In vivo antiplaque efficacy of combined antimicrobial dentifrice and rinse hygiene regimens

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ABSTRACT: Purpose: To evaluate using digital plaque image analysis the antiplaque efficacy of oral care regimens including use of antimicrobial toothpaste in combination with antimicrobial mouthrinse. Methods: 16 subjects completed the study protocol including: (1) initial treatment phase, all subjects used a standard sodium fluoride dentifrice with 2x/day brushing, (2) second treatment phase, subjects were randomized to two treatment groups: stannous fluoride/sodium hexametaphosphate dentifrice or sodium fluoride triclosan/copolymer dentifrice; (3) third treatment phase, the group using stannous fluoride dentifrice rinsed with alcohol-free cetylpyridinium chloride mouthrinse and the group using triclosan dentifrice rinsed with essential oil mouthrinse. During each phase, plaque levels were assessed in the morning before toothbrushing (AM), post-brushing in the morning (PB) and in the afternoon (PM). Results: Stannous fluoride dentifrice was superior to triclosan dentifrice in plaque growth inhibition between toothbrushing. Both mouthrinses provided additional plaque prevention benefits when used with antimicrobial dentifrices. The cetylpyridinium chloride mouthrinse and stannous fluoride dentifrice regimen was particularly effective, building accretive efficacy over time. Average plaque reductions exceeded 50% vs. sodium fluoride dentifrice alone. Chemotherapeutic dentifrices and rinses increase plaque control used alone and particularly in combination. The stannous fluoride-cetylpyridinium chloride regimen showed the greatest benefits. (Am J Dent 2008;21:189-196).

CLINICAL SIGNIFICANCE: This study showed that clinically proven antimicrobial toothpastes, though effective, may have efficacy further enhanced through combined use with selected mouthrinses.

Introduction

Gingivitis results from the development of microbial dental plaque infections at the gingival tooth interface initiating a host response. While specific microbial populations and pathogenicity have been correlated with disease processes, the generalized non-specific correlation between dental plaque and soft tissue disease is supported by the development of inflammation and bleeding in “experimental gingivitis” patients coupled with rapid resolution of disease upon reinstitution of hygiene measures. Although frequent and effective hygiene presents a proven and effective route to the maintenance of soft tissue health, for most patients complete plaque control is difficult with mechanical cleaning alone. As a result, gingivitis prevalence remains high, even among patients who visit the dentist routinely.

The addition of chemotherapeutic antimicrobials to dentifrices and mouthrinses presents an effective method to help patients improve their hygiene. In the United States, clinical studies have supported monograph or New Drug Application acceptance of a number of therapeutic oral products. Chemotherapeutic dentifrices with proven clinical efficacy marketed in the US include formulations containing stannous fluoride, triclosan and essential oils. Chemotherapeutic rinses with proven clinical efficacy marketed in the US include formulations based on chlorhexidine, essential oils and cetylpyridinium chloride. While both dentifrices and rinses have proven effective vehicles for the application of antimicrobials, these treatment forms may provide unique benefits. The advantage of dentifrice as a vehicle for therapeutic antimicrobials is that it is an easy, daily application which includes fluoride for control of dental caries. Toothpaste also serves to assist in the brushing regimen with surfactants in the removal of plaque. Despite these positives, dentifrice is a complicated vehicle for chemotherapeutic delivery in vivo. For example, a dentifrice is diluted significantly during use with saliva (a factor of 3-5 x by weight) and during the short time of application (the mean toothbrushing time is 40 seconds to 1 minute) patients may rinse or expectorate solubilized active ingredients before they can work. In addition, the high foaming properties of dentifrices which are popular with consumers can theoretically interfere with the penetration of the toothpaste ingredients. In contrast to dentifrices, mouthrinses present a very efficient delivery vehicle for therapeutic antimicrobials. Rinses are only marginally diluted on use (usually less than 20% by weight) and do not typically foam to the same degree as dentifrices during use. The volume of mouthrinse comfortably used is much larger than volume generated from toothpaste dilution with saliva. These combined properties make rinses efficient for delivery of therapeutics with particular focus on the penetration of ingredients between teeth.

The unique properties of rinses and pastes would suggest that they might be recommended as a combined regimen to achieve optimal clinical effects. Many patients already routinely use multiple oral hygiene products. For example, US surveys show that more than 95% of subjects report brushing their teeth at least once per day. These studies also report that up to 1/3 of patients floss in addition to toothbrushing and another 20-30% report that they routinely use mouthrinses. Despite consumer use of combinations of oral hygiene products, there are surprisingly few studies evaluating combined benefits of toothpaste and mouthwash antimicrobial regimens in providing
dental plaque control and gingival health protection. This study evaluated the effects of two chemotherapeutic dentifrice plus mouthrinse regimens on the prevention of dental plaque formation in vivo. One combination regimen included a commercial NaF/Gantrez/triclosan dentifrice combined with a commercial mouthrinse containing a fixed ratio of essential oils. This combination includes the over-the-counter antimicrobial dentifrice and mouthrinse with the longest history of U.S. use and which has been communicated directly to dental professionals and patients (http://www.listerineandcolgate.com/app/ListerineAnd Colgate/US/HomePage/OfferEnded.cvsp). The second treatment combination included two relatively new treatments available to professionals and patients, a stannous fluoride–sodium hexametaphosphate dentifrice paired with an alcohol-free cetylpyridinium chloride mouthrinse. These products were developed by the same manufacturer and are marketed under the Crest ProHealth product line.

**Materials and Methods**

**Subject qualifications** - Subjects entering the protocol were employed adults who participated in previous plaque testing at the Mason research facility. The use of the digital plaque image analysis (DPIA) methodology has been reviewed and approved by an Institutional Ethics Review Committee. Subjects signed informed consent for participation in the study. Subjects met the following entrance criteria:

1. Agreement not to use any other oral hygiene products than those assigned during the study, including mouthrinses, toothpastes, whitening or therapeutic chewing gums, or whitening formulations, etc. For subjects who floss regularly, they were allowed to floss between the posterior teeth only throughout the study. Subjects were asked to avoid chewing gum unless it was a daily habit which should be consistently maintained throughout the study.

2. No dental visits were planned for the duration of study participation;

3. No recent (2 weeks prior to the study) use of antibiotics;

4. No special dietary restrictions and/or diet adjustment expected during the trial;

5. No “color matched” restorations on facial surfaces of the anterior teeth;

6. No participation in other studies;

7. Good general health and good overall dental health (as self assessed and confirmed by subject disclosure that they have routine dental visits);

8. No general allergy problems or specific allergies to dyes;

9. Sufficient plaque levels during a pre-screening visit;

10. No pregnancy or nursing;

11. No adverse reactions to currently marketed oral care products.

**Test products** - The assigned toothpaste for Treatment Period 1 was standard Crest Cavity Protection Dentifrice (NaF, silica abrasive, regular flavor). This paste uses sodium fluoride at Food and Drug Administration monograph levels for cavity protection, standard abrasives, conventional levels of surfactant and preservatives and no specialized tartar control or antimicrobial ingredients. Accordingly, use of this paste could be considered to provide a typical standard of hygiene care. The dentifrices used in Treatment Period 2 were Crest Pro-Health for Group 1 and Colgate Total for Group 2. Crest Pro-Health dentifrice contains 0.454% stannous fluoride complemented with sodium hexametaphosphate (antitartar ingredient). Colgate Total dentifrice is a 0.243% sodium fluoride paste which contains triclosan antimicrobial delivered in a mixture with a copolymer of maleic anhydride/methyl vinyl ether (Gantrez Acid). Both pastes were supplied in overwrapped (blind) commercial packaging with study labeling containing instructions. For Treatment Period 3, Group 1 subjects added twice daily rinsing with Crest Pro Health mouthrinse and Group 2 twice daily rinsing with Listerine mouthrinse. Crest ProHealth is an alcohol-free therapeutic mouthrinse containing 0.07% cetylpyridinium chloride as active ingredient in an alcohol-free yet highly bioavailable formulation. Listerine is an alcohol based therapeutic mouthwash containing a fixed combination of essential oils including thymol, menthol, eucalyptol and methyl salicylate. Rinses were supplied in commercial bottles with over-labeling. Toothbrushes assigned for home use throughout the study were Oral-B 40 standard brushes. For supervised clinic brushing, subjects were provided disposable manual test brushes.

**Study schedule** - This study followed a randomized parallel group design with three treatment periods, preceded by 1 week of acclimation where the subjects used Crest Cavity Protection and a regular soft Oral-B 40 toothbrush. During the first treatment period subjects continued using Crest Cavity Protection dentifrice. Subjects brushed with assigned dentifrice each morning and before bed for 1 week. Prior to the second treatment period subjects were randomized based upon Period 1 plaque levels to one of the two treatment groups. The first group started brushing morning and evening with the stannous fluoride hexametaphosphate dentifrice (Crest ProHealth), the second group with triclosan containing dentifrice (Colgate Total). In the third treatment period, the group using stannous fluoride dentifrice started to rinse additionally twice a day, after the morning and evening brushing, with 20 ml of cetylpyridinium chloride mouthrinse. The group using triclosan dentifrice likewise rinsed with 20 ml of essential oils mouthrinse, again after the morning or evening brushing. Subjects were instructed not to rinse out the mouth with water and not to eat or drink for a minimum of 30 minutes after mouthrinse use. Treatment Period 3 lasted for 3 weeks. During each treatment week, subjects reported to the DPIA imaging lab for 3 days of replicate plaque evaluations. Each imaging day included assessment of plaque level in the morning before morning brushing (AM), post-brushing (PB) and again in the afternoon (PM). The study logistics are highlighted in Fig. 1.

**Dental plaque assessments**

**Subject instruction and imaging procedure** - For plaque evaluations, subjects were instructed to report to the imaging laboratory on the “grading morning” prior to any food/beverages and without oral hygiene. DPIA data collection followed standard procedures described previously. Subjects disclosed their plaque with fluorescein solution by carrying out a rinsing sequence including:

1. Rinse for 10 seconds with 25 ml of phosphate buffer.
2. Rinse for 1 minute with 5.0 ml of 1240 ppm fluorescein in phosphate buffer.
3. Rinse 3 x 10 seconds with 25 ml of phosphate buffer.
The phosphate buffer was comprised of 3.62 grams of mono-
sodium phosphate and 0.349 grams of disodium phosphate
diluted to 2 liters with deionized water. The final pH of this
mixture was 5.5. The solution was prepared fresh each day. The
buffer rinse was used to temporarily stabilize the pH of the
mouth and to ensure reproducible dye fluorescence.

The imaging system used in this study was similar to the
one described in detail previously by Sagel et al. It consisted
of two Balcar long wave UV flash units (model FX60) and
Balcar powerpack equipped with filters to remove the majority
of visible light. The filters had a peak light pass at 365 nm. The
flash units were positioned on either side of a chin rest at a 45
degree angle to reduce reflection to a minimum. The centrally
mounted camera (digital Fuji HC2500 3CCD, fitted with a
Fujinon lens A4x7.5BMD f2.8, 7.5-30 mm), was entirely
computer controlled. Figure 2 shows the system configuration.

Image analysis - Captured images were analyzed and classified
objectively using discriminant analysis to statistically classify
pixels into different anatomical categories (e.g. teeth, gums,
plaque on gums, plaque on teeth). Image pixels were collected
and assigned to respective designations. The analysis includes the ratio of plaque on teeth pixels to the total tooth pixels representing a percent “area” coverage estimate for each image. The computer designated plaque area is used to calculate a percentage dentition coverage estimate for the facial plaque image, which is then compared for treatment effects.

Statistical analysis - The efficacy analysis consisted of averaging on a per subject basis the 3 days of percentage plaque coverage measurements on teeth by treatment period, separately for AM, PB and PM images. A paired difference t-test was used to compare plaque coverage between weeks for each treatment group. ANCOVA was used to compare the plaque coverage between the treatment groups for each week using the Week 1 plaque level as a covariate. All statistical comparisons were two-sided using a 0.05 significance level.

Results

Seventeen subjects started the study and 16 subjects completed all 5 weeks and treatment periods, nine in the first group and seven in the second group. No product related side effects were reported. The baseline (Week 1) AM pre-brushing and PM plaque averages were approximately 14.5% and 13.7% respectively. Groups were balanced at baseline with no statistically significant differences at any measurement time (AM, PB, or PM). Tooth-brushing produced a 49.9% reduction in average morning plaque level (pre-brush vs. post-brush) for the group later assigned to stannous fluoride and 48.0% for the group later assigned to triclosan dentifrice. These statistically significant (P< 0.05) plaque reductions produced by brushing are similar to results seen in our previous research and help validate the DPIA methodology.19

Table 1 shows results from Period 2 where antimicrobial paste treatments were compared for efficacy versus baseline measurements obtained in Period 1. Stannous fluoride paste users showed significant reductions in mean dental plaque vs. baseline for both AM and PM plaque level. The AM plaque reduction was 18% and PM was 27% respectively. The difference in post-brush plaque level for the stannous fluoride dentifrice was 5.8% and was not significantly different from Period 1. Plaque levels in the triclosan dentifrice group did not show statistically significant (or numerically positive) changes for AM pre-brush, AM post-brush or PM plaque levels relative to Period 1. Brushing efficacy remained at 42% and 46% for stannous fluoride and triclosan dentifrice groups respectively.

Table 1 shows results from Period 3 of the study (Weeks 3-4-5) where the subjects were additionally using rinse. Plaque levels were again compared to the baseline Week 1 results. Users of stannous fluoride paste and cetylpyridinium chloride rinse showed significant reductions in plaque on the teeth in the AM, PM and now also PB rinse showed significant reductions in mean dental plaque levels in Period 1. The magnitude of efficacy was numerically larger, with a 48-64% reductions for AM, 57-69% for PM and 20-39% for PB. Interestingly, the antiplaque effects for the stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse regimen in-
### Table 2. Comparisons to Week 2 – Period 2 (% plaque coverage).

<table>
<thead>
<tr>
<th>Visit/Time</th>
<th>Treatment</th>
<th>N</th>
<th>Week 2-Period 2 Mean (SD)</th>
<th>Visit Mean (SD)</th>
<th>Δ* Mean (SD)</th>
<th>Percent reductiona</th>
<th>P valuesb</th>
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</thead>
<tbody>
<tr>
<td><strong>Week 3 – Period 3</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Pre-brushing</td>
<td>SnF₂ + CPC</td>
<td>9</td>
<td>11.3 (4.7)</td>
<td>7.2 (3.3)</td>
<td>-4.1 (2.1)</td>
<td>36.5</td>
<td>0.0003</td>
</tr>
<tr>
<td></td>
<td>Triclosan + EO</td>
<td>7</td>
<td>15.9 (4.0)</td>
<td>8.9 (2.3)</td>
<td>-6.9 (3.0)</td>
<td>43.8</td>
<td>0.0009</td>
</tr>
<tr>
<td>Post-brushing</td>
<td>SnF₂ + CPC</td>
<td>9</td>
<td>6.5 (3.2)</td>
<td>5.6 (2.1)</td>
<td>-1.0 (1.7)</td>
<td>14.8</td>
<td>0.1298</td>
</tr>
<tr>
<td></td>
<td>Triclosan + EO</td>
<td>7</td>
<td>8.6 (4.3)</td>
<td>6.7 (3.1)</td>
<td>-1.9 (1.9)</td>
<td>22.4</td>
<td>0.0363</td>
</tr>
<tr>
<td>Afternoon</td>
<td>SnF₂ + CPC</td>
<td>9</td>
<td>10.3 (5.2)</td>
<td>6.1 (2.0)</td>
<td>-4.2 (3.5)</td>
<td>40.7</td>
<td>0.0075</td>
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<td></td>
<td>Triclosan + EO</td>
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<td>13.1 (5.4)</td>
<td>8.1 (3.6)</td>
<td>-5.0 (3.0)</td>
<td>38.0</td>
<td>0.0049</td>
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<tr>
<td><strong>Week 4 – Period 3</strong></td>
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<td></td>
<td></td>
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<td>38.0</td>
<td>0.0049</td>
</tr>
</tbody>
</table>

* = Change from week 2 Period 2

a = Positive value for percent reduction was indicative of a decrease in plaque relative to Week 2 – Period 2.

b = Two-sided P-value for the comparison to Week 2 - Period 2 using a one-sample t-test.

NaF = Sodium fluoride control dentifrice; SnF₂ = Stannous fluoride sodium hexametaphosphate dentifrice; Triclosan = NaF triclosan dentifrice; CPC = alcohol-free cetylpyridinium chloride mouthrinse; EO = essential oil mouthrinse.

### Table 3. Treatment comparisons using ANCOVA (% plaque coverage).

| Visit/Time   | Treatment                  | N  | Week 1-Period 1 Mean (SE) | Visit adjusted Mean (SE) | Treatment difference (SE) | Percent reductiona | Two-sided P-valuesb |
|--------------|----------------------------|----|---------------------------|--------------------------|                         |                   |                   |
| **Week 2 – Period 2** |                            |    |                           |                          |                          |                   |                   |
| Pre-brushing | SnF₂                      | 9  | 13.8 (2.2)                | 11.8 (0.6)               | -3.4 (1.0)              | 22.4              | 0.0041            |
|              | Triclosan                 | 7  | 15.5 (1.6)                | 15.2 (0.7)               |                         |                   |                   |
| Post-brushing| SnF₂                      | 9  | 6.9 (1.1)                 | 7.0 (0.4)                | -1.0 (0.7)              | 12.5              | 0.1593            |
|              | Triclosan                 | 7  | 8.0 (1.4)                 | 8.0 (0.5)                |                         |                   |                   |
| Afternoon    | SnF₂                      | 9  | 14.2 (2.6)                | 9.9 (0.8)                | -3.8 (1.2)              | 27.6              | 0.0085            |
|              | Triclosan                 | 7  | 12.9 (1.3)                | 13.7 (0.9)               |                         |                   |                   |
| **Week 3 – Period 3** |                            |    |                           |                          |                          |                   |                   |
| Pre-brushing | SnF₂ + CPC                | 9  | 13.8 (2.2)                | 7.5 (0.6)                | -1.0 (0.9)              | 12.1              | 0.2710            |
|              | Triclosan + EO            | 7  | 15.5 (1.6)                | 8.5 (0.7)                |                         |                   |                   |
| Post-brushing| SnF₂ + CPC                | 9  | 6.9 (1.1)                 | 5.9 (0.4)                | -0.4 (0.7)              | 6.9               | 0.5351            |
|              | Triclosan + EO            | 7  | 8.9 (1.4)                 | 6.3 (0.5)                |                         |                   |                   |
| Afternoon    | SnF₂ + CPC                | 9  | 14.2 (2.6)                | 6.0 (0.7)                | -1.4 (1.0)              | 28.8              | 0.0385            |
|              | Triclosan + EO            | 7  | 12.9 (1.3)                | 8.4 (0.8)                |                         |                   |                   |
| **Week 4 – Period 3** |                            |    |                           |                          |                          |                   |                   |
| Pre-brushing | SnF₂ + CPC                | 9  | 13.8 (2.2)                | 5.9 (0.7)                | -1.5 (1.0)              | 20.4              | 0.1530            |
|              | Triclosan + EO            | 7  | 15.5 (1.6)                | 7.4 (0.7)                |                         |                   |                   |
| Post-brushing| SnF₂ + CPC                | 9  | 6.9 (1.1)                 | 4.8 (0.4)                | -0.7 (0.7)              | 12.4              | 0.3270            |
|              | Triclosan + EO            | 7  | 8.0 (1.4)                 | 5.5 (0.5)                |                         |                   |                   |
| Afternoon    | SnF₂ + CPC                | 9  | 14.2 (2.6)                | 4.9 (0.7)                | -1.0 (0.7)              | 31.5              | 0.0481            |
|              | Triclosan + EO            | 7  | 12.9 (1.3)                | 7.1 (0.8)                |                         |                   |                   |
| **Week 5 – Period 3** |                            |    |                           |                          |                          |                   |                   |
| Pre-brushing | SnF₂ + CPC                | 9  | 13.8 (2.2)                | 5.1 (0.7)                | -1.4 (1.0)              | 20.4              | 0.1530            |
|              | Triclosan + EO            | 7  | 15.5 (1.6)                | 7.2 (0.8)                |                         |                   |                   |
| Post-brushing| SnF₂ + CPC                | 9  | 6.9 (1.1)                 | 4.4 (0.5)                | -0.5 (0.7)              | 11.5              | 0.1538            |
|              | Triclosan + EO            | 7  | 8.0 (1.4)                 | 5.6 (0.5)                |                         |                   |                   |
| Afternoon    | SnF₂ + CPC                | 9  | 14.2 (2.6)                | 4.3 (0.8)                | -2.6 (1.2)              | 37.8              | 0.0432            |
|              | Triclosan + EO            | 7  | 12.9 (1.3)                | 6.9 (0.9)                |                         |                   |                   |

a = Means adjusted using the Week 1 plaque value.

b = Two-sided P-value for the treatment comparison of the difference in mean plaque value.

NaF = Sodium fluoride control dentifrice; SnF₂ = Stannous fluoride sodium hexametaphosphate dentifrice; Triclosan = NaF triclosan dentifrice; CPC = alcohol-free cetylpyridinium chloride mouthrinse; EO = essential oil mouthrinse.

Increased numerically with time of use e.g. Week 3, to 4 to 5. Users of the essential oil mouthrinse and triclosan dentifrice also showed plaque reductions in Period 3. The essential oil-triclosan regimen showed percentage reductions averaging from 42-52%,
37-47% and 17-28% and for AM, PM and PB plaque respectively. Differences were significant *versus* baseline, except for the PB score at Week 3 (Week 1 of the regimen) where *P*= 0.08. The essential oil-triclosan regimen also appeared to increase in activity over time but the effect was not numerically as substantial as for the stannous fluoride-cetylpyridinium chloride regimen.

Table 2 shows results from Period 3 (Weeks 3-4-5) now compared to the antimicrobial dentifrice Period 2 scores. Thus, we are looking at additive chemotherapeutic effects of mouthrinses to the antimicrobial dentifrices. Users of cetylpyridinium chloride rinse showed significant additive reductions in plaque on teeth in AM, PM and now also PB *vs.* use of the stannous fluoride paste alone in Period 2. The magnitude of efficacy was 36-56% reduction for AM, 41-58% for PM and 15-36% for PB. Again, the magnitude seemed to increase with time as effectiveness *vs.* baseline increased from Week 3, to 4 to 5. Users of the essential oil rinse also showed plaque reductions as compared with the use of the triclosan dentifrice. The essential oil rinse addition to the combination showed efficacy of 44-53% reduction in the AM, 22-33% for PB and 38-48% for PM plaque respectively. All therapeutic additions in effectiveness provided by rinses were statistically significant (*P*< 0.05) except for the Week 5 PB for cetylpyridinium chloride rinse (PB Week 1 *P*= 0.13).

Table 3 presents analysis comparing antiplaque efficacy of stannous fluoride dentifrice and triclosan dentifrice from Period 2 (Week 2) of the study. Users of stannous fluoride dentifrice showed significant reductions in dental plaque in AM, and PM evaluations with average reductions *vs.* the triclosan dentifrice of 22-28%. Post-brushing plaque levels were about 12% improved for stannous fluoride dentifrice users as compared with triclosan dentifrice users though this difference was not statistically significant (*P*= 0.16).

With respect to treatment regimens, Table 3 shows analyses comparing antiplaque efficacy of stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse as compared to triclosan dentifrice–essential oil mouthrinse for Period 3 (Weeks 3-5 of the study). Results showed a plaque preventive therapeutic advantage for subjects using the stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse regimen as opposed to users of the triclosan dentifrice–essential oil mouthrinse regimen. Treatment advantages for stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse numerically increased from Weeks 3-5 for AM, PM and PB scores. Differences in plaque development at the PM measuring point were statistically significantly different favoring the stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse combination throughout Period 3, *viz.* all Weeks 3-5. Differences in AM plaque numerically increased from Weeks 3-5 (12–20–30%) almost reaching significance at Week 5 (*P*= 0.06). Differences in PB plaque levels for stannous fluoride dentifrice–cetylpyridinium chloride mouthrinse combination numerically increased from Weeks 3-5 (7–12–21%).

A unique feature of these results was the apparent increase in efficacy of the treatment combinations over time. The study design of a 1-week period of dentifrice use followed by 3 weeks use of combined mouthrinse paste regimen was specifically chosen to improve the potential to see incremental bene-

**Discussion**

Dental plaque formation on the dentition provides a toxic challenge to adjacent gingival tissues prompting the initiation of gingivitis.1,5 Left untreated, inflamed and bleeding gingiva may progress to more serious periodontal conditions over time.2,21 The prevalence of gingivitis remains high in North America even in so called “healthy” demographic groups and is even more pronounced in need groups.3,20 Along with toothbrushes, a variety of chemotherapeutic toothpastes and mouthrinses are commercially available today which may provide improvements for the control of dental plaque formation and subsequent gingivitis. Stannous fluoride sodium hexametaphosphate dentifrice has shown significant clinical efficacy in the prevention of gingivitis and gingival bleeding, complementing a range of other clinical actions.22-24 Triclosan-copolymer dentifrice has likewise shown significant efficacy for control of plaque gingivitis and gingival bleeding.25,26 Alcohol free cetylpyridinium chloride mouthrinse has shown clinical efficacy for the prevention of gingivitis when used as an adjunct to daily toothbrushing.27-29 Essential oil mouthrinse has over a 100-year period of use and has clinically proven benefits for the control of dental plaque and gingivitis.29,32 Although professionals routinely recommend combinations of brushes, pastes, dental floss and rinses to their patients, few studies have examined the effects of combined regimens on health parameters. The available literature does suggest that therapeutic addi-

**Fig. 3.** Average plaque levels for subjects assigned to stannous fluoride dentifrice and cetylpyridinium chloride mouthrinse for Periods 1-3 of the study.
tion of treatment combinations may provide additive benefits to patients in the control of plaque and gingivitis. This study assessed plaque inhibitory effects of two antimicrobial treatment regimens: stannous fluoride dentifrice combined with cetylpyridinium chloride mouthrinse and triclosan dentifrice combined with essential oil mouthrinse.

The study results showed that stannous fluoride dentifrice was more effective for plaque inhibition than a triclosan dentifrice. The improved efficacy of stannous fluoride dentifrice to triclosan dentifrice in plaque prevention seen here is in agreement with limited studies comparing gingivitis effectiveness of these formulations, although both the cetylpyridinium chloride and essential oil mouthrinses provided enhanced activity to the antibacterial dentifrices. Overall, the antiplaque effectiveness of the stannous fluoride dentifrice combined with cetylpyridinium chloride mouthrinse appeared to be greater than the essential oil triclosan combination, though both were highly and significantly effective.

The DPIA study design permitted some unique comparisons by virtue of the collection of multiple daily plaque assessments at different times and longitudinally throughout treatment. At the very least, the combined features of DPIA provides an internal benchmark of clinical relevance since the treatment. At the very least, the combined features of DPIA provides an internal benchmark of clinical relevance since the concurrent measurement of pre- and post-brushing plaque levels helps to both validate the method and also investigate chemotherapeutic effects. With respect to the former, toothbrushing should provide plaque removal benefits of 30-60% based upon previous literature and indeed in this study brushing effects measured some 50% while using control dentifrice and antimicrobial dentifrice. In terms of dimensioning clinical effects, the comparative measures of plaque levels in regrowth periods (viz. AM pre-brushing, PM) to post-brushing levels suggest that combining chemotherapeutic regimens of stannous fluoride dentifrice and cetylpyridinium chloride mouthrinse provide substantial clinical effects, keeping plaque levels below those associated with regular hygiene overnight and through the afternoon. To our knowledge, this is the first time that chemotherapeutic effects have been contrasted in this manner and this brings particular relevance to practitioners and patients on the benefits of combined treatment regimens.

A large number of patients use a combination of oral hygiene products though these usually include toothbrushing with fluoridated dentifrice. In many instances practitioners make recommendations on the selection of antimicrobial toothpaste brands or alternatively the additional use of hygiene aids such as chemotherapeutic mouthrinses. The results shown here suggest that hygiene regimens may provide significant advantages to patients, in particular in combining low cost over the counter therapeutic pastes and rinses. Overall results show that chemotherapeutic dentifrices and rinses offer value to plaque control used alone and especially in combination. The relative impact of plaque reductions on oral health of patients obviously depends upon consistency in application of hygiene measures and also disease susceptibility. These results suggest further long term studies on combined benefits of chemotherapeutic and even mechanical hygiene modalities in various populations to help define treatment recommendations to assist patients in the longitudinal maintenance of their gingival health.

Acknowledgement: To Ms. Kathy M. Kozak for her skilled efforts of in study coordination logistics and digital plaque image analysis.

Conflict of interest and sources of funding: All authors are full-time employees of Procter & Gamble. No external funding, apart from the support of the authors’ institution, was available for the study.

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