

**Comparative evaluation of chlorhexidine and green tea as ultrasonic coolant on dental aerosols**

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**Abstract**

**Introduction:** To avoid transmission of infection during scaling, various chemotherapeutic agents have been utilized as a coolant. The use of antimicrobials as a coolant serves the dual purpose of not only reducing the bacterial count in aerosols but also helps in continuous irrigation of the treatment site which further enhances the gingival health.

**Objectives**

1. To compare the potency of green tea and chlorhexidine gluconate on reducing dental aerosols.
2. Quantitative assessment of microbial content of dental aerosols at right and left the dental chair.

**Materials And Method:** In this study 30 subjects were selected who fulfilled the inclusion criteria and were divided into three groups. Group 1: Ultrasonic scaling with 0.12% chlorhexidine (10 subjects), Group 2: Ultrasonic scaling with distilled water (10 subjects). Group 3: Ultrasonic scaling with green tea (10 subjects). At the baseline one blood agar plate was kept for 10 minutes in the fumigated chamber before ultrasonic

scaling, thereafter two blood agar plates were kept at a distance of 0.4 meters away on either side of the patient during ultrasonic scaling. Blood agar plates were kept for gravitometric settling of dental aerosols.

**Results:** It is found that Group 1 (chlorhexidine gluconate) showed effective CFU reduction followed by green tea and distilled water. More CFU were found on blood agar plates which were kept on right side in all the three groups.

**Conclusion:** Chlorhexidine gluconate is more effective in reducing dental aerosols when compared to green tea and distilled water.

**Keywords:** Chlorhexidine gluconate, green tea, distilled water, aerosols, colony forming unit.

**Introduction**

Dental professionals are at a higher risk for the spread of infection through splatter and aerosol because of transmission of the infection from the patient to health care providers. Various dental equipments such as the dental handpieces, air–water syringes, ultrasonic scalers, and air polishing units are known to produce the aerosols

during the procedures, and the published data indicate that they produce many folds increase in colony forming units (CFUs) when compared to pre- and post-operatively.

Miller in a study, concluded that aerosols generated from the patients' mouth contain millions of bacteria per cubic foot of air. King et al. reported that bacteria could be recovered 6 inches from the mouth of patient and the CFUs formed were significantly reduced when aerosol reduction device was used. Transmission of infection through splatter and aerosol has been considered a major risk factor for the dental professionals because of spread of the infection from the patient to health care providers. Various dental equipments such as the dental handpieces, air-water syringes, ultrasonic scalers, and air polishing units are known to produce the aerosols during the procedures.<sup>1</sup>

Aerosols are the suspension of liquid or solid particles containing viruses and bacteria which are suspended in gas for few seconds. The size of the particle may vary from 0.001 mm to more than 100 µm. The smallest particle size (ranging between 0.5 µm and 10 µm) has the greatest potential to penetrate the respiratory passages and the lungs, possessing the ability to transmit the disease. These microorganisms get aerosolized when come in contact with the dental equipment. Miller in a study concluded that aerosols generated from the patients' mouth contain millions of bacteria per cubic foot of air.<sup>1</sup>

Various approaches have been utilized to minimize the cross-contamination due to microbes in a dental office. This includes use of layered approach, surface decontamination, personal protective barrier use, immunization of dental staff, and preprocedural mouthrinses.<sup>2</sup> To avoid contamination, various chemotherapeutic agents have been utilized as a coolant. The use of antimicrobials as a coolant serves the dual purpose of not only reducing the bacterial count in

aerosols but also helps in continuous irrigation of the treatment site which further enhances the gingival health.

Among various studies evaluating the effect of various agents as preprocedural mouthrinse or as an ultrasonic coolant have been conducted, chlorhexidine has emerged as a gold standard.<sup>2</sup> However the research evaluating the effect of herbal mouthrinse is limited.

Green tea is one of the herbal agents which are known for their antioxidant and antibacterial properties. It is made solely with the leaves of *C. sinensis* that have undergone minimal oxidation during processing. The most abundant components in green tea are polyphenols, in particular, flavonoids such as the catechins. Major catechins found in green tea are epicatechin gallate (ECG), epicatechin (EC), epigallocatechin (EGC), and EGC gallate (EGCG). It has been shown to possess antibacterial, antioxidant, anti-inflammatory, antidiabetic, antiviral, and antimutagenic properties. Various studies have reported green tea to be efficacious against caries and periodontal diseases.<sup>6</sup>

Therefore, the present study mainly focuses on comparing the effectiveness of green tea and chlorhexidine as an ultrasonic coolant in comparison with distilled water.

### **Materials and Methods**

This study was a placebo-controlled, randomized clinical trial with a three-group parallel design. The study was conducted over a period of 3 months, and participants enrolled were selected from the outpatient Department of Periodontology. Ethical clearance was obtained from the Institutional Ethical Committee. A written informed consent was signed by all the patients. Participants who met the inclusion criteria were informed about the purpose of the study and each patient was provided with an informed consent, after explaining the nature and possible risk. Criteria for participation included patients having a minimum of 20 permanent functional teeth. Subjects with

mean probing depth  $\leq$  5mm and clinical attachment loss  $\leq$  3mm measured with Williams Periodontal Probe (Hufriedy) in at least 30% teeth sites were included.

### **Sample size calculation**

The sample size was calculated for  $\alpha$  error fixed at  $<5\%$  ( $P < 0.005$ ). Based on this calculation, the minimum sample size required in each group was 10 participants. Participants were enrolled in three groups.

### **Selection criteria**

The inclusion criteria of this study were as follows: (1) Systemically healthy patients (2) Participants diagnosed with moderate-to-severe gingivitis having a gingival index (GI) score of 2–3, and (3) Participants indicated for full-mouth scaling in single sitting.

The exclusion criteria of this study were as follows: (i) Systemic diseases like diabetes mellitus, heart diseases, rheumatoid arthritis, hepatitis and other systemic diseases that can alter the course of periodontal disease.

(ii) Diseases of oral hard and soft tissue except caries and periodontitis.

(iii) Use of tobacco in any form

(iv) Subjects on any medication taken within the last 6 months which may alter the periodontal status.

(v) Pregnant and lactating mothers.

Oral examination was carried out by measuring clinical parameters such as Probing Pocket Depth (PPD), Clinical Attachment Loss (CAL), Plaque Index (PI) and Gingival Index (GI) in patients with chronic periodontitis. All subjects were assigned to one of the three groups by using randomization table and consisted of 10 subjects in each group depending on different ultrasonic liquid coolants.

- Group I {test group}: 10 patients treated with ultrasonic scaling with 0.12% chlorhexidine in 0.06% dilution; 0.12% chlorhexidine gluconate is diluted in 1:1 ratio in 1 litre water to prepare ultrasonic liquid coolant.

- Group II {control group}: 10 patients treated with ultrasonic scaling with distilled water as a coolant.
- Group III {test group}: 10 patients treated ultrasonic scaling with green tea preparation [0.5% aqueous solution of green tea] diluted in 1:1 ratio in 1 litre water to prepare ultrasonic liquid coolant.

### **Study Design**

All treatment procedures were conducted in a closed operatory. It was fumigated for 48 hours before the procedure to prevent contamination.

At the baseline, one blood agar plate was kept for 10 minutes in the closed chamber before ultrasonic scaling. Patient was made comfortable in dental chair. Two blood agar plates were kept at a distance of 0.4 meters away on either side of the patient during ultrasonic scaling. The patients ultrasonic scaling was executed for 20 mins. The normal rate of flow of water in ultrasonic scaler is 20–30ml/min. The same rate of flow of water for each agent, while performing ultrasonic scaling was maintained. To assure that the room was free from aerosols appointments were scheduled in the morning around 10 am. For every scaling procedure, high vacuum suction was used. After the treatment, two coded blood agar plates were left uncovered for 20 min at the pre-designated sites for gravitometric settling of airborne bacteria. After gravitometric settling of aerosols, blood agar plates were transferred to laboratory for incubation at 37°C for 48 hours followed by colony counting procedure with the help of colony counter device by the microbiologist.

### **Statistical Analysis**

The Analysis of variance (ANOVA) test was used for continuous variables after confirming normality of the data distribution. Intergroup analysis of the CFU counts at right and left sides and the clinical parameters (GI and PI) were performed using Kruskal Wallis ANOVA whereas

intragroup analysis was done using Mann-Whitney U test. The statistical significance was defined as  $P < 0.05$ .

**Results**

This randomized, placebo-controlled, clinical trial was conducted over a period of 3 months between September 2019 to November 2019. This study included 30 patients and were randomly divided into chlorhexidine groups, distilled water and green tea; each group consisted of 10

subjects. The distribution of male and female participants according to different experimental group is shown in table 1. A total of 58.33% males and 41.67% females participated in the study. The mean  $\pm$  SD age of the patients included in the study was  $21.3 \pm 1.83$  years (table 2). There was no significant difference within the groups with respect to demographic characteristics ( $P > 0.05$ ).

Table 1 : Distribution of male and female in three study groups (1, 2, 3)

Sex	Group 1	%	Group 2	%	Group 3	%	Total
Male	5	50.00	5	50.00	6	60.00	16
Female	5	50.00	5	50.00	4	40.00	14
Total	10	100.00	10	100.00	10	100.00	30
Chi-square=0.0000 p=1.0000							

$P > 0.05$  considered statistically nonsignificant.

Table 2: Comparison of three study groups (1, 2, 3) with mean age by one way ANOVA

Groups	Means	SD	SE
Group 1	21.30	1.83	0.58
Group 2	21.80	2.39	0.76
Group 3	21.10	1.85	0.59
Mean age	0.3117		
SD age	0.7348		

Table 3 and 4 shows the intergroup and intragroup comparison of clinical parameters at baseline and after 3-months follow-up. At baseline, there were no difference statistically with regard to both GI and PI in all the three experimental groups. GI scores of Group I, Group II, and Group III were  $1.62 \pm 0.21$ ,  $1.51 \pm 0.21$ ,  $1.54 \pm 0.26$  respectively, which was statistically nonsignificant. After 3-months follow-up, these scores were reduced to  $1.00 \pm 0.09$ ,  $1.02 \pm 0.06$ ,  $0.97 \pm 0.26$  in Group I, Group II, and Group III, respectively. This reduction in GI scores was

statistically significant. At baseline, the PI scores of the participants in Group I, Group II, and Group III were  $1.79 \pm 0.22$ ,  $1.60 \pm 0.28$  and  $1.50 \pm 0.37$  respectively, which was statistically nonsignificant ( $P = 0.11$ ). These plaque scores were reduced to  $0.96 \pm 0.16$ ,  $0.99 \pm 0.14$  and  $0.90 \pm 0.23$  in Group I, Group II, and Group III, respectively, which was statistically significant ( $P < 0.05$ ). Intragroup analysis of both the clinical parameters (GI and PI) after 3-months follow-up is shown in table 3 and 4.

Table 3: Comparison of three groups (1, 2, 3) with respect to baseline and 3 months GI scores by Kruskal Wallis ANOVA

Groups	Baseline			3 months			Changes		
	Mean	SD	Mean rank	Mean	SD	Mean rank	Mean	SD	Mean rank
Group 1	1.62	0.21	17.85	1.00	0.09	15.15	0.62	0.19	17.80
Group 2	1.51	0.21	13.55	1.02	0.06	16.35	0.49	0.21	12.80
Group 3	1.54	0.26	15.10	0.97	0.19	15.00	0.57	0.21	15.90
% of change in Group 1							38.27%#, P=0.0050*		
% of change in Group 2							32.45%#, P=0.0051*		
% of change in Group 3							37.01%#, P=0.0050*		
H-value	1.2480			0.2160			1.7030		
P-value	0.5360			0.8980			0.4270		
Pair wise comparisons by Mann-Whitney U test									
Group 1 vs Group 2	p=0.2730			p=0.7337			p=0.2123		
Group 1 vs Group 3	p=0.4963			p=0.9397			p=0.6232		
Group 2 vs Group 3	p=0.7055			p=0.7624			p=0.4274		

\*p<0.05 indicates significant, SD- Standard deviation

Table 4 : Comparison of three groups (1, 2, 3) with respect to baseline and 3 months PI scores by Kruskal Wallis ANOVA

Groups	Baseline			3 months			Changes		
	Mean	SD	Mean rank	Mean	SD	Mean rank	Mean	SD	Mean rank
Group 1	1.79	0.22	20.05	0.96	0.16	15.25	0.83	0.16	20.9
Group 2	1.60	0.28	14.15	0.99	0.14	17.1	0.61	0.32	13
Group 3	1.50	0.37	12.3	0.90	0.23	14.15	0.60	0.23	12.6
% of change in Group 1							46.37%#, P=0.0050*		
% of change in Group 2							38.13%#, P=0.0052*		
% of change in Group 3							40.00%#, P=0.0050*		

Group 3			
H-value	4.3330	0.6300	5.7590
P-value	0.1150	0.7300	0.0560
Pair wise comparisons by Mann-Whitney U test			
Group 1 vs Group 2	p=0.1041	p=0.6232	p=0.0640
Group 1 vs Group 3	p=0.0697	p=0.7624	p=0.0258*
Group 2 vs Group 3	p=0.5454	p=0.4727	p=0.9699

\*p<0.05 indicates significant

Figure 1 and 2 show the graph where illustration of mean  $\pm$  SD of the CFUs formed is summarized. Table 5 shows the mean  $\pm$  SD scores of CFUs of all the three groups. In Group I, mean  $\pm$  SD scores of CFUs formed at the right side, and left side of the patients were  $560 \pm 485.80$  and  $280 \pm 379.47$ , respectively. In Group II, mean  $\pm$  SD scores of CFUs formed at the right side, and left side of the

patients were  $7300 \pm 4347.41$  and  $4600 \pm 4647.58$ , respectively. In Group III, mean  $\pm$  SD scores of CFUs formed at the right side, and left side of the patients were  $1900 \pm 2846.05$  and  $1360 \pm 3065.29$  (mean  $\pm$  SD), respectively [Table 4]. Also, table 4 shows the pairwise analysis of the CFUs formed at two standardized locations.

Table 5: Comparison of three groups (1, 2, 3) with respect to CFU counts at right and left sides by Kruskal Wallis ANOVA

Groups	Right side			Left side		
	Mean	SD	Mean rank	Mean	SD	Mean rank
Group 1	560.00	485.80	9.10	280.00	379.47	10.00
Group 2	7300.00	4347.41	22.60	4600.00	4647.58	23.20
Group 3	1900.00	2846.05	14.80	1360.00	3065.29	13.30
H-value	14.3570			14.4050		
P-value	0.0010*			0.0010*		
Pair wise comparisons by Mann-Whitney U test						
Group 1 vs Group 2	p=0.0019*			p=0.0009*		
Group 1 vs Group 3	p=0.0821			p=0.4057		
Group 2 vs Group 3	p=0.0233*			p=0.0126*		

\*p<0.05 indicates significant at 5% level

Figure 1: Intergroup comparison with respect to CFU counts at right and left sides

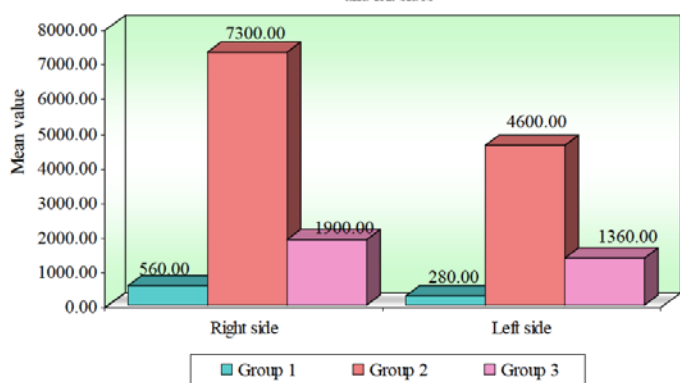
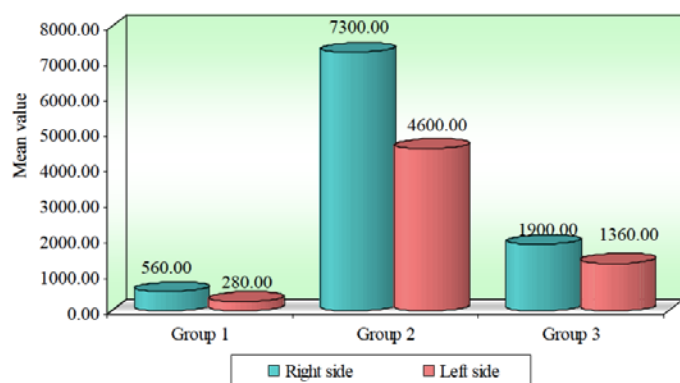


Figure 2: Intra group comparison with respect to CFU at right and left sides



## Discussion

The oral cavity offers an optimal habitat for millions of bacteria and viruses from the respiratory tract, saliva, and dental plaque. These microorganisms get aerosolized during the usage of ultrasonic scaler or air rotor of dental chair and are potentially capable of spreading infection in the dental office as well as operator and the assistant. Miller, in a study concluded that aerosols generated from the patients' mouth contain millions of bacteria per cubic foot of air. Thus, the present study was conducted evaluating the effect of chlorhexidine and green tea as an ultrasonic coolant as compared to distilled water on the reduction of microbial load in dental aerosols produced. In addition, their effect on the gingival and plaque status was also analyzed.

Various studies have reported that antimicrobial solutions when used as pre-procedural rinses can lead to decrease in the number of microorganisms aerosolized during clinical practice. Veksler et al., have evaluated that pre-procedural rinsing using 0.12% Chlorhexidine (CHX) gluconate reduces the magnitude of aerobic and facultative flora of oral cavity. However, the rinsing period for the preprocedural mouth rinsing varies between 30–60 s according to various studies.

On the other hand, the use of antimicrobial agents as an ultrasonic coolant provides continuous action of the agents over a longer period, thus bypassing the rinsing period as that of preprocedural mouth rinses. Also, the depth of penetration of the ultrasonic coolant is more and prolonged when compared to that of preprocedural mouth rinses, Apart from these reasons, the patient's compliance and subjective error in rinsing was also the reason why ultrasonic antimicrobial coolants were chosen in the current study. In the present study, chlorhexidine 0.12% and 0.5% aqueous solution of green tea were used. B. Meena Priya et al in 2019 compared the efficacy of the mouthwash containing green tea and chlorhexidine in the management of dental plaque-induced gingivitis and concluded that the green tea-containing mouthwash is equally effective in reducing the gingival inflammation and plaque to chlorhexidine. Based on this body of evidence, these two mouth rinses were used as ultrasonic coolant in the current study.

Chlorhexidine has antimicrobial property which is attributed to its action on the inner cytoplasmic membrane. Due to its broad spectrum antimicrobial activity and good substantivity, it is recommended as a gold standard for plaque control.

Green tea has been reported to be useful for the prevention of periodontal disease and maintenance of oral health. Various authors have reported the inhibitory effects of

catechin contained in green tea on periodontal pathogens, which provides the basis for beneficial effect of daily intake of green tea on periodontal health. Catechin present in green tea was found to have antiplaque and antibacterial properties and contributed in caries prevention and in gingival enlargement. Green tea is also a powerful antioxidant and has anti-inflammatory properties. Antioxidants play an important role in the control of gingival inflammation by inhibiting the oxidative stress. Green tea catechin inhibits the growth of *P. gingivalis*, *Prevotella intermedia* and *Prevotella nigrescens* and adherence of *P. gingivalis* on to human buccal epithelial cells. Green tea catechins with steric structures of 3-galloyl radical, EGCG, ECG and gallocatechin gallate, which are major tea polyphenols, inhibit production of toxic end metabolites of *P. gingivalis*. Furthermore, a study showed that green tea catechin, EGCG and ECG inhibit the activity of *P. gingivalis*-derived collagenase. Also, in other study green tea catechin showed a bactericidal effect against black pigmented, Gram-negative anaerobic rods, *Porphyromonas gingivalis* and *Prevotella* species, and the combined use of mechanical treatment and the application of green tea catechin using a slow-release local delivery system was effective in improving the periodontal status.

Jenabian et al. observed a significant decrease in both PI and BI in chronic generalized plaque-induced gingivitis patients receiving green tea or placebo. Based on these facts, we have designed our study in which chlorhexidine and green tea were used as ultrasonic liquid coolant and not as pre-procedural rinse.

King et al. reported that bacteria could be recovered 6 inches from the mouth of patient and the CFUs formed were significantly reduced when aerosol reduction device was used.

Overall, the results of the present study demonstrate that Chlorhexidine gluconate showed better CFU reductions when compared with green tea. Green tea also showed better CFU reductions when compared with distilled water. To the best of our knowledge, this is the first study where green tea is being used as an ultrasonic coolant and is compared with that of chlorhexidine. Thus, more randomized controlled clinical trials with larger sample size need to be conducted to validate these findings.

### **Conclusion**

Within the limitations of this study, this study concludes that chlorhexidine gluconate as an ultrasonic liquid coolant significantly reduces the microbial content of dental aerosols generated during scaling when compared with distilled water. Chlorhexidine gluconate showed better CFU reductions when compared with green tea. Green tea also showed better CFU reduction when compared with distilled water. Hence, green tea can also be used as an ultrasonic liquid coolant for reducing the number of dental aerosols during ultrasonic scaling. Moreover, green tea can also be promoted to be used as a mouthwash as it has no side effects.

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**Abbreviations:** Colony forming units (CFUs), Epicatechin gallate (ECG), Epicatechin (EC), Epigallocatechin (EGC), and EGC gallate (EGCG), Gingival index (GI), Probing Pocket Depth (PPD), Clinical Attachment Loss (CAL), Plaque Index (PI), Analysis of variance (ANOVA), Standard deviation (SD).