

## Comparative evaluation of the antimicrobial efficacy of herbal mouthwash versus chlorhexidine mouthwash

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### Abstract

**Context:** Chlorhexidine is considered as the "gold standard" anti-plaque agent over last three to four decades. Due to disadvantages of chlorhexidine, now a days an alternative is advisable, which is free from all the disadvantages of chlorhexidine.

**Aim and Objective:** The study aims at finding herbal alternatives to chemical mouthwashes to avoid harmful side-effects and improve patient compliance and over all oral health.

**Materials and Methods:** 160 dental students, 18 – 25 years of age were selected for this study from Santosh Dental College and Hospital, Ghaziabad. The study were divided randomly into two groups. Group -1 was advised to rinse with herbal mouthwash. Group-2 was advised to rinse with 0.12% chlorhexidine mouthwash.

**Result:** Lesser side-effects were observed with herbal mouth-wash as comparison with Chlorhexidine mouth-wash and also these two mouthwashes shown equal effectiveness.

**Conclusion:** The present study summarized that Herbal mouthwashes have proved to be excellent antiplaque agents. In the present study, it is concluded that the herbal mouthwashes are equally good antimicrobial agents compared to CHX mouthwash with lesser side-effects and equally effective as well.

**Keywords:** Dental plaque, chlorhexidine, oral rinse, herbal, mouthwash

### Introduction

Mouthwashes are aqueous solutions which are widely used as adjuncts to oral hygiene and in the delivery of active agents to the teeth and gums.<sup>(1)</sup> The main purpose of using mouthwashes is that they can be used at home as an effective home care system by the patient to enhance oral hygiene.<sup>(2)</sup> Mouthwashes have the ability to influence plaque formation and thus alter the course of gingival inflammation. Since 5-6 decades, Chlorhexidine has been used as a broad spectrum antiseptic Chlorhexidine is a gold standard against which other antiplaque and anti-gingivitis agents are measured.

In spite of its extreme utility, chlorhexidine is not routinely used due to its numerous side effects including brown discoloration of the teeth, restorative materials, dorsum of tongue and taste perturbation also. The most frequently reported oral mucosal symptoms associated with Chlorhexidine gluconate oral rinse are stomatitis, gingivitis, glossitis, ulcer, dry mouth, hyperesthesia, glossal oedema, and paraesthesia. Minor irritation and superficial desquamation of the oral mucosa have also been noted.<sup>(3)</sup>

Considering these drawbacks of CHX mouthwash, there is a need to explore natural and herbal products, which can be used to prepare mouthwashes with the pharmacotherapeutic

effects, same as chlorhexidine but with lesser side-effects.<sup>(4)</sup> So, alternative antiplaque agents have been developed.

Keeping the above idea in mind the present study was planned to assess the use of herbal products like Salvodora Persica, Belleric Myrobalan Bibhitaki and Betel Nagavalli as antimicrobial agents and to compare their efficacy and side-effects with 0.12% Chlorhexidine.

### Materials and methods

The present study was a clinical comparative study, which conducted over a period from Jan 2020 to Dec 2020. Saliva samples were collected for the study, from healthy Individuals, who were the students of Santosh Dental College, Ghaziabad. The sample size comprised of 160 healthy Individuals, between 18 – 25 years of age for this study. Individuals having gingival index of score 2 ( Loe & Silness, 1963 ) and plaque index of score 2 (Silness & Loe, 1964) were included in this study, who had moderate inflammation, mild erythematous gingiva and gingival bleeding on probing and had moderate layer of plaque present along the gingival margin . Individuals who had non-plaque induced gingivitis or periodontitis, who were on antibiotic therapy / steroid therapy or had uncontrolled systemic disorders were excluded from the study.

The sample was divided randomly into two groups. Group-1 was advised to rinse with chlorhexidine mouthwash for 14 days twice daily. Group-2 was advised to rinse with commercially available herbal mouthwash for 14 days twice

daily. Mouthwash and their ingredients are discussed in Table-1. For this study blood agar containing agar plates (fig 1) and Microbial colony counter (fig-2) were used.

**Table.1 : Mouthwashes, their active ingredients and mechanism of action<sup>(5)</sup>**

Mouthwash	Active Ingredients	Mechanism of action
Hexidine mouthwash	0.12% Chlorhexidine Gluconate	Chlorhexidine has an affinity for bacteria probably because of an interaction between the positively charged chlorhexidine molecule and negatively charged groups on the bacterial cell wall (e.g. phosphate groups). This interaction increases the permeability of the bacterial cell wall <sup>(6)</sup> and thus permits the agent to penetrate into the cytoplasm and cause the death of the microorganism <sup>(6)</sup> .
Herbal mouthwash	Miswak ( <i>Salvadora persica</i> ), Bibhutaki ( <i>Terminalia bellerica</i> ), Gandhapura Taila, Nagavalli ( <i>Piper Betle</i> )	Herbal mouthwash has antiplaque, analgesic, Antimicrobial, antiseptic <sup>(4)</sup> , and refreshing properties. It has active herbal ingredients that act against common strains of oral bacteria and fungi and prevent gum and tooth disease. It helps in the prevention and treatment of gum disease. Silica in Miswak acts as an abrasive material to remove stains <sup>7</sup> giving the teeth whiteness. Tannins also inhibit the action of glucosyl transferase thus reducing plaque and gingivitis. The alkaloid present in <i>Salvadora persica</i> is Salvadorine, which yields trimethylamine on hydrolytical cleavage. It exerts a bacteriocidal effect and stimulatory action on the gingiva. The mild bitter taste stimulates the flow of saliva, which is antiseptic. The sulfur compounds present in Miswak as shown by their pungent taste and smell have a bacteriocidal effect.

Saliva samples were collected before the use of mouthwash on the day 0 (Base line saliva sample). -shown in Fig-3 and fig 5. After 14 days, again saliva sample was collected. Counting of bacterial colonies were done from salivary samples before and after using the mouth wash. Each saliva sample was diluted with distilled water in a ratio by (1:1000). These samples were then inoculated on agar plates. Colonies of

bacteria were developed in a 1-2 days, depicted by fig 5 and fig-6 respectively of herbal mouthwash and chlorhexidine mouthwash, which then counted under microscope with the help of colony-counter to compare and determine the effectiveness of these two mouthwashes.

Also participant's compliance were noted by a questionnaire filled by them



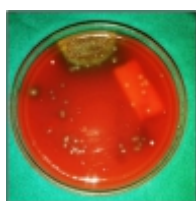
**Figure 1: Blood Agar Containing Gel Plate**



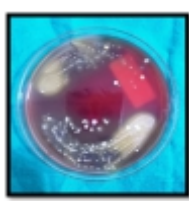
**Figure 2: Microbial Colony Counter**



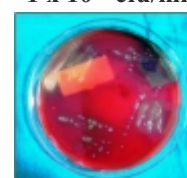
**Figure 3: Base Line Saliva Sample Microbial colony count  $1 \times 10^6$  cfu/ml**



**Figure 4: After Using Herbal Mouthwash Microbial Colony Count  $1.5 \times 10^3$  CfU/ml**



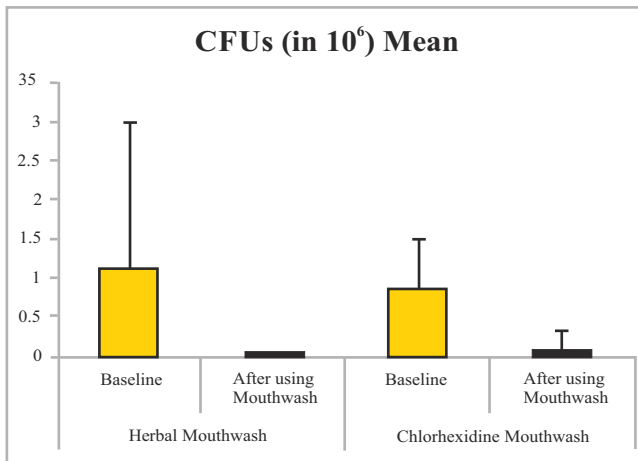
**Figure 5: Base Line Saliva Sample Microbial Colony Count  $1.8 \times 10^6$  CfU/ml**



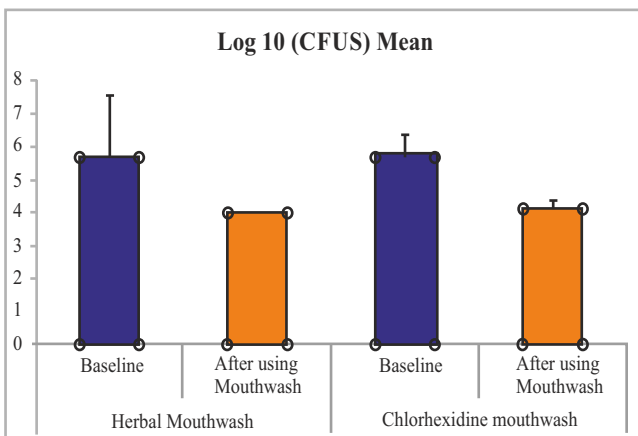
**Figure 6: After Using Chx Mouth Wash Microbial Colony Count  $1 \times 10^4$  CfU/ml**

**Results**

**Graph-1 shows** Mean distribution of CFUs of bacterial growth among herbal mouthwash and chlorhexidine mouthwash using individual subjects at before and after its usage showed bacterial colony counts of both mouthwashes were almost similar



**(Graph-1) Mean distribution of CFUs of bacterial growth**



**Graph-2 : Mean distribution of of CFUs of bacterial growth**

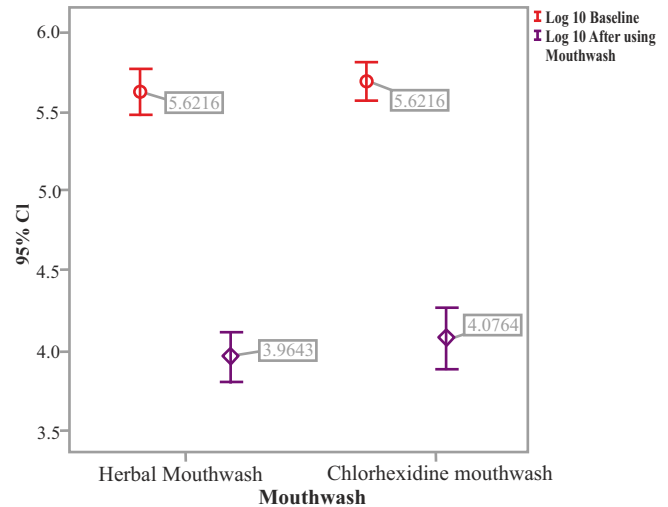
**Graph-2 shows** Mean distribution of logarithmic transformation of CFUs of bacterial growth among herbal mouthwash and chlorhexidine mouthwash using individual subjects at before and after its usage also showed almost equal results

**Independent Samples t Test**

The Independent Samples t Test is a parametric test that compares the means of two independent groups in order to determine whether there is statistical evidence that the associated population means are significantly different.

In the present study, Independent sample t test displays no statistically significant difference in logarithmic transformed mean CFUs of bacterial growth among herbal mouthwash and chlorhexidine mouthwash using individual subjects at

before and after its usage ( $P>0.05$ ) This indicates, herbal and chlorhexidine mouthwash are equally efficient in reduction of bacteria



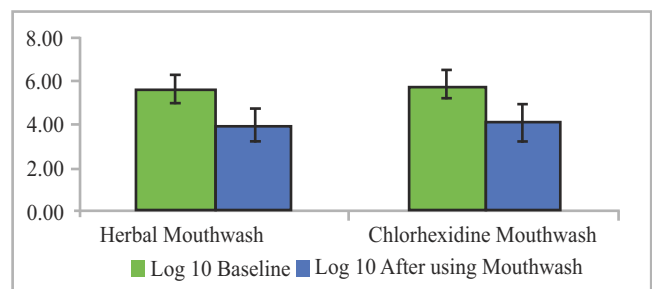
**Graph 3 shows** Distribution of logarithmic transformed mean CFUs of bacterial growth among herbal mouthwash and chlorhexidine mouthwash using individual subjects at before and after its usage. (Error Bar Chart).

**Table 2:** Pairwise comparison of means of logarithmic baseline with logarithmic after use of Herbal and Chlorhexidine mouthwash

		N	Mean	SD	Mean Difference	t value	P value
Herbal Mouthwash	Log10 Baseline	80	5.62	0.65	1.65	19.438	0.001*
	Log 10 After using Mouthwash	80	3.96	0.73			
Chlorhexidine Mouthwash	Log10 Baseline	80	5.69	0.53	1.61	16.77	0.001*

\*Statistical significance set at 0.05; N: Number of samples; SD: Standard deviation

**Graph 4:** Pairwise comparison of means of logarithmic baseline with logarithmic after use of Herbal and Chlorhexidine mouthwash



**Table 2 & Graph 4:** The paired sample t-test, sometimes called the dependent sample t-test, is a statistical procedure used to determine whether the mean difference between two sets of observations is zero or to find out the mean difference between to related continuous variable at two different intervals of time. In the present research, paired sample t-test was performed for the comparison of Means of logarithmic baseline with logarithmic after use of Herbal and Chlorhexidine mouthwash.

A statistically significant reduction of CFUs after using herbal mouthwash ( $3.96 \pm 0.73$ ) when compared to baseline ( $5.62 \pm 0.65$ ) bacterial CFUs (**t value = 19.438; P=0.001**). Similarly, after use of chlorhexidine mouthwash ( $4.08 \pm 0.86$ ) exhibited statistically significant lower bacterial CFUs when compared to the baseline ( $5.69 \pm 0.53$ ) bacterial CFUs (**t value = 16.77; P=0.001**)

### Discussion

The result of present study showed that both the mouthwashes, herbal and chlorhexidine were equally effective in terms of antimicrobial properties. Both mouthwashes were equally effective in reducing plaque and gingivitis. In this study patient compliance was noted to be much better in Herbal mouthwash as compared to chlorhexidine mouthwash as there were no side-effects.

In spite of its promising results, the present study also showed some limitations. The study was conducted on a definite age group, and also selection criteria was also defined by some pre-determined qualities. So, it was better if a broader age group was taken with no definitive qualities for wider spectrum. Though the present findings do not contraindicate, the use of chlorhexidine mouth rinses.

More advance studies are required on a broader age groups to determine the effectiveness of these two mouthwashes and compare their disadvantages, if any.

### Conclusion

The present study summarized that, Herbal mouthwashes have proved to be an excellent antiplaque agents. In the present study, it is concluded that the herbal mouthwashes are equally good antimicrobial agents compared to CHX mouthwash. These are made from popular herbs like Miswak (*Salvadora persica*) Bibhitaki (*Terminalia bellerica*) Nagavalli (*Piper betle*) and are very effective in inhibiting the

microbial growth without any noticeable side effects. However long term studies would provide better analysis the efficacy and side effects of the herbal mouthwashes. So, more clinical trials need to be carried out with broader age group and longer duration for better assessment.

### Declaration of conflicting interests

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