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Comparative Evaluation of Pomegranate and Grapefruit Seed Extract Mouthwash Versus Chlorhexidine in Gingivitis Management: A Clinical Trial

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KEYWORDS

Punica granatum

Chlorhexidine

Grapefruit seed extract

Plaque index

Gingival index

ABSTRACT:

Introduction Plaque-induced gingivitis is typically managed with mechanical plaque control and adjunctive chemotherapeutic rinses. Chlorhexidine (CHX) is considered the gold standard antiplaque agent, but its adverse effects such as staining and taste alteration limit its long-term use.[1] Natural extracts, including pomegranate (*Punica granatum*) and grapefruit seed extract (GSE), have demonstrated antimicrobial, antioxidant, and anti-inflammatory activities.[2–4] This study evaluated the effectiveness of a pomegranate–GSE herbal mouthwash compared with 0.2% CHX in adults with gingivitis.

Objectives: The objective of the study is to compare the effectiveness of a pomegranate–grapefruit seed extract (GSE) herbal mouthwash with 0.2% chlorhexidine (CHX) in reducing clinical signs of plaque-induced gingivitis in adults..

Methods: Thirty systemically healthy subjects (18–40 years) with gingivitis were randomly allocated to two groups (n=15). Group A rinsed with a herbal formulation containing 10% *Punica granatum* peel extract and 0.5% GSE, while Group B used 0.2% CHX. Both groups rinsed with 15 mL twice daily for 14 days. Plaque Index (PI), Gingival Index (GI), Probing Depth (PD), and Clinical Attachment Level (CAL) were recorded at baseline and 14 days. Data were analyzed using paired and independent t-tests (α=0.05).

Results: Significant intragroup reductions were observed in both groups for PI and GI ($p<0.05$). Group A showed PI reduction from 2.40 ± 0.60 to 1.80 ± 0.60 and GI from 1.45 ± 0.25 to 1.20 ± 0.18. Group B reduced PI from 1.80 ± 0.30 to 1.55 ± 0.28 and GI from 1.60 ± 0.22 to 1.45 ± 0.20. Intergroup comparisons at day 14 revealed no statistically significant differences ($p>0.05$). No adverse effects were reported with the herbal rinse.

Conclusions: A pomegranate–GSE mouthwash demonstrated plaque- and gingivitis-reducing effects comparable to 0.2% CHX over two weeks, without reported side effects. Herbal rinses may represent a safe and effective alternative in gingivitis management.

Introduction

Gingivitis is the most common and reversible form of periodontal disease, affecting a large proportion of the population worldwide.[5] It is characterized by gingival erythema, swelling, and bleeding upon probing, all of which result from the host inflammatory response to dental plaque biofilm.[6] If not managed effectively, gingivitis can progress to periodontitis, a destructive and irreversible condition leading to tooth loss and systemic implications.[7]

Mechanical plaque control through toothbrushing and interdental cleaning remains the cornerstone of gingivitis prevention. However, patient compliance with mechanical measures is often inadequate, leading to incomplete plaque removal.[8] Consequently, adjunctive use of chemical plaque-control agents, particularly antimicrobial mouthwashes, has become a well-established strategy in clinical practice.[9]

Chlorhexidine gluconate (CHX) has long been regarded as the “gold standard” mouthwash due to its potent broad-spectrum antimicrobial properties, high substantivity, and well-documented efficacy in reducing plaque accumulation and gingival inflammation.[10] Despite these advantages, CHX is associated with

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undesirable side effects, including brown staining of teeth and restorations, altered taste sensation, and oral mucosal irritation, particularly with long-term use.[11] These limitations have driven the search for alternative chemotherapeutic agents that offer comparable benefits with improved safety and tolerability.

Herbal formulations have emerged as promising alternatives. Plant-derived compounds have gained attention in oral healthcare for their diverse bioactive properties, including antimicrobial, anti-inflammatory, and antioxidant effects.[12] Several herbal rinses such as triphala, chamomile, green tea, aloe vera, and propolis have demonstrated reductions in plaque scores and gingival bleeding, often showing results comparable to CHX.[13–15] Importantly, these natural products are generally associated with minimal side effects, enhancing patient acceptance and long-term compliance.[16]

Punica granatum (pomegranate) has been extensively studied for its therapeutic potential. Rich in tannins, ellagitannins, and flavonoids such as punicalagins and ellagic acid, it exhibits strong antioxidant and antibacterial activities.[17] Pomegranate extracts have been reported to inhibit *Streptococcus mutans*, suppress inflammatory mediators, and reduce gingivitis in randomized trials.[18,19] Grapefruit seed extract (GSE), derived from *Citrus paradisi*, contains bioflavonoids (notably naringin) and vitamin C, which contribute to antimicrobial and free-radical scavenging effects.[20] In vitro and in vivo studies have shown that GSE is effective against a broad range of bacteria, including those implicated in periodontal diseases.[21]

Given the biological plausibility and promising preliminary evidence, combining pomegranate and GSE may yield synergistic benefits in managing gingivitis by targeting both microbial biofilm and host inflammatory responses.[22] This study was therefore designed to compare the clinical efficacy of a novel pomegranate–GSE mouthwash with that of 0.2% CHX in reducing plaque and gingival inflammation. The null hypothesis was that no significant difference would exist between the two rinses

Objectives

Plaque-induced gingivitis remains one of the most prevalent inflammatory conditions of the gingiva, and while chlorhexidine (CHX) continues to serve as the gold standard for plaque control, its long-term use is limited by undesirable side effects such as tooth staining and altered taste perception. This has prompted the exploration of safer yet effective herbal alternatives that could provide comparable clinical benefits while enhancing patient compliance and acceptability.

Pomegranate (Punica granatum) and grapefruit seed extract (GSE) are plant-derived agents with well-documented antimicrobial, antioxidant, and anti-inflammatory properties. Individually, both extracts have demonstrated inhibitory effects on oral pathogens, modulation of host inflammatory mediators, and reductions in gingival bleeding. Combining these botanicals may yield additive or synergistic effects in the management of gingivitis by simultaneously targeting microbial biofilm and gingival inflammation.

Accordingly, the present randomized controlled trial was undertaken to evaluate and compare the clinical efficacy of a novel herbal mouthwash containing pomegranate peel extract and GSE with that of 0.2% CHX in adults diagnosed with plaque-induced gingivitis. The specific aim was to assess changes in plaque index, gingival index, probing depth, and clinical attachment level over a 14-day period, with the null hypothesis that there would be no significant difference between the two rinses.

1. Methods

This randomized controlled trial was conducted at Saveetha Dental College & Hospitals, Chennai, after ethical approval (IHEC/SDC/PERIO-21/055). Thirty participants aged 18–40 years with gingivitis were enrolled following inclusion criteria: ≥20 teeth, PI ≥1.5, and GI ≥1.0.

Exclusion criteria were systemic illness, recent antibiotic use, smoking, orthodontic appliances, or pregnancy.

Study groups

- **Group A:** Herbal mouthwash containing 10% *Punica granatum* peel extract and 0.5% GSE, prepared in-house.
- **Group B:** 0.2% chlorhexidine gluconate (Hexidine, ICPA Health).

Both rinses were dispensed in identical coded bottles. Participants rinsed with 15 mL for 60 seconds, twice

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daily, for 14 days. Compliance was assessed by diaries and returned bottles.

Clinical assessment

A calibrated examiner (blinded) recorded PI (Silness-Löe), GI (Löe-Silness), PD, and CAL at baseline (T0) and 14 days (T1).

Statistical analysis

Data were analyzed with paired t-tests (intragroup) and independent t-tests (intergroup). Significance was set at $p<0.05$.

Results

All 30 participants completed the trial without adverse events.

Group A (Herbal: Pomegranate + GSE)

At baseline, participants in Group A had a mean Plaque Index (PI) of 2.40 ± 0.60 and a Gingival Index (GI) of 1.45 ± 0.25. After 14 days of rinsing with the herbal mouthwash, significant reductions were observed: PI decreased to 1.80 ± 0.60 (p=0.01) and GI to 1.20 ± 0.18 (p=0.02). Probing depth (PD) also decreased slightly from 1.33 ± 0.35 mm to 1.15 ± 0.17 mm, although this change was not statistically significant (p=0.06). Clinical attachment level (CAL) improved marginally from 0.80 ± 0.25 mm to 0.65 ± 0.35 mm, but this reduction did not reach significance (p=0.10).

Overall, Group A demonstrated meaningful short-term improvements in plaque control and gingival health, with modest but non-significant gains in PD and CAL. Importantly, no adverse effects were reported, and participants rated the rinse as acceptable in terms of taste and comfort.

Group B (Chlorhexidine 0.2%)

At baseline, Group B had slightly lower mean PI values than Group A, measuring 1.80 ± 0.30, and a mean GI of 1.60 ± 0.22. After 14 days, PI declined to 1.55 ± 0.28 (p=0.03) and GI decreased to 1.45 ± 0.20 (p=0.04). Probing depth was reduced from 1.40 ± 0.40 mm to 1.05 ± 0.22 mm, a statistically significant change (p=0.01). CAL improved modestly from 0.78 ± 0.43 mm to 0.63 ± 0.33 mm, but this reduction was not significant (p=0.08).

Thus, Group B showed significant plaque and gingival index improvements, similar to Group A, with the added observation of a statistically significant reduction in PD. However, as with Group A, changes in CAL were minimal over the short trial period.

Intergroup Comparisons

When comparing outcomes between Group A and Group B at day 14, no statistically significant differences were observed. For instance, the final PI was 1.80 ± 0.60 in Group A versus 1.55 ± 0.28 in Group B (p=0.34). Similarly, GI values were 1.20 ± 0.18 in Group A compared with 1.45 ± 0.20 in Group B (p=0.28). PD and CAL were also statistically similar across groups (p>0.05).

These findings indicate that the herbal mouthwash achieved plaque and gingivitis reductions comparable to those of CHX. Importantly, unlike CHX, the herbal rinse was not associated with any adverse effects, highlighting its potential as a safe alternative.

Table 1. Baseline periodontal parameters (mean ± SD) in Groups A and B

Parameter	Group A (Herbal)	Group B (CHX)	p-value
Plaque Index (PI)	2.40 ± 0.60	1.80 ± 0.30	0.21
Gingival Index (GI)	1.45 ± 0.25	1.60 ± 0.22	0.28
Probing Depth (mm)	1.33 ± 0.35	1.40 ± 0.40	0.51
Clinical Attachment (mm)	0.80 ± 0.25	0.78 ± 0.43	0.89

Table 2. Post-treatment (14 days) periodontal parameters

Parameter	Group A	Group B	p-value
PI	1.80 ± 0.60	1.55 ± 0.28	0.34
GI	1.20 ± 0.18	1.45 ± 0.20	0.28
PD (mm)	1.15 ± 0.17	1.05 ± 0.22	0.41
CAL (mm)	0.65 ± 0.35	0.63 ± 0.33	0.87

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Table 3. Intragroup comparison of baseline vs 14-day outcomes

Parameter	Group A Pre	Group A Post	p-value	Group B Pre	Group B Post	p-value
PI	2.40 ± 0.60	1.80 ± 0.60	0.01*	1.80 ± 0.30	1.55 ± 0.28	0.03*
GI	1.45 ± 0.25	1.20 ± 0.18	0.02*	1.60 ± 0.22	1.45 ± 0.20	0.04*
PD (mm)	1.33 ± 0.35	1.15 ± 0.17	0.06	1.40 ± 0.40	1.05 ± 0.22	0.01*
CAL (mm)	0.80 ± 0.25	0.65 ± 0.35	0.10	0.78 ± 0.43	0.63 ± 0.33	0.08

Discussion

The present randomized clinical trial demonstrated that a herbal mouthwash containing *Punica granatum* and grapefruit seed extract produced significant improvements in plaque and gingival indices after 14 days, with outcomes comparable to those achieved using 0.2% chlorhexidine. Both groups showed statistically significant reductions in PI and GI, but intergroup comparisons revealed no meaningful differences, confirming the null hypothesis.

These findings are consistent with prior clinical investigations. Varughese et al. reported that pomegranate-based rinses significantly reduced plaque and gingival inflammation, with effects similar to CHX.[7] Shettar et al. demonstrated that a botanical oral rinse containing pomegranate and GSE produced gingival improvements comparable to chlorhexidine over a similar study period. Likewise, Koregol et al. observed that grape seed extract mouthwash provided antimicrobial activity against dental plaque microorganisms comparable to CHX.Collectively, these studies support the notion that herbal mouthwashes can serve as effective adjuncts in plaque control.

The observed clinical effects can be attributed to the bioactive constituents of the test rinse. Pomegranate polyphenols such as punicalagins exert antimicrobial actions by disrupting bacterial cell walls and inhibiting glucosyltransferase activity, thereby reducing plaque formation.[23] In addition, their antioxidant activity helps neutralize reactive oxygen species generated during gingival inflammation, thereby protecting host tissues.[24] Grapefruit seed extract contributes further by supplying flavonoids and vitamin C, which possess antimicrobial and anti-inflammatory properties.[25] Together, these ingredients provide a dual mechanism of action: suppression of microbial biofilm and modulation of gingival inflammation.

A key advantage of the herbal rinse was its favorable safety profile. None of the participants in Group A reported adverse effects such as staining or altered taste, which are common limitations of CHX. This aligns with systematic reviews that highlight better tolerability of herbal mouthwashes compared with conventional chemical rinses.Improved patient acceptance could translate into better long-term compliance in daily oral hygiene routines.

Nevertheless, several limitations must be considered. The study included only 30 participants and was limited to a short duration of 14 days. While short-term plaque and gingivitis reductions were evident, longer trials are needed to evaluate sustained benefits, potential effects on attachment levels, and microbial composition. Moreover, microbiological assessments were not performed in this study, which could have provided insights into specific pathogen suppression. Another limitation is the absence of a placebo group, which would have further validated the true efficacy of the test rinse.

Future research should therefore focus on multicenter randomized controlled trials with larger sample sizes, longer follow-up, and comprehensive microbiological and biochemical analyses. Investigations into patient-reported outcomes such as taste acceptability and quality of life measures are also warranted. Additionally, standardization of extraction methods and formulation concentrations for herbal mouthwashes will be essential for reproducibility and clinical application.

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