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Efficacy Of Herbal And Chlorhexidine Mouthrinses On Salivary Ph Neutralization Following Beverage Induced Oral Acidity In Adults – A Randomized Control Trial

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Abstract
Background: *Acidic beverages are increasingly linked to dental erosion, caries, and oral health issues by lowering salivary pH below the critical level for enamel protection. Mouthwashes are used as adjuncts to neutralize oral acidity. With chlorhexidine as the gold standard, herbal mouthwashes are gaining traction due to fewer side effects. However, limited data compares their pH-restoring efficacy post-acidic challenge. This study evaluates and compares the neutralizing capacity of chlorhexidine and herbal mouthwashes on salivary pH.*
Materials and methods: *In this randomized controlled trial, 90 healthy adults (20–30 years) were divided into three groups: chlorhexidine mouthwash (Group I), herbal mouthwash (Group II), and water (Group III). Baseline unstimulated saliva was collected, followed by an acidic challenge (soft drink). Participants then rinsed with their assigned solution. Salivary pH was measured immediately post-rinse, at 15 and 45 minutes using a digital pH meter. Statistical analysis compared pH changes across groups.*
Results: *All groups showed a significant drop in salivary pH after the acidic challenge. Following rinsing, a significant increase in pH was observed in all groups. The chlorhexidine group demonstrated the highest rise in pH, reaching and maintaining baseline levels at all time intervals ($p < 0.001$). The herbal mouthwash group showed a modest increase in pH, remaining below neutral levels throughout the study period. The water group showed an intermediate effect, with pH values approaching but not sustaining baseline levels. No significant differences were observed between groups at baseline and after the acidic challenge.*
Conclusions: *Chlorhexidine mouthwash was found to be the most effective in restoring and maintaining salivary pH following an acidic challenge. Herbal mouthwash showed limited efficacy, while water provided moderate but less sustained benefits. Further studies are needed to explore the long-term effects and potential of herbal alternatives.*

Keywords: *Salivary pH, Chlorhexidine, Herbal mouthwash, Dental erosion, Acidic beverages, Oral health, Randomized control trial*

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I. Introduction

Acid-induced dental erosion, or erosive tooth wear, represents a major oral health challenge for many patients(1). In recent years, there has been a significant rise in the consumption of acidic beverages such as soft drinks, fruit juices, teas and coffees.(2) This trend has contributed to a noticeable increase in the incidence of dental issues, particularly dental caries, periodontal disease and dental erosion. Among these, dental erosion has become particularly common in young adults. (3,4)

The mechanism through which acidic drinks lead to dental issues can be largely attributed to two major factors. Firstly, the acids produced by the oral microorganisms while metabolising fermentable carbohydrates present in these beverages contribute to the demineralisation of tooth enamel. Secondly, the low pH of the drinks themselves. (5) After consumption of these drinks, the pH of oral cavity can fall below the critical level (~5.5). The drop in the pH can lead to softening of enamel and increased risk of erosion and carries.

Brushing immediately after consumption of these beverages is discouraged as it may further cause abrasion of the already softened enamel surface. Hence, alternative strategies are necessary (6) . One practical approach involves use of mouthwashes that may help in neutralising oral acidity. The use of medicaments before and after exposure has been extensively studied for their effectiveness in neutralising salivary pH and protecting enamel. Lindquist et al. included the use of neutralising agents in their research to evaluate their effectiveness in

restoring salivary pH to safe levels [7]. Their findings revealed that it can take up to 15 minutes for pH to return to pre exposure levels [8].According to Dehghan et al, rinsing with water and Chlorhexidine restore salivary pH to baseline levels immediately, with the Chlorhexidine group showing a significantly higher pH than the water group [1].

There is limited research directly comparing the effectiveness of herbal and chlorhexidine-based mouthwashes in modifying salivary pH levels following beverage-induced oral acidity. The consumption of acidic beverages such as soft drinks, fruit juices and energy drinks can temporarily lower the salivary pH, creating an acidic environment in the oral cavity. This decrease in pH may contribute to enamel demineralisation, dental erosion, and an increased risk of dental caries if the acidity persists for a prolonged period.

Mouthwashes are commonly used as an adjunct to routine oral hygiene practices to help neutralize oral acids, reduce microbial load, and maintain oral health. Chlorhexidine mouthwash is widely considered the gold standard due to its strong antimicrobial and plaque-inhibiting properties, whereas herbal mouthwashes are increasingly preferred because they are natural, have fewer side effects, and possess antimicrobial and anti-inflammatory benefits. However, despite their widespread use, the comparative effectiveness of these mouthwashes in restoring and maintaining salivary pH after acidic challenges remains unclear, highlighting the need for further research in this area.

This study aims to evaluate and compare the neutralising capacity of herbal and chlorhexidine-based mouthwashes on salivary pH following the consumption of beverages that induce oral acidity. Acidic beverages are known to temporarily lower the pH of saliva, creating an environment that may contribute to enamel demineralisation, dental erosion, and an increased risk of dental caries. Assessing the ability of different mouthwashes to counteract this acidic challenge is important for understanding their role in restoring the normal pH balance of the oral cavity.

By examining how herbal and chlorhexidine mouthrinses influence the salivary pH response after exposure to acidic beverages, this study seeks to determine their effectiveness in neutralising oral acidity and promoting oral health. The findings may help in identifying suitable preventive measures that can be incorporated into routine oral hygiene practices, thereby assisting individuals in maintaining a balanced oral environment and reducing the potential harmful effects of dietary acids on dental tissues.

II. Methodology

Study design and setting: The present study was a 3-arm, parallel group, randomized controlled trial conducted in Department of Public Health Dentistry of M.A. Rangoonwala college of dental sciences and research centre, Pune. The trial was conducted with approval of Institutional Ethics Committee (IEC) following all Infection control protocols. The null hypothesis (H_0) for this study states that there is no significant difference in the salivary pH increase between the chlorhexidine mouthrinse group and the herbal mouthrinse or water groups.

Study participants: Healthy adults aged 20 - 30 years were recruited for the study. Individuals were excluded if they had history of allergies or hypersensitivity to mouthwash ingredients, were pregnant or lactating, had any systemic disease, consumed antibiotics or immunosuppressive drugs in the past two weeks, had chronic dental disease like periodontitis, had undergone any dental treatment in the past two weeks. All participants signed a provided informed consent prior to the participation. Participants were instructed not to use any mouthrinse on the morning of the study.

Sample size: This randomized control trial was conducted on 90 individuals which were divided into 3 groups. The sample size was calculated based on comparison of mean salivary pH between three groups using one-way ANOVA. The formula used was:

$$n = \frac{2\sigma^2 (Z_{\alpha/2} + Z_{\beta})^2}{d^2}$$

Based on previous studies evaluating salivary pH changes[1], the calculated sample size was 27 participants per group, which was rounded off to 30 participants per group to compensate for possible dropouts. Thus, the total sample size was 90 participants (30 per group).

Randomization and Allocation Concealment: Participants were randomly allocated into three groups using a simple randomization by lottery method to ensure unbiased allocation. Allocation concealment was achieved using sequentially numbered, opaque, sealed envelopes (SNOSE technique). Each envelope contained the group assignment and was opened only at the time of intervention. Blinding was maintained at the participant level (single-blind design), where participants were unaware of the type of mouthrinse administered.

Study procedure: Appointments for sample collection were scheduled in the morning between 8:30 and 10:00 am to minimise the influence of daily variations in salivary composition and pH. Upon arrival, participants were instructed to remain seated and relaxed before the procedure. An unstimulated whole saliva sample of approximately 10 mL was first collected from each participant to establish a baseline salivary pH measurement. Unstimulated saliva was collected by asking the participants to allow saliva to accumulate naturally in the mouth and then expectorate into a sterile collection container without chewing or stimulation.

Following the baseline collection, participants were subjected to an acidic challenge to simulate the effect of acidic beverage consumption on the oral environment. This was achieved by asking them to rinse their mouth with 30 mL of a cold drink for two minutes, ensuring adequate exposure of the oral cavity to the acidic beverage. Immediately after this procedure, another saliva sample was collected to assess the change in salivary pH resulting from the induced oral acidity.

After the acidic challenge, participants were randomly assigned to one of three treatment groups: 15 mL of chlorhexidine mouthwash, 15 mL of herbal mouthwash, or 15 mL of water as a control rinse. Participants were instructed to rinse with the assigned solution for a standardised duration and then expectorate. To reduce bias, the participants were blinded to the treatment assignments so that they were unaware of which mouthrinse they received.

Subsequently, saliva samples were collected at multiple time intervals to monitor the recovery of salivary pH over time. Samples were obtained immediately after rinsing and again at 15 minutes and 45 minutes post-rinsing. During the entire study visit, participants were instructed not to consume any food or beverages to avoid external factors that could influence salivary pH levels. All collected saliva samples were analyzed using a high-accuracy digital pH meter with a precision of 0.01 to ensure reliable and accurate measurement of salivary pH changes throughout the study period.

Statistical Analysis: Data were entered into Microsoft Excel and analyzed using Statistical Package for the Social Sciences (SPSS) version XX (IBM Corp., Armonk, NY, USA). Descriptive statistics were expressed as mean, standard deviation (SD), and standard error (SE). Normality of data was assessed using the Shapiro–Wilk test. Intergroup comparisons of mean salivary pH at different time intervals were performed using one-way Analysis of Variance (ANOVA). Post hoc analysis was carried out using the Tukey test for pairwise comparison between groups. A p-value < 0.05 was considered statistically significant.

III. Result

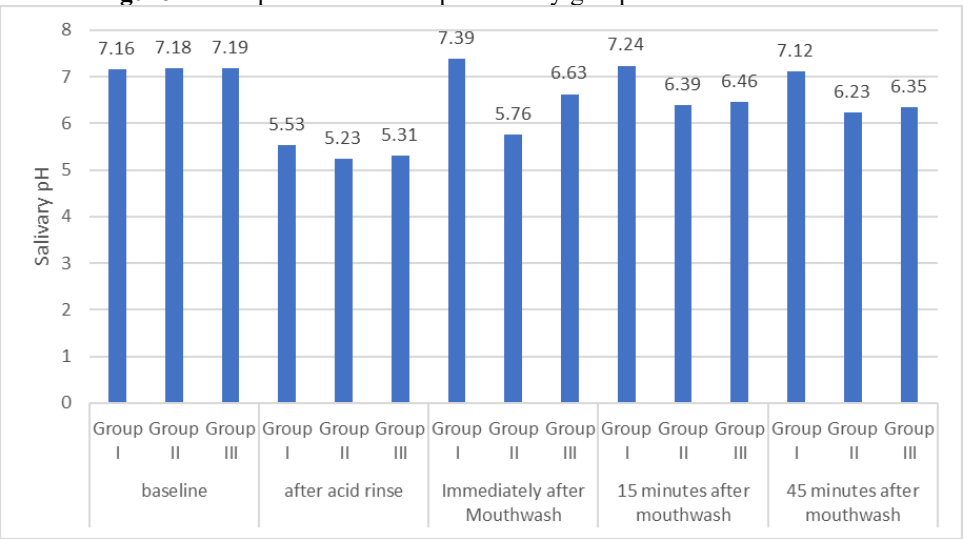
Table 1: Demographic Characteristics and Clinical Findings of Study Participants

Parameter	Group I (n=30)	Group II (n=30)	Group III (n=30)
Gender Distribution			
Females – n (%)	22 (24.4%)	22 (24.4%)	26 (28.9%)
Males – n (%)	8 (8.9%)	8 (8.9%)	4 (4.4%)
Age (years)			
Mean ± SD	23.1 ± 0.885	23.3 ± 0.828	23.3 ± 0.837
Minimum	21	22	21
Maximum	25	25	24
Number of Decayed Teeth			
Mean ± SD	1.23 ± 0.504	2.00 ± 1.259	1.33 ± 0.606
Minimum	1	0	1
Maximum	3	4	3

Table 2: Comparison of Mean pH in study groups at different intervals

pH time points	Groups	N	Mean	SD	SE	F value	P value
baseline	Group I	30	7.16	0.23	0.041	0.128	0.880
	Group II	30	7.18	0.24	0.044		
	Group III	30	7.19	0.25	0.045		
after acid rinse	Group I	30	5.53	0.40	0.073	4.692	0.13
	Group II	30	5.23	0.37	0.068		
	Group III	30	5.31	0.31	0.057		
Immediately after Mouthwash	Group I	30	7.39	0.17	0.031	671.84	<.001*
	Group II	30	5.76	0.17	0.031		
	Group III	30	6.63	0.17	0.031		
15 minutes after mouthwash	Group I	30	7.24	0.13	0.024	318.27	<.001*
	Group II	30	6.39	0.17	0.030		
	Group III	30	6.46	0.16	0.029		
45 minutes after mouthwash	Group I	30	7.12	0.14	0.026	303.41	<.001*
	Group II	30	6.23	0.159	0.0290		
	Group III	30	6.35	0.174	0.0317		

Figure 1: Comparison of Mean pH in study groups at different intervals



A total of 90 participants were included in the present study. The study population was predominantly female with females constituting 77.7% (24.4% in group 1, 24.4% in group 2 and 28.9% in group 3) and males 22.2% (8.9% in groups 1, 8.9% in group 2 and 4.4% in group 3) (Table 1). The mean age of participants in Group 1 was 23.1 years, in Group 2 was 23.3 years and the Group 3 was 23.3 years with ages ranging between 20-30 years (Table 2). All participants attended the clinical visit. No adverse event was reported during the trial. Salivary ph at different time intervals is shown and listed with statistical results. (Table 3) (Figure 1)

There was no significant difference in salivary pH among the 3 groups at baseline. After rinsing with acidic drink, the pH significantly dropped in all 3 groups. Salivary pH was raised immediately after rinsing with the provided mouth-rinse in all three groups, with the pH significantly higher in Group 1. In the Chlorhexidine mouth rinse group, the mean pH after acidic challenge was 5.53 which was immediately raised to 7.39. Then, after 15 mins, the pH dropped to 7.24 and after 45 mins, pH dropped to 7.12. This shows that the pH was maintained at the baseline values throughout the experiment. In the Herbal mouthrinse group(Group 2), the mean pH after acidic challenge was 5.23 which was raised to 5.76 immediately after rinsing with mouthwash. After 15 mins, the pH value was raised to 6.39 and after 45 mins, the values were 6.23 indicating that the pH was acidic throughout the experiment. However, initially, the pH dropped close to the critical pH but after 15 and 45 mins the pH was slightly raised. In the water group (Group 3), the mean pH after acidic challenge was 5.31 which was raised to 6.63 immediately after rinsing. After 15 mins, the pH was 6.46 and after 45 mins, the pH was 6.35. This indicates that initially, the pH raised close to the baseline level, pH dropped after 15 and 45 mins intervals.

According to the statistics, the p values of pH measured at baseline levels and after acidic challenge were not statistically significant. This shows that there was uniformity in all three groups during those two steps of the experiment. After rinsing with the given mouthrinses, comparative analysis was made and found that chlorhexidine gives a greater increase in salivary pH with a P = <0.001 which is found to be statistically significant.

The null hypothesis for this study was rejected ; there was a significant difference in the increase of salivary pH in chlorhexidine mouthrinse group when compared with herbal mouthrinse and water groups.

IV. Discussion

This study was undertaken to address the critical role of salivary pH in preventing conditions exacerbated by frequent acidic challenges from diet and beverages that lower oral pH below the critical threshold of 5.5. It compares the pH-neutralizing effects of chlorhexidine, herbal mouthwashes, and water post-acidic challenge to identify rinses that rapidly restore neutral pH and reduce oral health risks.

According to a study conducted by Manikandan et al., in 2021, comparison was made in the salivary pH after rinsing with chlorhexidine and herbal mouthrinses. The results stated that herbal mouthrinse elevated the pH better than chlorhexidine mouthrinses[9]. In the present study, the Chlorhexidine group gave better results than the Herbal group. However, the herbal mouthrinse taken in that study were tea tree oil and lemongrass oil based which is not readily available in the market. Therefore, no direct comparison was possible. The herbal mouthrinse used in this study contains Potassium nitrate, Clove, Menthol, etc which are readily available in herbal formulations in the market.

According to a study conducted by Dehghan et al., in 2015, the pH was measured using listrene, a two step mouth rinse and water after acidic challenge. The results were such that, after 15 and 45 mins, pH values of

all three groups went back to baseline values with no significant difference among the 3 groups. They included only 12 participants which is insufficient for generalising to a large population. The number of visits in that study were more than one and the participants were subjected to acidic challenge during each visit which increases the exposure to acid. [(1)]

In contrast to the findings of Maria Polyakova et al., who reported no significant differences among the tested rinsing solutions, the present study demonstrated a statistically significant improvement in pH following the use of chlorhexidine. Additionally, the salary pH was restored to the baseline levels and maintained overtime, with clearer group-wise differences observed across intervals, supporting stronger clinical relevance[(5)]

This study has several notable strengths that enhance its internal validity and clinical relevance. First, the use of a randomized controlled design with three well-defined intervention groups (chlorhexidine, herbal mouthrinse, and water) allows for direct comparison of pH-neutralizing effects under standardized conditions. Second, the inclusion of repeated salivary pH measurements at multiple time points (immediately after rinsing, 15 minutes, and 45 minutes) provides a dynamic picture of pH recovery and helps distinguish transient from sustained effects. Third, the protocol controlled for major confounders by standardizing time of day, dietary restrictions during the visit, and rinse duration, and by excluding participants with systemic diseases or recent dental procedures that could influence salivary composition or pH. Finally, the relatively large sample size (n = 90) and the use of a high-accuracy digital pH meter improve the reliability and precision of the observed pH differences.

Despite these strengths, the study has several limitations that should be acknowledged. First, the study population was limited to healthy young adults aged 20–30 years, which may limit the generalizability of findings to older adults, children, or individuals with systemic diseases or compromised salivary function. Second, the herbal mouthrinse formulation used (containing potassium nitrate, clove, menthol, and related ingredients) does not represent the full spectrum of herbal products available, so results may not be extrapolated to other herbal rinse types.

V. Conclusion

Within the limitations of the present study, it can be concluded that all three rinsing agents—chlorhexidine mouthwash, herbal mouthwash, and water—demonstrated the ability to increase salivary pH immediately after an acidic challenge. However, the extent and sustainability of pH recovery varied significantly among the groups.

Chlorhexidine mouthwash exhibited the most effective and consistent neutralizing capacity, rapidly restoring salivary pH to baseline levels and maintaining it within the neutral range over time. In contrast, the herbal mouthwash showed only a modest increase in pH, with values remaining below neutral throughout the observation period, indicating a comparatively limited buffering effect. The water rinse demonstrated an intermediate effect, producing an initial rise in pH close to baseline levels but failing to sustain neutrality over time.

These findings suggest that chlorhexidine mouthwash is more effective in neutralizing oral acidity and maintaining salivary pH following exposure to acidic beverages. Therefore, it may serve as a beneficial adjunct in preventing enamel demineralization and reducing the risk of dental erosion and caries in individuals frequently consuming acidic drinks.

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