Winter 2006

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The Comparative Effects of 0.12% Chlorhexidine and Herbal Oral Rinse on Dental Plaque-Induced Gingivitis

Elizabeth N Southern, Gayle B McCombs, S Lynn Tolle and Ken Marinak

Purpose. The purpose of this study was to determine the effects of two oral rinses—one 0.12% chlorhexidine rinse (CHX) and one herbal rinse (HBR)—on gingival health status over time.

Methods. Sixty-three participants were randomly assigned to one of three treatment groups: CHX, HBR, or placebo. For three months, participants rinsed twice daily (morning and evening) with ½ ounce of allocated rinse after brushing and flossing. Individuals were given the same type of soft bristle toothbrush and whitening toothpaste. No attempt was made to modify participants' routine oral care, except they were advised to refrain from use of any other oral rinse for the duration of the study. Data were collected at baseline (B), month one (1), two (2), and three (3) utilizing the Gingival Index (GI), Plaque Index (PI), and bleeding on probing (BOP). A full mouth periodontal probing was performed at baseline and at the completion of the study. A soft tissue oral assessment was completed at each visit. CHX, HBR, and placebo data were compared between three time intervals, B-1, B-2, and B-3. Statistical analysis was conducted by means of multiple regression using generalized linear models. Paired comparison tests—ANOVA followed by a post hoc Tukey test—were used to confirm results.

Results. CHX was the only oral rinse to demonstrate a statistically significant effect on the reduction of mean GI, BOP, and PI scores when compared to placebo. CHX demonstrated a 31% reduction in the proportion of GI scores between B-2 and a 29% reduction between B-3 (p=.003 and p=.012, respectively). CHX demonstrated a 19% reduction of BOP sites between B-1, 32% reduction between B-2, and 29% reduction between B-3 (p=.028, p=.000, and p=.005, respectively). CHX demonstrated a 20% reduction in PI scores between B-1, and a 28% reduction between B-2 (p=.005 and p=.032, respectively). The effects of HBR on reducing mean GI, BOP, and PI scores were not statistically greater than placebo at any time during the study.

Keywords: Dental plaque, gingivitis, chlorhexidine, oral rinse, herbal, mouthrinse

Introduction

Bacterial plaque is the primary etiological cause of chronic gingivitis. Many patients, due to difficulty maintaining thorough oral hygiene, accumulate significant amounts of bacterial plaque containing virulent pathogens. Effective oral care is important for all individuals; it is especially important for those who are compromised due to poor dexterity, an immune system deficiency, and/or are undergoing chemotherapeutic or radiation therapy. Despite one’s best efforts, mechanical aids may fail to adequately remove plaque biofilm or “reduce the bacteria below the patient’s threshold for disease.” For these individuals, a therapeutic mouthrinse is often recommended as an adjunct to mechanical plaque control to help maintain gingival health.
A variety of oral rinses are available to consumers either by prescription (Rx) or over-the-counter (OTC). The increasing popularity of herbal or "natural" products has led companies to include these in their oral care product lines. Since herbal products may be purchased OTC, they have attracted millions of consumers who are looking for an alternative mouthrinse; however, more research is needed to determine the effectiveness and safety of these products.

To date, little research has been conducted on the comparative effects of 0.12% chlorhexidine versus herbal mouthrinses on plaque-induced gingivitis and plaque biofilm accumulations. The body of evidence pertaining to herbal products is small; therefore, tests should be conducted to gain evidence regarding their effectiveness and safety and to substantiate product claims. Fischman concluded that mouthrinse sales are closely related to whether a consumer likes the taste, smell, or color, and whether the consumer feels like the product leaves their mouth feeling fresh and clean. Depending on the effectiveness of the ingredients and patient compliance, evidence suggests that mouthrinses may act as beneficial adjunctive aids, especially for those who do not achieve optimal plaque control.

Review of the Literature

Current research indicates that plaque biofilm, a complex three-dimensional arrangement of bacteria in a self-sustaining community, has been associated with the initiation and progression of gingivitis and the onset of periodontitis. Van Dyke defined gingivitis as the "marginal inflammation of the gingiva comprising an inflammatory cell infiltrate, reversible destruction of collagen, and the clinical appearance of redness and swelling." Plaque-induced gingivitis begins at the gingival margin, and the virulent pathogens can progress throughout the gingival unit. Irreversible damage may occur when microbes migrate deeper into the epithelium. Antimicrobial agents may aid in disrupting pathogenic bacteria associated with plaque, thus aiding in the control of gingivitis. Healthy oral flora is influenced by effective plaque control; however, the pathogenic degree of the bacteria in plaque also plays a significant role, as does the host response, immune status, and amount of time that plaque remains on the tooth. Local and systemic factors should also be considered when addressing the host response in reducing the virulence of plaque biofilm since this interaction has the potential to exacerbate the gingival disease process.

Kornman states that there are three ways to treat or prevent gingivitis: 1) elimination of all clinically detectable plaque; 2) reduction of plaque below the individual's threshold for disease; and/or 3) alteration of the microbial succession in supragingival plaque. However, the depth that a mouthrinse effectively penetrates into a periodontal pocket of 1-6 mm is approximately 21% of the total pocket depth; therefore, the adjunctive use of a therapeutic oral rinse will best aid in altering the bacteria in subgingival plaque in individuals with gingivitis rather than periodontitis.

The present study focused on two specific mouthrinses: OTC herbal rinse (HBR) and 0.12% chlorhexidine rinse (CHX), both claiming to be effective in the treatment of gingivitis. Chlorhexidine (0.12%) has the American Dental Association (ADA) Seal of Acceptance and is Food and Drug Administration (FDA) approved for the reduction of plaque and gingivitis. Chlorhexidine also has long-standing research to substantiate its safety 13-16 and efficacy whereas the HBR mouthrinse used in this study is a newer, less researched product. Many studies have been conducted on 0.12% chlorhexidine mouthrinses; however, little research has been conducted to determine the efficacy of herbal mouthrinses and their ability to control plaque-induced gingivitis. Despite commonly known side effects such as temporary loss of taste; staining of the teeth, restorations, and mucosa; dry, sore mucosa; bitter taste; and a slight increase in supragingival calculus formation, CHX is considered the "gold standard" of antimicrobial rinses because of broad-spectrum antibacterial activity and substantivity of 8-12 hours. Chlorhexidine contains 11.6% alcohol, whereas the HBR tested in this study is a non-alcoholic, sugar-free product. In certain individuals whose oral health is compromised, or are recovering alcoholics, the addition of an effective therapeutic herbal oral rinse may be a valuable adjunct to brushing and flossing. Cost and accessibility are additional factors to be considered when selecting the appropriate oral rinse. The HBR and CHX mouthrinses used in this study were similar in price and quantity,
although the CHX product can only be obtained by prescription. Presently, the HBR oral rinse sells at a major pharmacy chain for $6.79 (8 oz) and the CHX product retails for approximately $15.00 (16 oz).

The manufacturer of the HBR product evaluated in this study claims it does not contribute to staining, tartar buildup, or taste alteration; possesses the ability to kill germs as effectively as the leading CHX prescription mouthrinse; and reduces gingivitis. The ingredients in the HBR include filtered spring water, vegetable glycerin, echinacea, goldenseal, calendula, aloe, bloodroot, grapefruit seed extract, citric acid, spearmint oil, peppermint oil, and cinnamon. Therapeutic benefits from these ingredients have not been extensively tested in a clinical setting. However, research conducted by Kaim et al suggests that HBR has an antimicrobial effect against A. viscosus and S. mutans in vitro. Additional research conducted by Scherer et al demonstrated HBR reduced gingival bleeding after three months of use as compared to placebo.

Methods and Materials

The protocol was reviewed and approved by the Old Dominion University Institutional Review Board. Qualifying participants signed informed consent, in duplicate, with one copy given to the participant and the other retained. Sixty-three individuals were recruited via flyers and campus emails. Individuals who met the following inclusion criteria were enrolled: GI score of 2 or 3, 18 years of age or older, and in general good health. Potential participants were excluded if they presented with advanced periodontal disease (AAP IV or greater), history of antibiotic use in the last 90 days, need for antibiotic pre-medication, or anterior facial restorations; used a daily anti-gingivitis rinse within the last 3 months; were pregnant, a smoker, or not in good general health. Participants were randomized to one of three product groups (CHX, HBR, or placebo). Individuals were identified by code numbers throughout the study.

At each appointment, the health history was reviewed and a soft tissue oral examination was performed. At the conclusion of the study, participants were evaluated for the need of a dental prophylaxis. This service was provided to participants who exhibited staining or calculus deposits attributed to the use of the study product.

A randomized design was utilized over a three-month period to evaluate the effects of 0.12% CHX, HBR, and placebo mouthrinses on gingival health and plaque accumulation. Information from product literature and previous studies suggest that the time period for initial treatment of a patient undergoing 0.12% CHX mouthrinse therapy should be limited to three months; then, the individuals’ gingival health status should be reevaluated. Numerous gingivitis studies have been conducted using an experimental period of three months or less; therefore, a three month study design was used for this study.

Data collection:

Baseline: Oral Exam (OE), Plaque Index (PI), Gingival Index (GI), Bleeding On Probing (BOP), and Probing Pocket Depth (PPD)

Month 1: OE, PI, GI, and BOP

Month 2: OE, PI, GI, and BOP

Month 3: OE, PI, GI, BOP, and PPD

In an attempt to replicate "real life" comparison group(s where a majority of consumers who purchase mouthrinse products have no recent history of a dental prophylaxis, no prophylaxis was conducted prior to study initiation. At baseline, participants were randomly assigned to one of three treatment groups: Group I (placebo), Group II (HBR), and Group III (0.12% CHX). All individuals were given the same type of soft toothbrush (Crest Complete, Procter and Gamble Co. Inc., Cincinnati, OH) and fluoride whitening toothpaste (Crest Dual Action Whitening Cool Mint, Procter and Gamble, Co., Inc. Cincinnati, OH) to decrease the possible side effects of staining and lessen examiner bias. Participants were instructed to use the assigned mouthrinse in conjunction to their normal oral hygiene routine. Product identifiers were removed from all containers. No attempt was made to modify participants' regular daily oral hygiene routine except they were advised not to use any other oral rinse for the duration of the study. Standardized written and oral rinsing instructions were provided.
Participants received periodic telephone calls to encourage protocol compliance and retention.

Data were collected using three periodontal assessment tools to assess gingival inflammation (GI), plaque accumulations (PI), and bleeding (BOP). A full mouth periodontal probing was performed at six sites per tooth (mesiofacial, facial, distofacial, mesiolingual, lingual, and distolingual) at baseline and at the end of the study using UNC-12 periodontal probes (Hu-Friedy Manufacturing Co., Inc., Chicago, IL). GI, BOP, and PI scores were obtained from the same six sites. The examiner and participants were blinded to product allocation. A single calibrated dental hygienist was used to collect all data. A research assistant was responsible for product allocation and supervising rinsing procedures. New products were dispensed at each appointment in unmarked bags and unused product was collected.

The Plaque Index (PI) was used to measure plaque accumulation. A score of 0-3 was assigned to six sites per tooth using the following criteria:\n
0 = No plaque on gingival margin.
1 = A film of plaque is supragingival, and adheres to the free gingival.
2 = Moderate plaque is present supragingivally and subgingivally.
3 = Heavy plaque is present supragingivally and subgingivally.

The Gingival Index (GI) was used to determine severity and location of gingivitis. A score from 0-3 was assigned to six sites per tooth, using the following criteria:\n
0 = Normal gingiva. Pale pink color, normal stippling, gingiva firm when probed. Gingival margin located on enamel or apical to CEJ.
1 = Mild inflammation. Slight changes in color of gingiva- more reddish than normal, slight edema. No bleeding on probing.
2 = Moderate inflammation. Gingiva is red to reddish-blue with moderate edema present and glazing. Bleeding on probing is present.
3 = Severe inflammation. Marked redness, edema, and ulceration. Tendency towards spontaneous bleeding.

Greenstein stated that bleeding on probing is an objective way to assess for clinical, bacteriologic, and histopathologic changes, since bleeding indicates areas infiltrated by inflammatory cells and histopathologic alterations due to the epithelial proliferation that occurs in early to established gingival lesions. Therefore, in the present study, BOP was independently scored as positive when bleeding was detected after 15 seconds, when stimulated by a periodontal probe.

Statistical Analysis

Statistical analysis was conducted by means of multiple regression using generalized linear models to analyze the paired difference between baseline and month one (B-1), month two (B-2), and month three (B-3). CHX and HBR were compared to placebo. Paired comparison tests, ANOVA followed by a post hoc Tukey test, were used to confirm the results. Data sets included continuous proportional data and ordinal data. Comparing the paired difference in proportion allowed participants to act as their own control. Change in proportion was calculated by subtracting the baseline proportion of unhealthy observations from the proportion of unhealthy observations following allotted months of rinsing. A negative value indicates that the proportion of scores decreased. Predictors of demographic data were analyzed separately. Pre-statistical analysis determined that age and race had an influence on GI scores; therefore, these factors were controlled for in the final data analysis. Age and race exhibited no statistically significant influence on PI or BOP. The p level was set at <0.05, a 95% confidence level, to minimize Type I error.
Results and Discussion

Sixty-three participants, 57% male (n=35) and 43% female (n=28) with a mean age of 25 years (sd=7.67), of whom 61% were Caucasian, 27% Asian, and 12% African American, were enrolled. During the course of the study, one subject withdrew from each group due to personal reasons, resulting in a final sample size of 60 (30 in each group).

Gingival Health (GI)

Randomization produced similar equivalent baseline groups that exhibited overall GI scores of 2 or 3. Analysis of GI scores indicate that CHX exhibited a statistically significant reduction when compared to placebo in the proportion of mean GI scores at B-2 and B-3 (p=.003 and .012, respectively) (Table I). These data support the work of Grossman who found a 29% reduction of gingival bleeding following three-month use of CHX. HBR did not reveal a statistically significant reduction greater than placebo in the proportion of GI scores at any time during the study. This is a conflicting comparison to the results by Scherer et al that indicated a 26.9% reduction in gingivitis following rinsing with HBR for three months.

The greatest significant decrease in the proportion of GI scores, occurred in the CHX group from B-2 as indicated by a 31% reduction. One explanation for this comparison may be examiner bias; on the other hand, participants may have been enthusiastic about the study, resulting in greater compliance to rinsing regimens, which decreased as the study progressed.

Bleeding (BOP)

CHX exhibited a statistically significant reduction in BOP greater than placebo throughout the study as indicated by the decrease in proportion of positive bleeding sites between B-1, B-2, and B-3 (p=.028, .001, and .005, respectively) (Table II). HBR did not have a statistically significant effect greater than placebo on decreasing positive bleeding sites at any point in the study. CHX showed a comparable 29% reduction of both BOP and GI scores at B-3, which is expected since bleeding and gingival inflammation are reflected in GI scores. Ultimately, CHX was the only mouthrinse that demonstrated a statistically significant effect on the reduction of bleeding when compared to placebo.
In the present study, BOP data suggest that CHX had significant clinical effects on the improvement of gingival health. Over time, CHX effectively reduced clinical evidence of gingivitis as indicated by a statistically significant reduction in GI and BOP scores. The greatest decrease in BOP was evident from B-2 in the CHX group as indicated by a 32% decrease in positive bleeding sites.

**Table II. Change in Proportion of Positive BOP Scores as Compared to Placebo Over Time**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Mean</th>
<th>B-1</th>
<th>B-2</th>
<th>B-3</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBR</td>
<td>0.160</td>
<td>-0.236</td>
<td>-0.171</td>
<td></td>
</tr>
<tr>
<td>sd</td>
<td>0.090</td>
<td>0.149</td>
<td>0.116</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>0.100</td>
<td>0.059</td>
<td>0.955</td>
<td></td>
</tr>
<tr>
<td>se</td>
<td>0.044</td>
<td>0.046</td>
<td>0.041</td>
<td></td>
</tr>
<tr>
<td>CHX</td>
<td>-0.185</td>
<td>-0.315</td>
<td>-0.291</td>
<td></td>
</tr>
<tr>
<td>sd</td>
<td>*0.172</td>
<td>*0.146</td>
<td>*0.144</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>*0.028</td>
<td>*0.000</td>
<td>*0.005</td>
<td></td>
</tr>
<tr>
<td>se</td>
<td>0.043</td>
<td>0.045</td>
<td>0.040</td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>-0.087</td>
<td>-0.147</td>
<td>-0.173</td>
<td></td>
</tr>
<tr>
<td>sd</td>
<td>0.144</td>
<td>0.138</td>
<td>0.124</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>0.006</td>
<td>0.000</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>se</td>
<td>0.031</td>
<td>0.032</td>
<td>0.029</td>
<td></td>
</tr>
</tbody>
</table>

* Bold indicates a statistically significant reduction, p=.05

In the present study, BOP data suggest that CHX had significant clinical effects on the improvement of gingival health. Over time, CHX effectively reduced clinical evidence of gingivitis as indicated by a statistically significant reduction in GI and BOP scores. The greatest decrease in BOP was evident from B-2 in the CHX group as indicated by a 32% decrease in positive bleeding sites.

**Plaque (PI)**

Only CHX had a statistically significant effect greater than placebo on reducing the proportion of PI scores of 2 or 3 (Table III). CHX demonstrated a 20% reduction in PI scores between B-1, and a 28% reduction between B-2 (p=.005 and .032, respectively). Although the CHX group demonstrated a 28% decrease in PI scores from B-3 (p= 0.098), this reduction was not significantly different from placebo. HBR did not have a statistically significant effect greater than placebo on decreasing the proportion of PI scores at any time during the study.
In the majority of cases, participants were examined at the same time of day to reduce extraneous variables in plaque accumulation, such as the length of time between home care and data collection. Variations in plaque accumulations may have been influenced by the Hawthorne effect or the tendency of participants to improve behavior because of the expectations created by the situation. On the other hand, as the study progressed, participants may have become more lax with their oral care resulting in an increase of PI scores.

**Probing Pocket Depth (PPD)**

Full mouth periodontal probing measurements were obtained at baseline and month three (B-3). Statistical analysis showed no statistically significant change in PPD in any of the groups.

**Summary and Conclusions**

The purpose of this study was to determine the comparative effects of 0.12% CHX and HBR to placebo on gingival health and plaque biofilm accumulations over time. The ability of an oral rinse to be retained in the oral cavity and maintain potency over an extended length of time has been debated. Lang stated that the substantivity of an antimicrobial agent needs sufficient contact time with a microorganism in order to inhibit or kill it. Chlorhexidine, with a substantivity of 8-12 hours, is considered to be highly effective; whereas, the substantivity of the HBR is unknown. Conclusions from the present study suggest that CHX was effective in killing virulent bacteria found in plaque biofilm, and is evident by a statistically significant reduction in the proportion of GI scores and BOP sites in the CHX group. The greatest significant decrease in the proportion of GI scores occurred in the CHX group between B-2 as indicated by a 31% reduction. There is not enough statistically significant evidence to suggest that HBR had a greater effect in reducing GI scores than placebo. During the study, extraneous variables such as length of study and busy schedules may have reduced participants' enthusiasm, resulting in less compliance to rinsing regimens, or perhaps participants who enrolled with minimal gingivitis may have exhibited healthier gingival status over less time.

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**Table III. Change in Proportion of PI Scores of 2 or 3 as Compared to Placebo Over Time**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>B-1</th>
<th>B-2</th>
<th>B-3</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBR</td>
<td>Mean</td>
<td>-0.107</td>
<td>-0.197</td>
</tr>
<tr>
<td></td>
<td>Sd</td>
<td>0.107</td>
<td>0.142</td>
</tr>
<tr>
<td></td>
<td>P value</td>
<td>0.250</td>
<td>0.306</td>
</tr>
<tr>
<td></td>
<td>Se</td>
<td>0.053</td>
<td>0.069</td>
</tr>
<tr>
<td>CHX</td>
<td>Mean</td>
<td>*-0.196</td>
<td>*-0.0275</td>
</tr>
<tr>
<td></td>
<td>Sd</td>
<td>*0.251</td>
<td>*0.286</td>
</tr>
<tr>
<td></td>
<td>P value</td>
<td>*0.005</td>
<td>*0.032</td>
</tr>
<tr>
<td></td>
<td>Se</td>
<td>0.052</td>
<td>0.068</td>
</tr>
<tr>
<td>Placebo</td>
<td>Mean</td>
<td>-0.046</td>
<td>-0.126</td>
</tr>
<tr>
<td></td>
<td>Sd</td>
<td>0.098</td>
<td>0.190</td>
</tr>
<tr>
<td></td>
<td>P value</td>
<td>0.216</td>
<td>0.012</td>
</tr>
<tr>
<td></td>
<td>Se</td>
<td>0.037</td>
<td>0.048</td>
</tr>
</tbody>
</table>

* Bold indicates a statistically significant reduction, p=.05*
Results indicate that CHX had a statistically significant effect on decreasing the paired difference in the proportion of BOP sites compared to placebo, as indicated by the decrease in positive BOP scores between B-1, B-2, and B-3. The greatest significant decrease in BOP sites was evident in the CHX group between B-2 as indicated by 31% reduction. There is not enough statistically significant evidence to suggest that HBR is more effective than placebo in its ability to reduce BOP over time.

The results of this study indicate that only CHX had a significant effect on decreasing moderate to heavy plaque at B-1 and B-2 when compared to placebo. Although the greatest reduction in PI scores occurred between B-3 for CHX, the results were not statistically different than placebo. The largest significant decrease in PI scores occurred in the CHX group between B-2 as indicated by a 28% decrease in the proportion of PI scores. A 16% reduction of PI scores between B-3 in the placebo group may be due to the Hawthorne effect as participants reached the end of the study. There was no statistically significant decrease in PI scores in the HBR group when compared to placebo at any time during the study.

Little research has been conducted on the comparative effects of herbal oral rinses and chlorhexidine oral rinses. With the proliferation of herbal oral care products, it is important for clinicians to make evidence-based decisions when making product recommendations. Research by Kaim et al suggests that there are certain ingredients in herbal oral rinses that exhibit evidence of anti-inflammatory and anti-fungal therapeutic effects. These components include echinacea, goldenseal, and grapefruit seed extract. However, research needs to be conducted to determine the substantivity of HBR, as well as determine its antimicrobial effects on gingivitis, plaque biofilm accumulation, and related bacteria.

Based on the results of this study, it is important to note that 0.12% CHX mouthrinse effectively reduced the clinical symptoms of plaque-induced gingivitis, and had a statistically significant effect on the reduction of plaque scores. Data from this study suggests that CHX is more effective than HBR in reducing clinical indicators of gingivitis when compared to placebo. Data does not support HBR product claims suggesting that it is as effective as CHX. Therefore, individuals who are looking for a “natural,” sugar-free, non-alcohol mouthrinse should be advised that there is little research to support the effectiveness of herbal oral rinses. Dental professionals will benefit from this body of evidence as they seek to make evidence-based product recommendations.

Suggestions for future studies include: (1) address the substantivity of HBR mouthrinses; (2) incorporate microbial analysis in order to draw conclusions about antibacterial effects; (3) expand study population to include broader disease status and varied age groups; (4) extend study to six months; (5) add stain and calculus indices, (6) increase internal validity by supervising rinsing; and (7) determine if the whitening toothpaste interacted with components of the mouthrinse.

Acknowledgements

Researchers are grateful to Russell Bogacki, DDS, MS, assistant professor at Virginia Commonwealth University School of Dentistry for statistical support and Deborah Z. Williams for research assistance.

Notes

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