or 10 ml water. All the patients underwent supragingival ultrasonic scaling for a minimum of 30 min. The aerosol collection was done using a blood agar plate. The blood agar plates were kept approximately 12 inches from the patient's mouth. The microbial culture was analyzed. The colony-forming unit (CFU) counting in all three groups was assessed using one-way ANOVA test to compare among the groups (p value <0.001). The intergroup comparison was done using the post hoc Tukey test. **Result.** There was a marked reduction in the CFU in the CHX group in all three areas. This was followed by Befresh™ (Sagar Pharmaceuticals) mouthwash. There was no reduction in the CFU of the water group. **Conclusion.** This study proves that a regular preprocedural mouth rinse could significantly reduce the majority bacteria present in aerosols generated by the use of an ultrasonic unit, and Befresh™ mouth rinse was found to be equally effective in reducing the aerosol contamination to 0.2% CHX gluconate.

1. Introduction

The dissemination of oral microbes following various dental procedures has been a concern in clinical practice. Equipment such as ultrasonic scalers, high-speed handpieces, or three-way syringes might cause the spread of aerosol in the environment [1–4]. The aerosol from dental procedures is

composed of water, microbes, tissue, tooth dust, and fluids such as saliva and blood. The spread of aerosol might relate to cross-infection in the dental clinic, resulting in impairment of the health status of patients, dental professionals, and dental assistants. Within a general dental practice, numerous procedures are performed on a daily basis that results in the production of aerosols and splatter. The

concentration of dental aerosol is more in ultrasonic scaling procedures compared to the cavity cutting procedure. Most of the aerosols produced during treatment procedures have diameters $\leq 5 \mu\text{m}$, and they can gain entry in our mouth and respiratory system or can come in contact with skin and mucous membrane. They can be lodged in the lungs and can be dangerous [5].

Various steps have been considered to prevent aerosol-related cross-infection, such as immunization for dental professionals and assistants, sterilization, disinfection, and use of protective barrier and preprocedural mouthwash [6–10]. Using a preprocedural rinsing with an antimicrobial agent prior to the dental procedure is another easy, economical, and safe method to produce a reduction in overall bacterial load during dental treatment.

Among the mouthwashes, chlorhexidine (CHX) is considered the gold standard substance in controlling oral biofilm growth in the oral cavity or microbial spread by oral aerosols [3, 7, 11–13] due to its broad antibacterial spectrum [12, 14] and substantivity of 8 to 12 hours [12].

However, other antiseptics have also been used as preprocedural mouthwashes, such as essential oils [15] and cetylpyridinium chloride (CPC) [13]. On the other hand, herbal mouth rinses with their natural ingredients offer a safe and effective option that should be optimally made use of [6]. The main benefit of using herbal preparations is that they do not contain alcohol or sugar, which are present in over-the-counter products and which possess the ill effects of causing bacterial growth resulting in halitosis or bad breath. In the present study, an herbal mouth rinse Befresh™ which is free from alcohol, CHX, povidone-iodine, and sugar was compared with 0.2% CHX mouthwash as a preprocedural rinse. It contains *Cinnamomum zeylanicum*, *Mentha spicata*, *Syzygium aromaticum*, and *Eucalyptus globulus*. *Cinnamomum zeylanicum* oil has antimicrobial, antioxidant, anti-inflammatory, and anticancer properties. *Mentha spicata* oil has antifungal property [3]. *Syzygium aromaticum* contains potassium, manganese, iron, selenium, vitamin A, K B6, and C. Cloves of *Syzygium aromaticum* has antimicrobial, anti-inflammatory, and analgesic property. It controls halitosis. *Eucalyptus globulus* is used as a dental solvent. It gives a clean aroma. It has antibacterial and anti-inflammatory effects.

Thus, the aim of this study was to assess and compare the efficacy of bacterial aerosol contamination generated by ultrasonic scaling following preprocedural rinse with commercially available herbal mouthwash, 0.2% CHX, and water.

2. Materials and Methods

2.1. Study Design. The present study was a single-center, triple-blinded, placebo-controlled, randomized, three-group parallel design study. This study was approved by the Human Subject Ethics Board of Manipal College of Dental Sciences, Mangalore, Manipal Academy of Higher Education, Manipal, India. It was conducted in the department of periodontology in the same institution, in accordance with the Helsinki Declaration of 1975, as revised in 2000. The study is registered at clinical trials.gov as CTRI/2019/01/

017038. The study is reported in compliance with the CONSORT guidelines.

2.2. Study Population. The sample size for the study was calculated based on a previous study by Rani et al. [4]. Thirty systemically healthy patients above 18 years of age with a minimum of 20 teeth and diagnosed with mild-moderate form of chronic periodontitis (showing more than 30% of sites with clinical attachment loss $\leq 4 \text{ mm}$) were included in the study (Figure 1). The exclusion criteria were any known systemic disease or conditions, smokers, pregnant and lactating mother, patients who underwent any periodontal treatment in last 6 months or received antibiotics or non-steroidal anti-inflammatory drugs (NSAIDs) in the past 8 weeks, and patients who are already using mouthwash or who have hypersensitivity to CHX mouthwash. All the included study participants were informed about the study and a written consent was obtained regarding the same.

2.3. Randomization and Blinding. Randomization was done using a computer-generated table. The included patients were divided randomly into three groups (group A–C) of ten patients each to receive 0.2% CHX gluconate mouthwash (group A), herbal mouthwash (group B), and water (group C), respectively, as a preprocedural rinse. In order to avoid contamination in the dental clinic, one patient per day was treated. This is a triple-blinded study in which the participant, the operator who performed ultrasonic scaling, and the person who did the microbial analysis were blinded from the allocated groups. The enrollment and allocation were done by a masked examiner (V. S) who was a part of the study. Ultrasonic scaling and assistance were done by the two investigators (A. K and V. R) of the study. The microbial analysis of the agar plates was done by S.N. and A.H. In all three groups, assigned mouthwash (colourless) was placed in a similar opaque bottle and was administered to blind the operator.

2.4. Sample Collection and Clinical Procedure. Demographic details (age and gender) and periodontal clinical examination (using a Williams probe Hu-Friedy, Chicago, IL, USA) of all study participants were recorded prior to the beginning of the procedure. The clinical attachment level (CAL) and probing pocket depth (PPD) were measured at mesiobuccal, mesiolingual, midbuccal, distobuccal, distolingual, and midlingual sides of each tooth, excluding third molars. The plaque index (PI) [16] was used to record the presence of plaque at distofacial, facial, mesiofacial, and lingual surfaces. Only plaque of the cervical third of the tooth was evaluated. The gingival condition was assessed using the gingival index (GI) [17] at four surfaces: distofacial papilla, facial margin, mesiofacial papilla, and entire lingual gingival margin. All clinical data were collected by a single examiner, who had been calibrated prior to the commencement of the study. All three group patients were advised to use 10 ml allotted liquid for a minute as a preprocedural rinse prior to ultrasonic scaling for a minimum of 30 minutes. A blood agar plate was

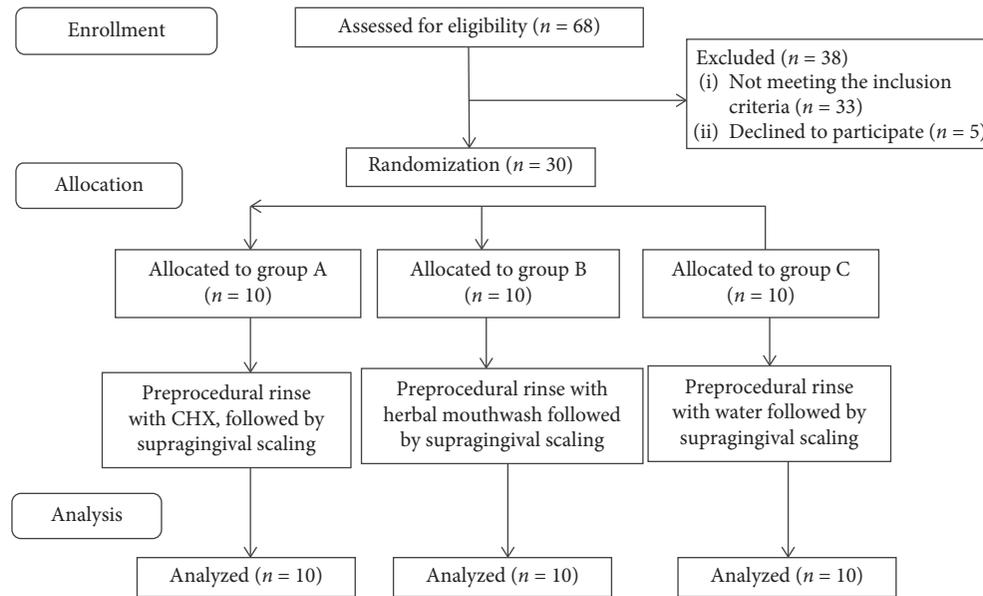


FIGURE 1: Patient flow diagram.

used to collect the airborne microorganisms as it is a valid medium for culturing airborne bacteria. Before ultrasonic scaling, the blood agar plates were positioned at the patient's chest area, at the dentist's chest area, and at the assistant's chest area with the help of double-sided adhesive tapes. The average distance was approximately 12 inches from the patient's mouth to the blood agar plate. Full mouth supragingival ultrasonic scaling was done by the same operator for all patients. Each patient underwent scaling for a duration of 30 minutes. Postscaling for 10 minutes, the blood agar plates were kept in the designated area so that the aerosol will get settled in the agar plate. The agar plates were incubated at 37°C for 48 hours. CFU in the agar plates was counted in the Microbiology Department, Kasturba Medical College, Mangalore, Manipal Academy of Higher Education, Manipal, India. The collected data were subjected to statistical analysis. The number of CFU was considered as the primary variable in the present study.

2.5. Statistical Analysis. Statistical analysis was done using SPSS software version 20. Mean \pm SD (standard deviation) was calculated for all variables. One-way ANOVA test was done to compare among the groups. The intergroup comparison was done using the post hoc Tukey test.

3. Results

The demographic and clinical parameters of the study participants are explained in Table 1. No significant differences were found among groups for any of the demographic variables. Comparison of three mouthwashes in reducing CFU revealed that there is a sharp decrease in the number of colonies after rinsing with CHX mouthwash with a p value of <0.001 (Figure 2 and Table 2).

TABLE 1: Demographic and clinical parameters among the three groups.

Details	Group A	Group B	Group C	p value
Age	42.43 \pm 6.3	43.73 \pm 5.5	44.00 \pm 6.0	NS
Gender (M/F)	5/5	5/5	5/5	NS
GI	1.89 \pm 0.6	1.85 \pm 0.5	1.87 \pm 0.6	NS
PI	2.09 \pm 0.46	2.19 \pm 0.38	2.08 \pm 0.39	NS
PPD	5.78 \pm 0.78	5.72 \pm 0.67	5.74 \pm 0.68	NS
CAL	6.16 \pm 0.62	6.19 \pm 0.63	6.2 \pm 0.62	NS

M: male; F: female; NS: nonsignificant; GI: gingival index; PI: plaque index; PPD: probing pocket depth; CAL: clinical attachment level.

There was a decrease in the number of colonies after rinsing with herbal mouthwash but not as significant as CHX (p value = 0.002). There was no decrease in the number of colonies after rinsing with water (Figure 2). The number of CFU was more in patients' chest area followed by operators' chest area and assistants' chest area. The CFU was maximum with a preprocedural rinse with water and minimum with CHX mouthwash and herbal mouthwash (Figure 3). Though there was a difference in the number of CFU between herbal and CHX mouthwashes, it was not statistically significant (Table 2 and Figure 1).

4. Discussion

Aerosol dissemination leads to the transmission of infectious agents and potentially harmful substances in the dental clinic environment. Effective infection control is key for the safe and successful dental practice. In the dental treatment room, the microbial aerosol peak concentration was associated with scaling procedures (47%) followed by cavity preparation (11%) [2, 9]. Both the high-speed air rotor and the ultrasonic scaler, which works with water, have the propensity of generating numerous airborne particles, which in turn have detrimental effects on both the clinician and the

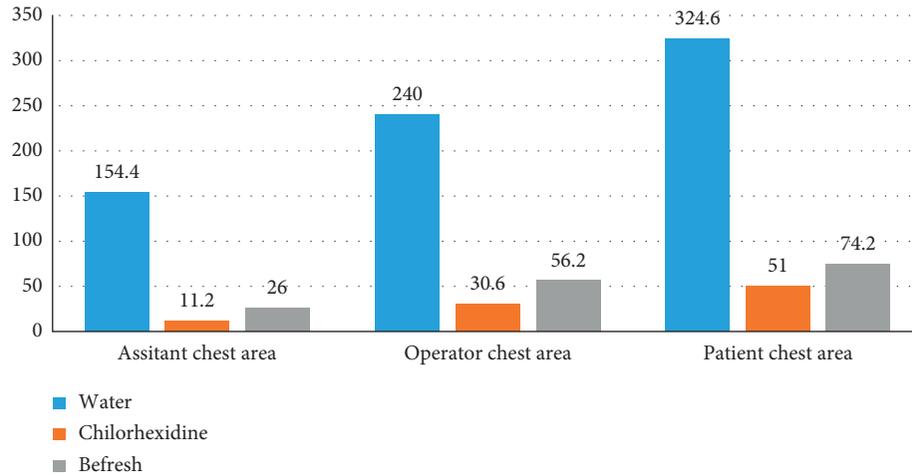


FIGURE 2: CFU (Colony Forming Unit) among three groups in different locations during the ultrasonic scaling procedure.

TABLE 2: Mean CFU/plate according to treatment and locations during the procedure.

Location	Groups	Mean \pm SD	<i>p</i> value
Assistant chest area	Group A	11.20 \pm 3.96	<0.001
	Group B	26.00 \pm 7.17*	
	Group C	154.40 \pm 28.08 [†]	
Operator chest area	Group A	30.60 \pm 3.97	<0.001
	Group B	56.20 \pm 20.13*	
	Group C	240.00 \pm 29.36 [†]	
Patient chest area	Group A	51.00 \pm 4.52	<0.001
	Group B	74.20 \pm 14.80*	
	Group C	324.60 \pm 36.43 [†]	

*Nonsignificant between groups A and B; [†]significant between groups B and C; [†]significant between groups A and C.

patient [10, 11]. 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 [11].

Dental plaque, the primary etiologic agent of diseases of the periodontium, comprises of both bacteria and viruses in a gelatinous matrix. Disruption of plaque biofilm occurs with the pressure of food, the friction of soft tissues, or by ultrasonic scalers [12]. Aerosols from ultrasonic devices are highly contaminated by bacteria and viruses leading to transmission of various other diseases. It could also result in contamination of other instruments which could be a source of infection to other patients [6]. Various preprocedural mouth rinses have been tried out for a reduction in bacteria load aerosol production. However, CHX has always been the best choice in reducing aerosol contaminants [2]. Advances in the dental field have seen an overwhelming use of herbal products in recent times. Plant products act as antimicrobial agents and have been considered as an alternative to antimicrobials. Thus, the present study was planned to compare the efficacy of herbal mouthwash in comparison with 0.2% CHX mouthwash and water as a preprocedural rinse prior to ultrasonic scaling.

The results of the present study showed that there was a highly significant reduction of bacterial CFU in both group A (0.2% CHX) and group B (herbal mouthwash); however, there were no significant results in the reduction of bacterial CFU in group C (water), which was in accordance with the study by Ammu A. et al. (2019) [18]. In the present study, the CFU was maximum in the patient chest area followed by operator chest area (Table 2 and Figure 2). This finding is in accordance with Rani et al. (2014) [4] whereas contrary to the finding of Purohit et al. (2009) [15]. The use of CHX or essential oil-containing mouthwash for one minute before a dental procedure has been shown to significantly lower the bacterial load and contamination of the operative area and staff. CHX is an effective antiseptic agent for planktonic microbes in saliva and mucous membranes, but CHX is not free of its side effects [19].

In a study by Gupta et al. (2014) [20], the efficacy of CHX 0.2% mouthwash and herbal mouthwash was superior to the control group (water). They concluded that CHX is better than herbal mouthwash as a preprocedural rinse. The present study is in accordance with Gupta et al.'s study in terms of superior aerosol bacteria reduction with both CHX and herbal mouthwash. However, in contrast to the former study, there was no significant difference found between CHX and herbal mouthwash in the present study. This finding is similar to an earlier study by Rani et al. (2014) [4].

Cross-contamination of water lines of the dental chair can also occur from the contamination of aerosols. Narrow bore water lines, stagnation of water, and dental chair unit heating from over usage are considered as a source of contamination [21]. Hence, in this study, dental chair unit bottles were disinfected before the dental procedure. In addition, before the procedure, the flushing of water was done for 2 minutes with the usage of high-speed evacuators. This was done similar to previous studies [6, 22].

Preprocedural oral rinsing with an antiseptic mouthwash significantly reduces the bacteria in aerosol which is generated during dental procedures [13]. The risk of cross-contamination with infectious agents in the dental operatory can be reduced to a greater extent by following preprocedural rinse [13, 14].

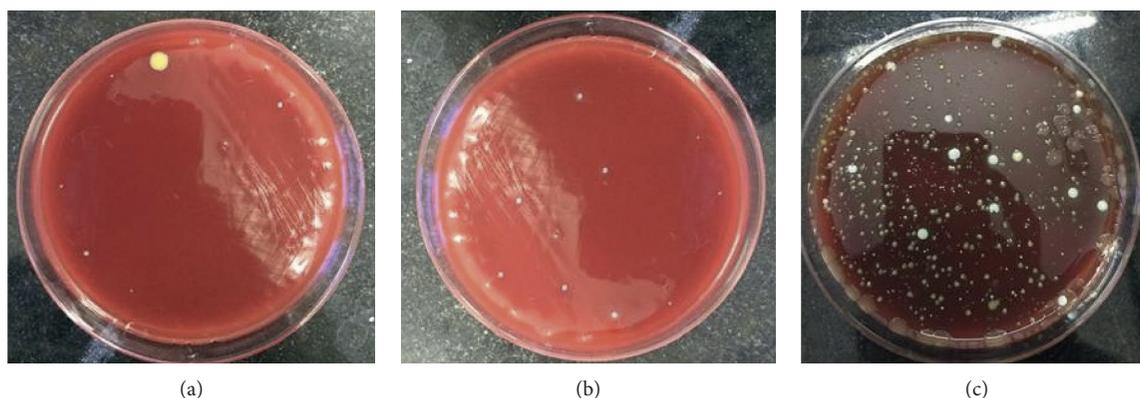


FIGURE 3: Blood agar plates: (a) group A; (b) group B; (c) group C.

5. Conclusion

Reduction in bacteria present in aerosol during ultrasonic scaling was found with the herbal rinse (Befresh™) and it was found to be as effective as CHX. Considering the fact that herbal rinses contain more natural ingredients and better accepted by patients, many more studies have to be conducted using these agents to prove their further beneficial effects. The CFU estimation in the present study includes only aerobic bacteria capable of growth on blood agar plates; anaerobic bacteria and viruses that require specialized media were not isolated, which needs to be addressed in further investigations.

Immunization of a dental team against biological hazards in their workplace through specific (vaccines) or nonspecific (e.g., gamma-globulin) immunization is mandatory. Total elimination of aerosol during ultrasonic scaling is not possible. Aerosol production during ultrasonic scaling is very dangerous to the patient, the operator, and the public at large. Therefore, along with other barrier techniques, preprocedural rinsing should be incorporated as a mandatory practice in all dental setups.

Data Availability

The data used to support the findings of this study are included in the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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