

Commercially available chemical and herbal mouthwash and its efficacy against oral microorganisms

Abstract

Aim: The present study was done to determine the activity of Naturally prepared licorice mouthwash in comparison to chlorhexidine mouthwash.

Introduction: Maintenance of oral hygiene is very important in preventing the growth of a sticky film of bacteria and food particles that accumulates on teeth and hence Mouthwashes are prescribed in dentistry for treatment of several oral conditions and for prevention against various microorganisms. *Licorice* is derived from "liquiritiae" which is shrub or herb and the yellow colour of it is due to the flavonoids content of the plant, which includes liquiritin, isoliquiritin and other compounds. Herbal Mouthwashes (mouth rinses) are solutions or liquids intended to reduce the microbial load in the oral cavity.

Materials and methods: This study was conducted in Saveetha Dental Hospital, Chennai, Tamilnadu, India. The liquorice was extracted and formulated as mouth wash. **Figure 1** denotes the Saliva samples were obtained from 20 volunteers (10 test group, 10 control group) and then, the participants were asked to rinse their mouth with freshly prepared licorice mouthwash (test group), chlorhexidine mouthwash (control group) and then paired saliva samples were obtained immediately after and before the mouthwash and finally the Colony forming units were recorded for pre and post usage of both the herbal and commercially available mouthwash.

Results: Study showed that there was a mean reduction in the colony-forming units with herbal mouthwash and Chlorhexidine mouthwashes. Herbal mouthwash showed reduction in colony forming growth after usage of herbal mouthwash.

Keywords: *Glycyrrhiza glabra*; Chlorhexidine; Dental caries; *licorice* mouthwash, innovative technique, energy green synthesis.

Running title: Effectiveness of chemical and herbal mouthwash against oral microorganisms.

INTRODUCTION

Mouthwashes are prescribed in dentistry for treatment of several oral conditions and for prevention against various microorganisms(1). Maintenance of oral hygiene is very important in preventing the growth of sticky film of bacteria and food particles that accumulates on teeth. Oral hygiene measures include mechanical aids like toothbrushes, floss, interdental cleansers and chemotherapeutic agents like mouthwashes, dentifrices and chewing gums. Mouthwashes (mouth rinses) are solutions or liquids intended to reduce the microbial load in the oral cavity.(2,3) *Glycyrrhiza glabra*: The genus includes nearly 20 species native to North , South America, Australia, Europe, Asia etc. The English name *licorice* is derived from "liquiritiae" which is shrub or herb and the yellow colour of it is due to the flavonoids content of the plant, which includes liquiritin, isoliquiritin and other compounds. *Glycyrrhiza glabra* is a commonly used herb in Ayurvedic medicine. Various Studies indicate that *Glycyrrhiza glabra* Linn possesses vast properties like antimalarial, antispasmodic ,antibacterial, antioxidant, anti-inflammatory and anti hyperglycemic properties. Various others properties includes antiulcer, antiviral, antihepatotoxic, antifungal and herpes simplex have also been studied.(4,5)

Chlorhexidine is regarded as the "gold standard" anti-plaque agent(6) However, it is not a "Magic Bullet" due to certain side effects like tooth staining, taste disturbance, etc(1,6,7) Most of the mouthwashes available in the market contain alcohol and other chemicals such as chlorhexidine gluconate and triclosan. These chemicals cause various side effects ranging from taste disturbance to allergic contact stomatitis. To overcome such side effects, nontoxic herbal mouthwashes using various herbs and plant extracts are introduced. Natural herbs like *Glycyrrhiza glabra*, *triphala*, *tulsi patra*, *jyeshthamadh*, *neem*, *clove oil*, *pudina*, *ajwain*, and lots of more used either alone or together are scientifically proven to be safe and effective against various oral health problems like bleeding gums, halitosis, mouth ulcers, and preventing cavity .

The major strength of those natural herbs is the absence of any side effects and they do not contain alcohol and sugar found in their counter products. The microorganisms prey on these ingredients, releasing by-products and causing halitosis. Thus, herbal mouth rinses promote better oral hygiene and health(7) The present study compared the effectiveness of an herbal

mouthwash with different brands of commercially available mouthwashes in reducing the oral bacterial count.

MATERIALS AND METHODS

This study was conducted in Saveetha Dental Hospital, Chennai, Tamilnadu, India and 28g of freshly prepared *Glycyrrhiza glabra* (*athimathuram*) and was mixed with 450ml of distilled water and were boiled for 10 minutes. Then the solution was strained and the strained liquid was stored in a sterilised vessel.(8)

TEST GROUP:

Saliva samples were obtained from 10 volunteers and then, the participants were asked to rinse their mouth with freshly prepared *licorice* mouthwash and then two saliva samples were obtained immediately after and before the herbal mouthwash.

CONTROL GROUP:

Saliva samples were obtained from volunteers and then the participants were asked to rinse their mouth with chlorhexidine, the saliva samples were collected immediately after and before the mouthwash. After collecting saliva samples, the samples were sent to the microbiology laboratory immediately. **Figure 1** denotes the collected samples before and after using mouthwash and saline was added in the test tubes and culture plates were used to see the reduction of oral pathogens before and after Licorice mouthwash and then Colony forming units were recorded for pre and post usage of the mouthwash with the help of colony counter app.

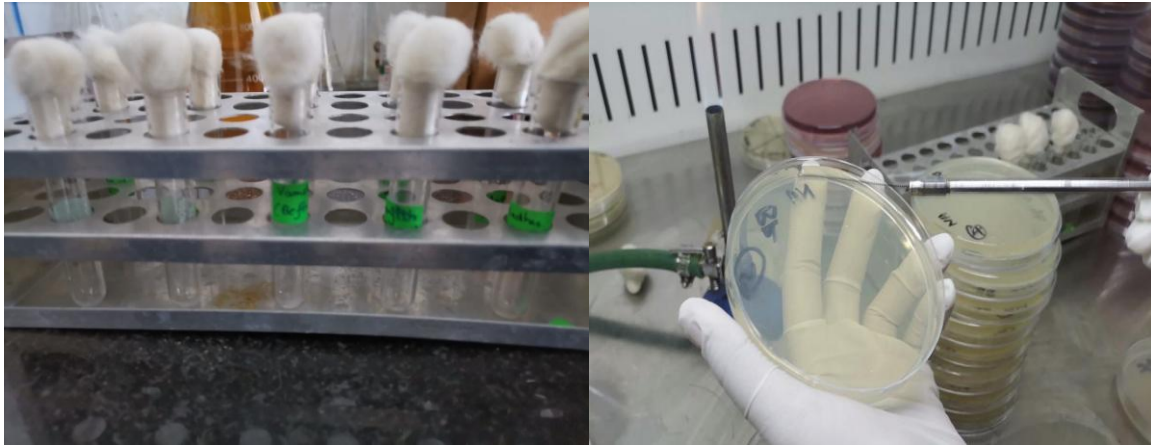


Figure 1:Represents the saliva samples before and after using mouthwash along with saline (left) and Nutrient Agar with salivary samples (right)

RESULTS AND DISCUSSION:

UNDER PEER REVIEW

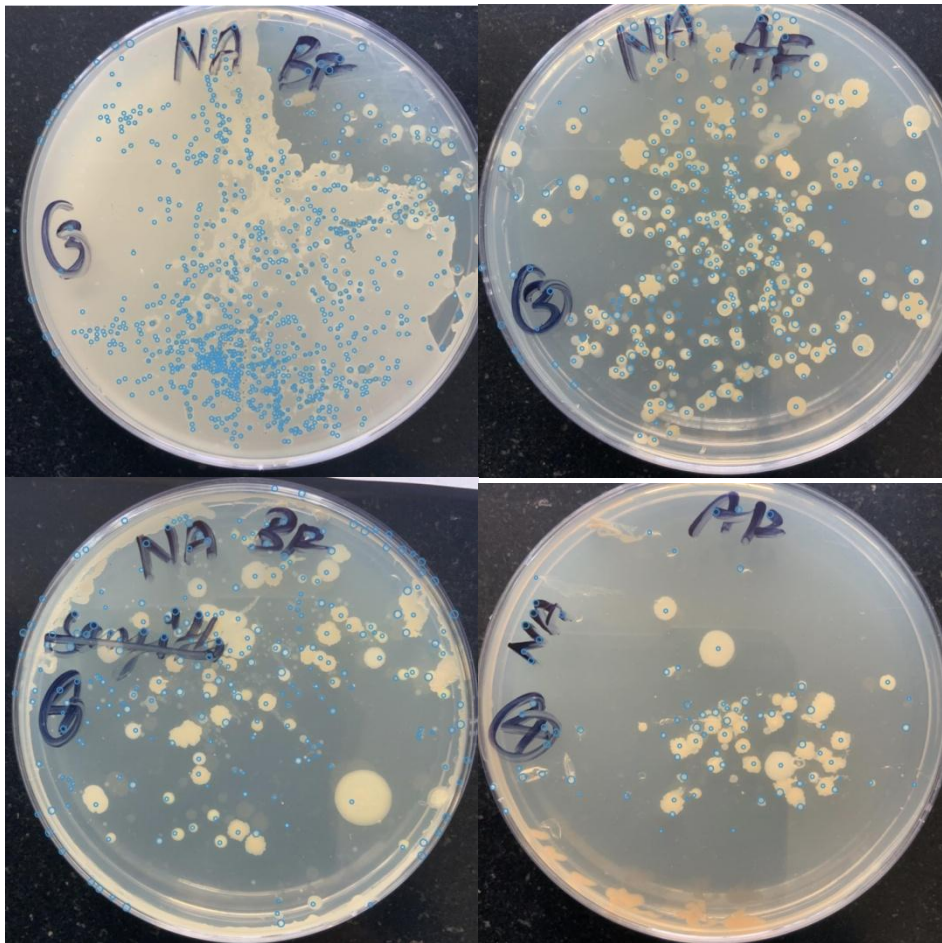


FIGURE 2:This depicts the Colony forming growth of saliva samples in Nutrient Agar

PARTICIPANTS	CFU BEFORE USING MOUTHWASH	CFU AFTER USING MOUTHWASH
PARTICIPANT 1	225	101
PARTICIPANT 2	418	256

PARTICIPANT 3	860	307
PARTICIPANT 4	246	89
PARTICIPANT 5	362	214
PARTICIPANT 6	240	150
PARTICIPANT 7	180	78
PARTICIPANT 8	420	230
PARTICIPANT 9	356	214
PARTICIPANT 10	200	130
MEAN VALUES	350.7	176.9

TABLE 1: Depicts the number of colony forming units before and after using herbal mouthwash.

SAMPLE	CFU BEFORE USING MOUTHWASH	CFU AFTER USING MOUTHWASH
SAMPLE 1	330	25
SAMPLE 2	420	1
SAMPLE 3	190	5
SAMPLE 4	185	6
SAMPLE 5	140	15
SAMPLE 6	700	28
SAMPLE 7	106	2
MEAN VALUES	295.86	11.714

TABLE 2: Depicts the Mean values of CFU before and after Chlorhexidine mouthwash

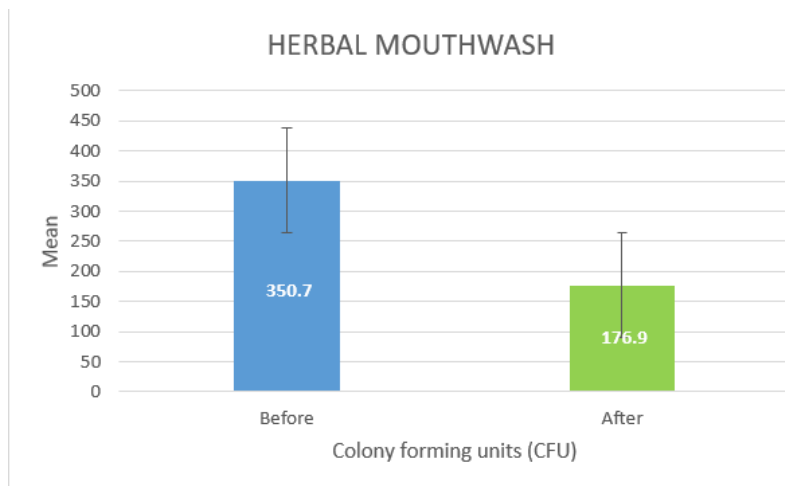


FIGURE 3 : Error graph depicts the CFU before and after usage of Herbal mouthwash. X axis represents the number of participants involved in this study, Y axis represents the CFU. Blue colour denotes the CFU before using mouthwash, green colour depicts the CFU after usage of mouthwash. From this it is inferred that the majority of the participants showed more CFU before mouthwash when compared to CFU after herbal mouthwash.

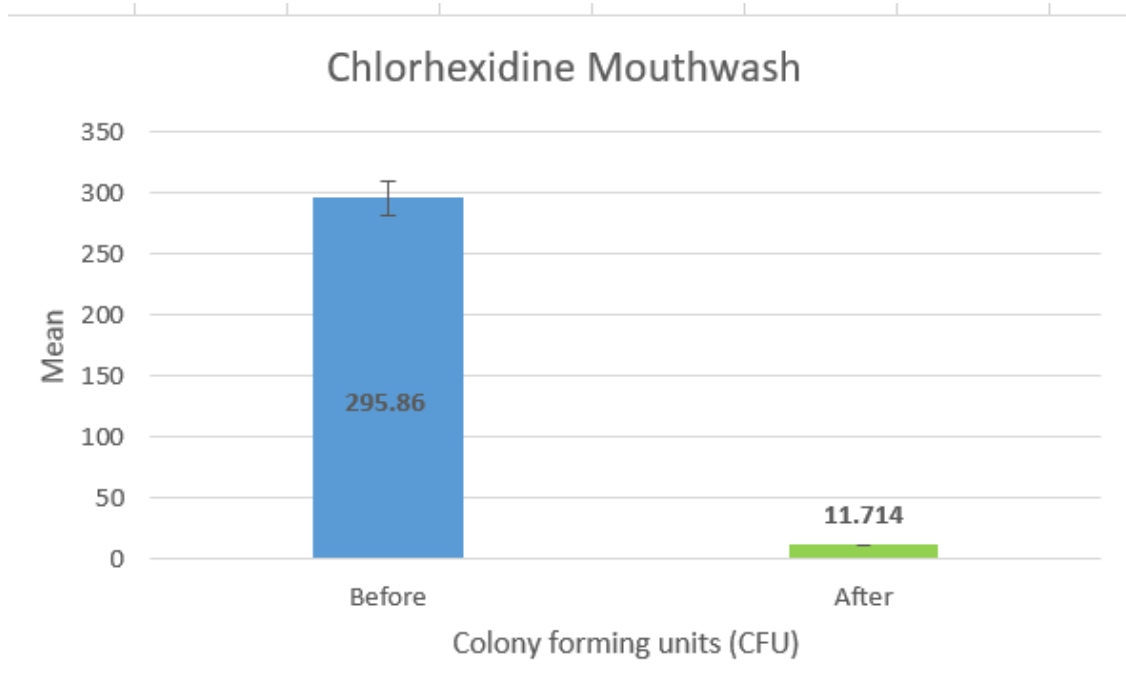


FIGURE 4 : Bar graph depicts the CFU of before and after the Chlorhexidine mouthwash. X axis represents the number of samples taken in this study, Y axis represents the CFU. Blue colour denotes the CFU before using mouthwash, green color depicts the CFU after usage of mouthwash. From this it is inferred that the majority of the participants showed more CFU before mouthwash with the least in CFU after mouthwash.

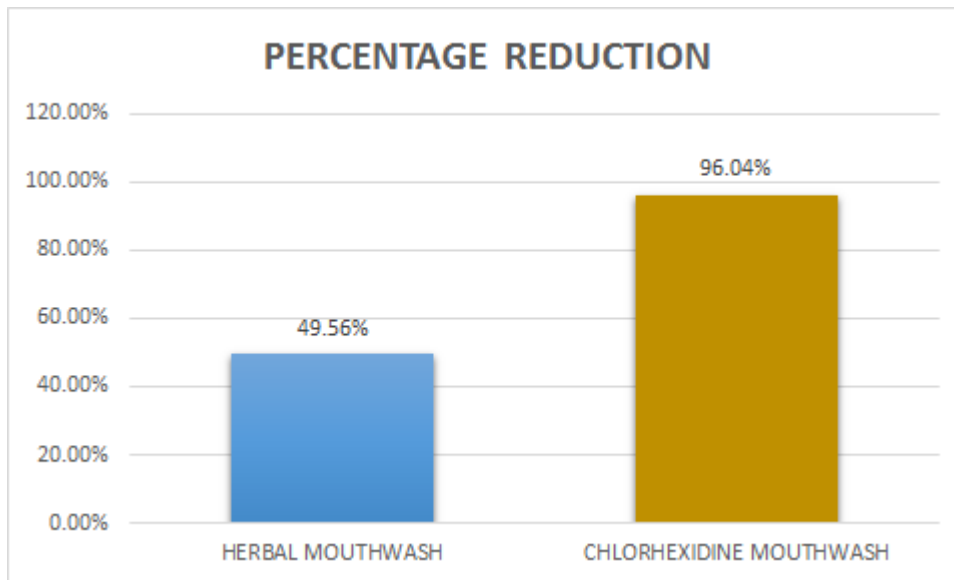


FIGURE 5: Bar graph Depicts the mean percentage value reduction of herbal and chlorhexidine mouthwash. Blue represents the herbal mouthwash and yellow represents the chlorhexidine mouthwash. From this it is shown that chlorhexidine mouthwash showed maximum reduction compared to herbal mouthwash.

In our present study **Figure 2** represents Nutrient agar which is used in procedures commonly performed for the cultivation of microbes and supporting growth of a wide range of non-fastidious organisms and its popular because it can grow a variety of types of bacteria and fungi, and contains many nutrients needed for the bacterial growth and in our present study, the efficacy of the herbal mouthwash and Chlorhexidine mouthwash in reducing oral pathogens were assessed. In our present study **Table 1** and **figure 3** explains the total Colony forming unit before and after using herbal mouthwash where the mean value before using herbal mouthwash was 350.7 and the mean after using herbal mouthwash was 176.9.

In **Figure 5** the mean reduction percentage is estimated to be 49.56%. From our study **Table 2** and **figure 4** explains the total Colony forming unit before and after using Chlorhexidine mouthwash which was considered as a negative control in our study where the mean value before using Chlorhexidine mouthwash was 295.86 and the mean value after using Chlorhexidine mouthwash was 11.714, in figure 5 mean reduction percentage is estimated to be 96.04%. While in a study Decker et al. improved antiplaque strategies by using Chlorhexidine mouthwash in combination with plaque. Chlorhexidine (0.1%) was used as the positive control, saline was the negative control, and two CHT derivatives were used against streptococcus sanguis. Their results proved that Chlorhexidine and CHT combination was more stronger than Chlorhexidine alone because of the bioadhesive properties of CHT and the antibacterial activity of Chlorhexidine.(9)

Andre et al. demonstrated the bactericidal activity against *Streptococcus mutans* growth on dentures as a biofilm by using cepacol. On the other hand a product named pearl drops were

also tested in this study which showed negative results in accordance with the standard(10) Al Bayati ,Sumaiman's et al demonstrated with Listerine® mouthwash for 30 s which results in decrease of bacterial counts in saliva.. They reported that the aqueous extract of *Salvadora persica* showed strongest antibacterial activity against *Streptococcus faecalis*(11) AM Khalessi et al in 2004 demonstrated the efficacy of herbal mouth wash in controlling the plaque formation(12)Limitation of the study is small sample size and in vitro.Our team has extensive knowledge and research experience that has translate into high quality publications(13–24),(25–29). In future similar study in large scale productions for targeted drug delivery to treat and prevent a wide array of oral microbial infections.

CONCLUSION

The results of our study and also **figure 5** shows a mean reduction in the colony-forming units with herbal mouthwash and Chlorhexidine mouthwashes and showed that both mouthwashes gave reduction in colony forming growth after its usage. Though chlorhexidine shows higher percentage reduction (98 %) the herbal mouthwash has reduced the count upto 50 percent.

NOTE:

The study highlights the efficacy of "Ayurveda medicine" which is an ancient tradition, used in some parts of India. This ancient concept should be carefully evaluated in the light of modern medical science and can be utilized partially if found suitable.

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