

## Contact Angle Analysis Of Herbal Mouthwashes - An In Vitro Study

Research Article

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### Abstract

**Introduction:** Dental plaque is a diverse microbial colony found on the tooth surface and it can be hard to remove. Thus several chemical plaque control methods like mouthwashes which are adjuvant to those mechanical plaque removal methods have gained widespread popularity in the world. Mouthwashes are effective in reducing plaque buildup and maintain good oral hygiene thereby reducing gingivitis and periodontitis. Nowadays, herbal mouthwashes are widely used because of their additional anti-inflammatory and antioxidant properties, which could further benefit gingival health. The contact angle quantifies the wettability of a particular material on a solid surface. Contact angle and wettability are inversely related to each other. The aim of this study is to evaluate the contact angle of three different herbal mouthwashes and determine their wettability with the surface of the tooth.

**Materials and Methods:** Three groups of herbal mouthwashes, 6 samples each totalling 18 samples were dispensed from a microsyringe on the facial surface of the microtomed tooth and the contact angles were measured with the help of the Ossila Goniometer. The obtained values from the samples were determined and the data was exported to SPSS software version 20.0 and was statistically analyzed. One way Anova test was used to find the significance and multiple intergroup comparisons were done using Tukey post hoc test. P-value less than or equal to 0.05 is considered to be significant.

**Results:** Comparing the three mouthwashes, Closeup herbals mouthwash had a low average contact angle of 23.55. The association of the contact angle between three mouthwashes was statistically significant ( $p$ -value = 0.010 < 0.05).

**Conclusion:** With the limitations of the study, it can be concluded that the Close up herbals mouthwash has higher surface wettability and lowest average contact angle.

**Keywords:** Wettability; Contact Angle; Herbal, Mouthwash; Innovative Measurement; Goniometer.

### Introduction

Dental plaque is a diverse microbial colony found on the tooth surface. They are embedded in a matrix of polymers and saliva [1]. Once the tooth is cleaned, a biofilm consisting of proteins and glycoproteins is rapidly absorbed on the tooth surface 4-12 hours after brushing. They collectively form a dental plaque when the receptors on salivary molecules get exposed to the bacteria and are absorbed on the surface of the tooth. Subsequently, secondary colonizers adhere to the already existing early colonizers through specific molecular interactions. These can involve protein-protein or carbohydrate-protein (lectin) interactions and this

process contributes to bacterial succession. Constant colonization of plaque on the tooth surface and soft tissues leads to a reversible condition called gingivitis. An inflammatory response is due to excessive dental biofilm colonization and inadequate brushing habits. It is one of the most prevalent oral diseases worldwide causing halitosis, bleeding, and pain [2].

Periodontitis is the most severe form of gingivitis causing sensitivity and receding gums causing periodontal pockets creating a place for more accumulation of plaque and tartar. Therefore, effective tooth brushing is essential to remove food debris, which prevents the further development of plaque. Plaque can also be

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removed mechanically by scaling and root planning but several studies show that mechanical removal is not alone sufficient for the removal of oral flora which is responsible for gingival and periodontal diseases. Thus several chemical plaque control methods like mouthwashes which are adjuvant to those mechanical plaque removal methods have gained widespread popularity in the world. Studies pertaining to mouthwashes prove that they are effective in reducing plaque buildup and maintain good oral hygiene thereby reducing gingivitis and periodontitis. Chlorhexidine (CHX) is a broad-spectrum antiseptic and is considered to be the gold standard for dental plaque control. But it can stain the tooth and tongue and cause adverse effects in oral mucosa after prolonged use [3]. Thus in search of alternatives, herbal medicines derived from botanical sources play a major role. Plant-derived products have played an important role in dentistry for a long period and they inhibit microbial growth, reduce and soothe irritation, and relieve pain [4]. Studies reveal that it has positive results in plaque and gingivitis control [5]. Herbal mouthwashes are prepared with extracts and essential oils from therapeutic plants, containing a mixture of active agents such as catechins, tannins, and sterols. Compared with the antimicrobial mechanisms, herbal mouthwashes can have additional anti-inflammatory and antioxidant properties, which could further benefit gingival health [6].

The contact angle is a measurable angle of a drop of liquid that forms on the surface of the solid. It also quantifies the wettability of a particular material on a solid surface [7]. Measurement of the contact angle is used to determine the solid-liquid and vapor-solid interfacial tensions present in the materials which shows the bonding relationship between two surfaces. The contact angle is used as a method for determining the hydrophilicity of material [8]. Lower contact angles indicate increased wettability or greater hydrophilicity, while greater contact angles indicate limited wettability or greater hydrophobicity. A larger contact angle also indicates a weak adhesive force between the liquid and the solid [9].

The aim of this study is to evaluate the contact angle of herbal mouthwashes and determine their wettability with the surface of the tooth.

### Materials and Methods

This in vitro study was done at white lab - saveetha dental college. Three groups of herbal mouthwashes, 6 samples each totalling 18 samples were dispensed from a microsyringe on the facial surface of the microtomed tooth and the contact angles were measured with the help of the Ossila Goniometer. The obtained values from the samples were determined and the data was exported to SPSS software version 20.0 and was statistically analyzed. One way Anova test was used to find the significance and multiple intergroup comparisons were done using Tukey post hoc test. P-value less than or equal to 0.05 is considered to be significant.

### Results

The Ossila goniometer was used to measure different contact angles. Our study is first of its kind in the literature, no studies have analysed this parameter in dentistry. It is known that contact angle and wettability are inversely proportional to each other. Comparing the three mouthwashes in our study, Closeup herbals mouthwash had a low average contact angle (23.55), followed by Himalaya's hiora mouthwash (32.12) and KP Namboodiris herbal mouthwash had highest average angle compared to the other two (35.79) (Table 1) (Figure 1). So, Closeup herbals mouthwash is known to have high wettability. The test used in finding significance was one way ANOVA. The association of the contact angles between three mouthwashes in our study was found to be statistically significant ( $p\text{-value} = 0.010 < 0.05$ ) (Table 2). Multiple intergroup comparisons using Tukey post hoc test were done to compare between the intergroups (Table 3).

**Table 1. Table shows the average contact angles of 6 samples from each herbal mouthwash on the facial surface of the microtomed tooth and also represents the mean average value for each mouthwash.**

SAMPLE NUMBER	K P NAMBOODIRIS	HIMALAYA'S HIORA	CLOSE UP HERBALS
1	24.77	37.39	19.29
2	37.38	36.67	24.64
3	40.79	33.58	16.25
4	26.74	28.66	22.13
5	42.95	27.98	29.03
6	42.15	28.46	29.98
MEAN	35.79	32.12	23.55

**Table 2. Table shows the mean, standard deviation, and significance of the herbal mouthwashes.**

	N	Mean	Std. Deviation	95% confidence interval for mean		Significance
				Lower bound	Upper bound	
KP Namboodiris	6	35.797	8.032	27.367	44.226	0.01
Himalaya's Hiora	6	32.123	4.315	27.595	36.652	
Closeup herbals	6	23.553	5.404	17.882	29.225	
Total	18	30.491	7.804	26.610	34.372	

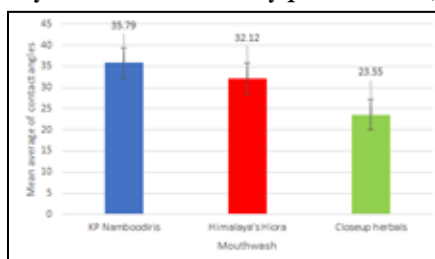
One way ANOVA test was used to find the significance. P-value less than or equal to 0.05 is considered to be statistically significant.

**Table 3.** Table shows the multiple inter-group comparisons between the mouthwashes.

Groups		Mean difference	95% confidence interval		Significance
			Lower bound	Upper bound	
KP Namboodiris	Himalaya's Hiora	3.673	-5.504	12.850	0.564
	Closeup herbals	12.24333*	3.066	21.420	0.009
Himalaya's Hiora	KP Namboodiris	-3.673	-12.850	5.504	0.564
	Closeup herbals	8.57	-0.607	17.747	0.069
Closeup herbals	KP Namboodiris	-12.24333*	-21.420	-3.066	0.009
	Himalaya's Hiora	-8.57	-17.747	0.607	0.069

Multiple intergroup comparison was done using Tukey post hoc test. P-value less than or equal to 0.05 is considered to be statistically significant.

**Figure 1.** Bar graph depicts the average mean for the contact angle of three different mouthwashes. X-axis represents different herbal mouthwashes and Y-axis represents the mean average of the contact angles. Blue denotes KP Namboodiris, red denotes Himalaya's hiora and green denotes closeup herbals. Comparing three mouthwashes, closeup herbals had a low average contact angle and better wettability. The association of the contact angles between three mouthwashes was statistically significant (One way ANOVA with Tukey post hoc test; p-value= 0.010 < 0.05).



**Discussion**

Our team has extensive knowledge and research experience that has translated into high-quality publications [10-29]. Surface wettability has an important role in many biological, chemical and physical processes. The contact angle is the angle at the interface where water, air and solid meet. Low contact-angle values demonstrate a tendency of the water to spread and adhere to the surface, whereas high contact-angle values show the surface's tendency to repel water [30]. The most common method for surface-wetting characterization is sessile-drop goniometry, due to its simplicity. It is performed by recording a video of a water drop on a solid surface and determining the contact angle from the images of the video by a fitting procedure. The method determines the contact angle from the shape of the droplet and can be applied to a wide variety of materials, from biological surfaces to tooth, polymers, metals, ceramics, minerals and so on.

With a rise in the incidence of oral diseases there is continuous research to identify effective and safe oral hygiene aids for patients' self-care. Mechanical plaque control by tooth brushing methods and interdental aids may not be enough to completely remove dental plaque from tooth surfaces. Antimicrobial mouthwashes may help to improve plaque control and gingival health by reducing dental plaque from difficult to reach areas. Though mouthwashes have been in use for prevention as well as curative purposes, their side effects and affordability are of growing concern [31, 32]. For individuals with existing disease, the use of mouthwashes or dentifrice to deliver antimicrobial and antiplaque agents has only limited or no effects on the subgingival flora [33]. The use of natural products is always recommended to reduce the usage of chemicals causing human and environmental risk. The

commercially available mouthwashes contain many chemicals causing various ill effects. Their constituents include water which is the chief constituent, ethanol, dyes, surface active agents, zinc chloride/acetate, aluminum potassium sulfate (astringent) and phenolic compounds, quaternary ammonium compounds and essential oils such as oil of peppermint (as antibacterial agents) among others [34]. Commercial mouthwashes also cause taste disturbance, tooth staining, sensation of a dry mouth and discoloration. Alcohol-containing mouthwashes worsen halitosis by causing dryness of mouth. Soreness, ulceration, chemical burn and redness may sometimes occur (e.g., aphthous stomatitis and allergic contact stomatitis) if the person is allergic or sensitive to mouthwash ingredients such as preservatives, coloring agents, flavors and fragrances. To overcome such side effects, the World Health Organization advises researchers to investigate the possible use of natural products such as herb and plant extracts.

Herbs and plant extracts have been used in oral hygiene products for many years [35]. The present study was done using non-alcoholic herbal mouthwashes containing extracts of tea tree, meswak & betel and tulsi & cardamom. Tea tree oil (TTO) derived from the paperbark tea tree is being used in medicinal and dental products because of its antibacterial and antiinflammatory activities. These activities were shown to be related to the active ingredients such as 1,8-cineole and terpinen-4 ol. There are very few studies regarding the effects of TTO on periodontal tissues. The results of our study indicate that closeup herbals mouthwash containing extracts of tea tree had the lowest average contact angle of 23.55 which in turn has the highest wettability compared to the other mouthwashes. A previous study done by Rahman et al., supports the use of tea tree oil as an anti-plaque agent in comparison with chlorhexidine [36]. Another study conducted by Soukoulis and Hirsch tested the effect of TTO gel on gingivitis and plaque. They reported significant reductions in gingival in-

flammation with TTO gel. However, TTO gel did not reduce the plaque scores. Hence they concluded that TTO has no plaque inhibitory effect, but anti-inflammatory effect on gingivitis and can be a useful adjunct to chemotherapeutic periodontal therapy which could not relate to this study much [37].

Hiora is a herbal mouthwash manufactured by the Himalaya Drug Company ; each gram contains 5.0 mg of Pilu, 10 mg of Bibhitaka, 10 mg of Nagavalli (Piper betel), 1.2 mg of Gandhapura taila, 0.2 mg of Ela, 1.6 of Peppermint satva and 0.4 mg of Yavani-satva. It is claimed that it acts as an oral antiseptic and prevents tooth decay and also prevents bad breath and reduces plaque and gingivitis. The results of our study indicate that Himalaya's Hiora has the second highest average contact angle (32.12) which in turn has a poorer wettability than Close up herbals. Aspalli et al evaluated the antiplaque and antigingivitis effects of herbal mouthwash in the treatment of plaque-induced gingivitis and found that it can be effectively used as an adjunct to mechanical therapy with less side effects which could not relate much to our study [38]. Another study conducted by Ramamurthy and Mg et al concluded that Hiora and Chlorhexidine mouthwashes were equally effective in the treatment of gingivitis which could not relate to our study [39]. Further studies need to be undertaken with a more emphasis on herbal products in order to show the effectiveness and hence prove its merit.

## Conclusion

The results of the study proved that the Close up herbals mouthwash has higher surface wettability and lowest average contact angle. Hence more evidence pertaining to the usage of herbal products need to be done with more clinical and randomized control trials on a larger scale to continue their development and usage.

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