# Effects of an Ascorbic Acid–Derivative Dentifrice in Patients With Gingivitis: A Double-Masked, Randomized, Controlled Clinical Trial

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**Background:** Reactive oxygen species might be associated with the onset and progression of gingival inflammation. The aim of this study is to investigate the effect of a dentifrice containing L-ascorbic acid 2-phosphate magnesium salt (APM), a long-acting ascorbic acid derivative with antioxidant properties, on gingival inflammation.

**Methods:** The clinical effects of APM were investigated in a multicenter, randomized, parallel-group, controlled clinical trial comprising 300 individuals with gingivitis. Half of the participants were given an APM-containing dentifrice and half were given a control dentifrice. The primary outcome was the gingival index (GI) at 3 months. Secondary outcomes included gingival redness as an indicator of the degree of local gingival inflammation, gingival bleeding as a measure of the gingivitis severity index, and total antioxidant activity of the saliva.

**Results:** Under the intent-to-treat analysis, GI did not significantly differ between the groups ($P = 0.12$). However, under the per-protocol analysis, GI was significantly lower in the APM group ($P = 0.01$) than in the control group. In the APM group, gingival redness was significantly lower, and the difference from the baseline gingivitis severity index was significantly greater ($P = 0.04$ and $P = 0.02$, respectively). The total antioxidant activity of the saliva was significantly higher in the APM group ($P = 0.03$). The incidence of adverse events did not significantly differ between the groups ($P > 0.15$).

**Conclusion:** These findings indicate that the regular application of an APM-containing dentifrice could reduce gingival inflammation. *J Periodontol 2015;86:27-35.*

**KEY WORDS**

**Antioxidants; controlled clinical trials, randomized; dentifrices, ascorbic acid; gingivitis; reactive oxygen species; saliva.**

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The local immune response in periodontal tissues plays an important role in preventing the onset and progression of periodontal disease. However, continuous and/or excessive immune responses are a primary factor in periodontal tissue breakdown. The incidence and progression of periodontal diseases are associated at least in part with alterations in the polymorphonuclear leukocyte (PMN) defense system, as well as with bacterial infection and proliferation.[1] PMNs release reactive oxygen species (ROS) during phagocytosis to eliminate bacteria. However, it is well known that PMNs in patients with periodontal disease have decreased phagocytic ability and produce ROS in excess.[2-5] This imbalance in the defense system is involved in the process of periodontal tissue breakdown.[6]

Antioxidant enzymes in saliva such as peroxidase, catalase, and superoxide dismutase, as well as low molecular weight antioxidants such as uric acid and vitamins, play central roles as ROS scavengers.[7] The total antioxidant activity (TAO) of saliva is lower in patients with periodontitis than in healthy individuals,[1] but appropriate periodontal treatment can increase TAO.[8] Thus, salivary antioxidant levels may be closely

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associated with the onset and progression of periodontal disease. The authors postulated that maintaining the oxidant/antioxidant balance is important for periodontal health and can reduce the onset and rate of periodontal disease progression.

Ascorbic acid (AA) is a major antioxidant that performs several functions. The initial signs of an inflammatory response, such as gingival redness and swelling, increase when dietary intake of AA is insufficient.[9] These gingival changes have been attributed to blood vessel damage caused by deficient collagen production and collagen degradation. Some studies have shown an inverse relationship between plasma AA levels and the prevalence of periodontitis.[10-12] However, there is no clear evidence that local application of AA is effective in improving periodontal health.[13]

L-Ascorbic acid 2-phosphate magnesium salt (APM), a long-acting derivative of AA that is hydrolyzed by endogenous phosphatases, has antioxidant and immunomodulatory properties. This derivative has a higher affinity for skin than AA and thus is more effective than AA in terms of cellular uptake, antioxidant activity, and ability to stimulate collagen synthesis and fibroblast proliferation in tissues.[14,15] The authors have found that APM enhances the bacterial phagocytosis of PMNs (unpublished observations). It has been reported that APM has high penetration of skin and mucosal membranes compared with AA and enhances AA concentration in tissues.[16] APM has not been used clinically for oral healthcare, so this clinical study is the first to investigate the effects of APM on gingivitis. The authors have previously tested the efficacy of APM at reducing inflammation in animal models and found that it reduces the inflammation caused by ROS in guinea pig skin[17] and inhibits gingival inflammation in beagle dogs. These results indicate that topical application of APM might reduce gingival inflammation in humans. The authors thus conducted an exploratory clinical trial on 33 patients with gingivitis at two facilities (approved by the institutional review boards of Osaka University, Nihon University School of Dentistry at Matsudo, and Lion Corporation). This trial showed that a dentifrice containing 0.3% APM decreased GI scores (unpublished observations). The authors therefore extended the research by conducting a multicenter, randomized controlled trial on the clinical effect and safety of a dentifrice containing APM in individuals with gingival inflammation.

## MATERIALS AND METHODS

This study is designed in accordance with the guidance for evaluation of gingivitis published by the U.S. Food and Drug Administration (FDA).[18] This 3-month multicenter, double-masked, randomized controlled trial was conducted according to the Guidelines for Good Clinical Practice. The Consolidated Standards of Reporting Trials (CONSORT) checklist is available as supporting information. Study protocols were approved by the institutional review boards of Lion Corporation and the respective participating facilities before initiation (Lion Institutional Review Board approval number 81). The ClinicalTrials.gov identifier is NCT02102295.

### *Sample Size Calculation*

The sample size necessary to detect a reduction in gingival inflammation, calculated from an exploratory 3-month clinical trial of 33 individuals (difference: 0.13; SD: 0.33; power calculation: 0.8) was 240 individuals. Allowing a margin for dropouts, the initial clinical cohort comprised 300 participants.

### *Patients*

This trial was conducted at the following dental facilities: Osaka University Dental Hospital, Nihon University Dental Hospital, Nihon University Hospital School of Dentistry at Matsudo, and Tohoku University Dental Hospital (Table 1). The authors explained details of the trial to 305 patients, who subsequently provided written informed consent to participate. The authors registered 300 patients (152 males and 148 females, aged 20 to 64 years; mean: 38.5 ± 10.2 years) who met the following inclusion criteria: 1) aged 20 to 64 years (inclusive); 2) ≥16 permanent teeth; and 3) a mean baseline gingival index[19] (GI) ≥0.5 without severe gingival inflammation. Severe gingival inflammation was determined when patients were deemed upon an oral inspection to require professional therapy based on periodontal symptoms such as severe swelling, redness, bleeding, and pus discharge in the gingiva. The exclusion criteria included: 1) treatment with antibiotic medication within 1 month before the trial; 2) participation in any other clinical trial; 3) women who were pregnant or of childbearing potential; 4) use of medication that could influence gingival tissue; and 5) being judged unsuitable for this study by the examiner. According to FDA guidelines, patients who took antibacterial and/or anti-inflammatory drugs during the trial were excluded from the intent-to-treat (ITT) population. The registered participants were randomly assigned to two groups that received a dentifrice with or without APM.

### *Randomization*

All examiners, patients, and suppliers were masked to the group assignment. A randomization code number was assigned to each patient by a contract research organization¶ using permuted-block randomization, and no information was revealed until the trial ended.

¶ CMIC, Tokyo, Japan.

**Table 1.**

## Dental Facilities and Examiners

| Facility | No. of Examiners | No. of Patients |
|---|---|---|
| Osaka University Dental Hospital | 11 | 80 |
| Nihon University Dental Hospital | 10 | 80 |
| Nihon University Hospital, School of Dentistry at Matsudo | 5 | 80 |
| Tohoku University Dental Hospital | 9 | 60 |

*Test Dentifrices*

One group received a test dentifrice containing 0.3% APM[#] and 950 ppm fluoride. The other group received the same dentifrice without APM (control). The dentifrices were indistinguishable in terms of appearance, flavor, and packaging. Test dentifrices and toothbrushes** (standard type with regular bristles) were provided to the patients at the start of the trial (0 months) and at a 1-month review.

*Procedure*

Thirty-five examiners were involved in this study (Table 1). The authors held meetings with every examiner from each of the participating facilities. In addition, an explanatory meeting for all examiners was held at each facility before the trial began. At these meetings, the trial protocol was confirmed, and clinical evaluations were standardized among facilities. At all facilities, SK explained the detailed methods to all examiners. To maximize inter- and intra-examiner reproducibility, the authors trained all examiners to the same scoring standards using standardized photographs of gingival tissue and by direct examination of the gingival tissues of volunteers. All examiners repeated measurements of clinical scores of several volunteers, and the concordance rate was calculated. The authors continued until the rate of concordance was >90%. Regarding measurement of redness, the authors standardized the evaluation of a range of color changes. Finally, each examiner reconfirmed their understanding of the clinical criteria using case photos immediately before every inspection. Each participant was assigned to an examiner and was inspected by that person throughout the trial: at baseline, 1 month, and 3 months. Unstimulated whole saliva was collected before each clinical examination.

The registered participants were instructed to brush their teeth twice a day in their customary manner using the toothbrushes provided. They were also cautioned not to use any other oral hygiene products, such as mouthwash or dental floss, during the test period, and to fast for 1 hour before each clinical examination.

*Outcome Measures*

The primary outcome was GI at 3 months to assess gingival inflammation. Secondary outcomes included gingival redness as an indicator of the degree of local chromatic changes in the gingiva and gingival bleeding as a measure of the gingivitis severity index (GSI). The TAO of saliva was assessed as the ferric reduction ability of plasma (FRAP). The examiners confirmed GI and redness by comparison with evaluation criteria using a scoring manual with standard photographs at each clinical inspection. All clinical parameters were assessed by the same examiners throughout the trial. The authors standardized the sequence as follows: 1) saliva collection, 2) examination of GI, and 3) examination of gingival redness. The GSI score was calculated using GI scores.

**Clinical scores.** Based on the results of the exploratory clinical trial in which the authors evaluated mesio-buccal areas of all teeth, the authors judged that for the GI and GSI scores, the mesio-buccal areas of all teeth as representative teeth when the authors examined 300 patients (including dropouts) were statistically evaluable. Gingival inflammation was assessed using the following scores: 0 = no inflammation; 1 = mild inflammation with a slight change in color, little change in texture, and no gingival bleeding on probing (BOP); 2 = moderate inflammation with moderate glazing, redness, edema, hypertrophy, and gingival BOP; and 3 = severe inflammation with marked redness and hypertrophy and a tendency for spontaneous bleeding and ulceration.[19] The GI score was calculated by dividing the summed scores of individual sites by the number of examined sites. The GSI score was calculated by dividing the number of sites scored as 2 or 3 in the GI by the total number of sites examined in each patient.[20] Before starting this clinical trial, the authors decided to analyze GSI in patients who had gingival bleeding at baseline to evaluate the effect of the APM and control dentifrices at 1 month (APM group: n = 115, control group: n = 120) and 3 months (APM group: n = 110, control group: n = 116). Gingival redness in the buccal area around each tooth was scored as follows: 0 = absent (normal gingival color); 1 = present at the interdental papilla; 2 = present at the marginal gingiva; and 3 = present at the attached gingiva.[21] The gingival redness score was calculated by dividing the summed scores of individual sites by the number of examined sites. Individual oral hygiene status was determined from plaque control records (PCRs).[22]

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**Degree of salivary oxidation.** Unstimulated whole saliva samples (3 mL) collected from the participants by passive flow were immediately stored at –80°C until analysis. The TAO of saliva was then measured using FRAP.[23] Briefly, saliva samples were separated by centrifugation at 10,000 × g for 10 minutes, and supernatants were diluted two-fold with saline. Aqueous solutions with known $Fe^{II}$ concentrations ranging from 0.125 to 2 mmol/L were used for calibration. The antioxidant reagent comprised 25 mL of 0.3 mol/L acetate buffer (pH 3.6); 2.5 mL of 0.01 mol/L 2,4,6-tripyridyl-*s*-triazine in 0.04 mol/L HCl; and 2.5 mL of 0.02 mol/L $FeCl_3 \cdot 6H_2O$. Antioxidant reagent (150 μL) was added to 96-well plates containing 20 μL $Fe^{II}$ standard, diluted saliva, or undiluted saliva, and then absorbance was measured at 595 nm using a plate reader.†† Salivary antioxidant levels were calculated as:

$$Y = (S - B)/A,$$

where $Y$ is the antioxidant level (FRAP, mmol/L), $S$ is sample absorbance, $B$ is the intercept of the linear regression model, and $A$ is the slope of the linear regression model.

### *Safety Evaluation*

The safety evaluation population included individuals who used the test dentifrice and underwent at least one oral cavity inspection. Examiners evaluated the safety of each dentifrice, and the incidence of adverse events was calculated from oral cavity and systemic medical findings throughout the trial period, as confirmed by visual assessment and interviews.

### *Statistical Analyses*

The primary outcome (GI at 3 months) was analyzed in ITT and per-protocol populations. Secondary outcomes (GSI, gingival redness, and TAO of saliva) were analyzed in the per-protocol population only. The per-protocol population excluded patients from the ITT population who took medications, underwent professional scaling, or did not use the assigned dentifrice during the trial. The GI, gingival redness scores, and TAO of saliva were assessed using analysis of covariance. Baseline-adjusted GI and redness scores of the groups were compared at 1 and 3 months. The GSI score and the PCR score were analyzed using Mann–Whitney *U* test. The GSI score was analyzed in individuals with gingival bleeding at baseline. The authors evaluated differences between the four dental facilities and between sexes by using the interaction in the analysis of covariance with primary outcome. Baseline scores for each dentifrice were compared using a *t* test. The incidence of adverse events was compared between the groups using Fisher exact test. Among the baseline characteristics of the patients, sex and smoking habits were compared between the groups using Fisher exact test, and age was compared using a *t* test. In this study, values of $P<0.05$ are considered significant. All data were statistically analyzed using software.‡‡

## RESULTS

### *Study Population*

Figure 1 shows the flow of participants through each stage from enrollment to statistical analysis. Among 305 participants who applied for this clinical trial, five were excluded because they withdrew consent or did not meet inclusion criteria. Three hundred participants were randomly assigned to receive either the APM or the control dentifrice. The ITT population, which excluded patients who did not undergo oral cavity inspection, comprised 294 participants at 1 month (145 in APM group, 149 in control group) and 292 at 3 months (144 in APM group, 148 in control group). The per-protocol population, which excluded those in the ITT population who deviated from the protocol, comprised 285 participants at 1 month (142 in APM group, 143 in control group) and 273 at 3 months (135 in APM group, 138 in control group). The baseline characteristics of the participants, including age, sex, and smoking habits, did not significantly differ between the two groups (Table 2). The authors checked the patients' medications and plaque-retentive factors and confirmed that none were taking oral contraceptive medication or calcium antagonist agents or had plaque retentive factors that could affect the evaluation of gingival status. Furthermore, patients who took AA supplements on a daily basis were not registered in this study.

### *Clinical Scores*

The primary outcome in the ITT population, namely the adjusted mean GI score, was lower for the APM group than for the control group at 3 months, but the difference did not reach statistical significance ($P = 0.12$). However, in the per-protocol population, the primary and secondary outcomes, as well as the adjusted mean GI and gingival redness scores, were significantly lower ($P = 0.01$ and $P = 0.04$, respectively) for the APM group than for the control group at 3 months (Table 3). Before this analysis, the authors confirmed a significant difference between periods and the interaction between period and group by using a repeated measures analysis. The reduction in GSI from baseline was significantly greater in the APM group than in the control group at 3 months ($P = 0.02$; Table 3). The individual oral

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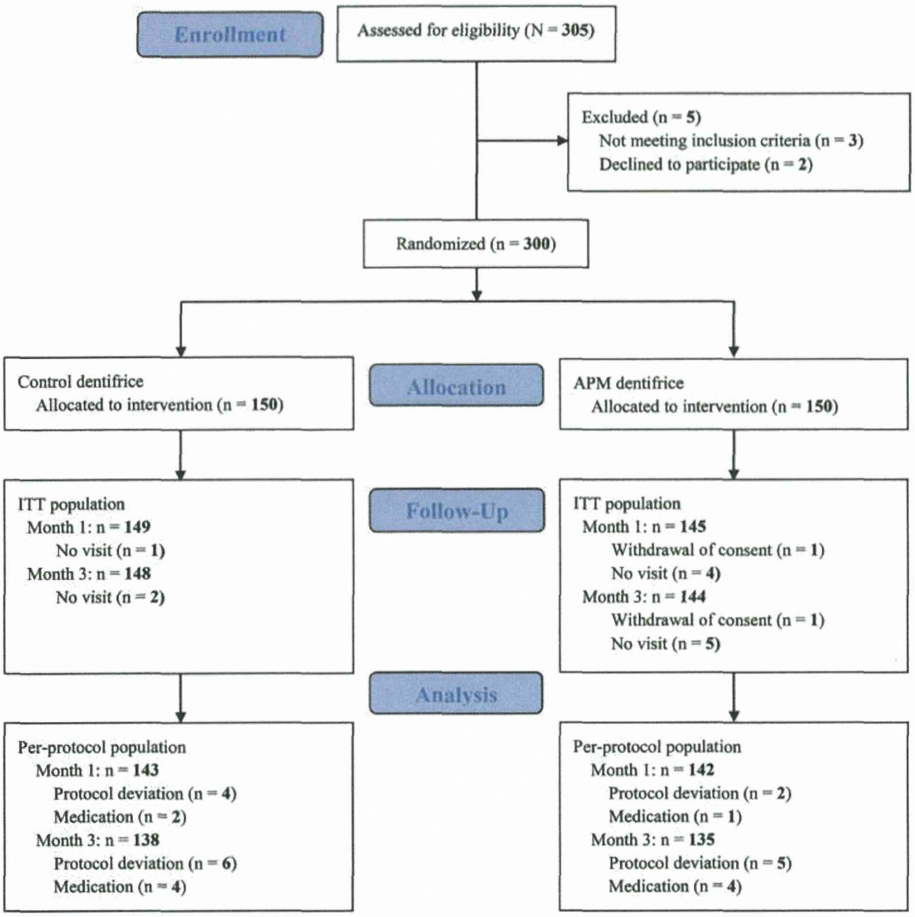


**Figure 1.**
*CONSORT 2010 flow diagram.*

hygiene status as assessed by PCR did not significantly differ between the groups at 3 months ($P>0.15$). The effects of each dentifrice on all clinical scores did not significantly differ among the four dental facilities or by sex.

### *Safety Evaluation*

None of the patients experienced adverse events severe enough to cause withdrawal from the trial. Adverse events that might have been related to the dentifrice, such as oral cavity irritation, were found in one patient in the APM group and four patients in the control group. All adverse events disappeared as the trial continued. The incidence of adverse events did not significantly differ between the groups (APM versus control: 0.7% versus 2.7%, $P>0.15$).

### *Degree of Salivary Oxidation*

The authors assessed the TAO of saliva by measuring FRAP values. The 3-month adjusted mean FRAP value of the APM group was significantly higher than that of the control group ($P = 0.03$) (Table 4).

## DISCUSSION

The onset and progression of periodontal disease result from a combination of factors, including periodontopathic bacteria and the host response. Most dentifrices that aim to prevent gingivitis contain antibacterial agents such as triclosan/copolymer and stannous fluoride to inhibit plaque formation and protect gingival health.[24] Although these dentifrices have been proven effective against gingivitis,[25,26] new strategies that act on the host response and reduce gingival inflammation may be needed in view of the high prevalence of periodontal disease.

The present clinical trial evaluated the effects of an antioxidant dentifrice containing 0.3% APM on gingival inflammation. The aim of this study is to evaluate the effect of an antioxidant dentifrice on the inflammatory status of periodontal tissues, including the effect on the whole marginal gingiva.

The baseline GI, gingival redness, and FRAP scores did not significantly differ among the APM and control groups. This finding suggests that the gingival inflammation, gingival chromatic change, and antioxidant levels of the patients at the start of the trial were similar. The GI scores, redness, and GSI were also not significantly different between groups in the ITT population ($P = 0.12$ and 0.69, respectively). However, the APM dentifrice significantly improved GI, redness, and GSI at 3 months in the per-protocol population. The per-protocol population excluded patients who took medications such as antibacterial or anti-inflammatory drugs, who underwent professional scaling, or who did not use the assigned dentifrice. In the per-protocol population, four patients from each group who took medications were excluded from the study at 3 months (Fig. 1). Five patients in the APM group and six in the control group who deviated from the protocol were also excluded (Fig. 1). Therefore, the authors postulate that improvement in gingival inflammation resulting from these medications might have affected the study results in the ITT population. The aim of this study is to evaluate the effects of the APM dentifrice, which

**Table 2.**

## Patient Characteristics

| Characteristic | APM Dentifrice | Control Dentifrice | P* |
|---|---|---|---|
| Number of patients | 150 | 150 | |
| Sex (% of total) | | | 0.73 |
| Males | 52.0 | 49.3 | |
| Females | 48.0 | 50.7 | |
| Mean age (years) ± SD | 38.1 ± 10.5 | 38.9 ± 9.8 | 0.50 |
| Smoking habits (% of total) | | | 0.88 |
| No | 81.3 | 80.0 | |
| Yes | 18.7 | 20.0 | |
| Mean GI ± SE | 1.22 ± 0.03 | 1.17 ± 0.03 | 0.23 |
| Mean gingival redness ± SE | 1.10 ± 0.04 | 1.12 ± 0.04 | 0.74 |
| Mean GSI ± SE | 0.34 ± 0.02 | 0.30 ± 0.02 | 0.25 |
| Mean total antioxidant activity in saliva ± SE | 0.423 ± 0.02 | 0.413 ± 0.02 | 0.71 |

* Not significantly different between the groups ($P > 0.15$).

**Table 3.**

## Clinical Scores at 1 and 3 Months (per-protocol population)

| | Analysis at 1 Month | | | Analysis at 3 Months | | | |
|---|---|---|---|---|---|---|---|
| Treatment | n | Baseline mean ± SE* | Result† | n | Baseline mean ± SE* | Result† | P‡ |
| GI§ | | | | | | | |
| APM | 142 | 1.21 ± 0.03 | 0.88 ± 0.02 | 135 | 1.22 ± 0.03 | 0.73 ± 0.03 | 0.01 |
| Control | 143 | 1.16 ± 0.03 | 0.93 ± 0.02 | 138 | 1.16 ± 0.03 | 0.84 ± 0.03 | |
| Gingival redness‖ | | | | | | | |
| APM | 142 | 1.10 ± 0.04 | 0.85 ± 0.02 | 135 | 1.09 ± 0.04 | 0.69 ± 0.03 | 0.04 |
| Control | 143 | 1.12 ± 0.04 | 0.88 ± 0.02 | 138 | 1.13 ± 0.04 | 0.78 ± 0.03 | |
| GSI¶ | | | | | | | |
| APM | 115 | 0.40 ± 0.03 | 0.17 ± 0.02 | 110 | 0.41 ± 0.03 | 0.21 ± 0.02 | 0.02 |
| Control | 120 | 0.35 ± 0.03 | 0.15 ± 0.02 | 116 | 0.34 ± 0.03 | 0.15 ± 0.02 | |

* Baseline means and SE for 1- and 3-month per-protocol populations.
† Results for GI and gingival redness are adjusted mean ± SE from analysis of covariance with the baseline score as the covariate; results for GSI are mean of reduction from baseline ± SE.
‡ *P* values indicate the differences between the APM group and the control group at 3 months.
§ *P* value of GI scores at baseline versus 3 months was <0.001.
‖ *P* value of gingival redness scores at baseline versus 3 months was <0.001.
¶ The GSI score was analyzed in individuals with gingival bleeding at baseline. The *P* values of GSI scores at baseline versus 3 months was 0.04.

has antioxidant activity, on gingival inflammation in the absence of antibacterial agents. The authors used PCR scores to examine whether plaque accumulation influenced the clinical effects and found no significant difference in bacterial adhesion rates between the groups. This finding suggests that the improved clinical scores in the APM group were not the result of better brushing to remove adherent bacteria but instead reflected the physiologic effects of APM. The GI, redness, and GSI scores in the per-protocol analyses indicated that APM could be a valuable adjunct to antigingivitis dentifrices.

The TAO of saliva was assessed using FRAP assays to determine how the continuous use of a dentifrice containing antioxidants affects periodontal antioxidant status. The antioxidant profile of saliva significantly differs from that of gingival crevicular fluid (GCF)[27] because the major antioxidants in saliva are uric acid and ascorbate, whereas that in GCF is reduced glutathione.[28,29] Whole saliva is more

**Table 4.**

## Total Antioxidant Activity in Saliva at 1 and 3 Months in the Per-Protocol Population

| | Analysis at 1 Month | | | Analysis at 3 Months | | | |
|---|---|---|---|---|---|---|---|
| Treatment | n | Baseline mean ± SE* | Adjusted mean ± SE† | n | Baseline mean ± SE* | Adjusted mean ± SE† | P‡ |
| APM | 140 | 0.426 ± 0.014 | 0.435 ± 0.014 | 132 | 0.430 ± 0.013 | 0.452 ± 0.015 | 0.03 |
| Control | 142 | 0.413 ± 0.014 | 0.415 ± 0.013 | 137 | 0.413 ± 0.014 | 0.405 ± 0.015 | |

* Baseline means and SE for 1- and 3-month per-protocol populations.
† Adjusted means and SE of 1 and 3 months from analysis of covariance with the baseline score as the covariate.
‡ *P* value indicates the difference between the APM group and the control group at 3 months.

relevant than GCF to analyses of antioxidants because it contains GCF, immune cells, and tissue metabolites.[30,31] Many studies have determined periodontal antioxidant status by analyzing saliva because it is a more practical, albeit surrogate, analytical fluid.[32] Therefore, the authors assessed antioxidant activity using unstimulated saliva. Several assays have been used to assess the TAO of saliva, e.g., the 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) equivalent antioxidant capacity assay,[33] FRAP, and the enhanced chemiluminescent assay.[34] However, each assay measures only a few antioxidant species, and it is therefore very difficult to clarify the TAO of saliva. The FRAP assay directly measures antioxidants with a reduction potential below that of the $Fe^{3+}/Fe^{2+}$ couple[35,36] and is convenient for evaluating the TAO associated with ascorbate, but not with glutathione. Because the major antioxidant species are uric acid and ascorbate in saliva and glutathione in GCF, the authors investigated the effect of the APM dentifrice on the antioxidant status of saliva using the FRAP assay. However, this assay measures only a few species and is thus not representative of TAO in vivo. Sculley et al. reported that the method used for saliva collection and storage can influence its antioxidant capacity at analysis.[31] Indeed, Chapple et al. suggested that it is necessary to centrifuge saliva and store it at –80°C under liquid nitrogen.[34] In this study, unstimulated whole saliva samples were collected by passive flow, and samples were immediately stored at –80°C until analysis. The TAO of the saliva was determined using the FRAP assay. The mean concentration of TAO was significantly higher in the APM group than in the control group (Table 4). Participants fasted for only 1 hour before the clinical examination. Therefore, dietary antioxidant intake might have affected the TAO of the saliva. To the best of the authors' knowledge, this is the first evaluation of the effect of a dentifrice containing an antioxidant on the TAO of unstimulated whole saliva. Thus, further studies are warranted to clarify the effect on oral antioxidant status.

Several recent reports[37,38] have implicated ROS in the progression of periodontal disease, thus indicating the importance of antioxidant activity. Tsutsumi et al. suggested that APM has greater antioxidant activity than AA.[14] Furthermore, APM can enhance intercellular AA in human gingival fibroblasts and thus accelerates Type I collagen synthesis and inhibits IL-8 production by suppressing intercellular ROS. In a preliminary study, the present authors examined the penetration of APM through the epithelial barrier using a hamster cheek pouch model. The cheek pouch was treated with a three-fold diluted extract of the APM dentifrice for 10 minutes, after which APM present in the epithelial tissue was measured by high performance liquid chromatography. Penetrating APM was detected in the epithelial tissue. These findings indicate that the local application of a dentifrice to deliver APM to the gingival tissues can reduce gingival inflammation and enhance salivary antioxidant status through the multiple functions of APM.

The present study finds that APM, administered in a dentifrice, improves the symptoms of gingivitis and salivary antioxidant levels. In this study, the control dentifrice has the effect of decreasing gingival inflammation; this means that brushing affected gingival inflammation. In this situation, the difference in adjusted GI mean scores between the APM dentifrice and control dentifrice at 3 months was 13.1%. Moreover, in patients with mild-to-moderate inflammation, the GI score of the APM group was significantly lower in a time-dependent manner, compared with the control group. Accordingly, the authors would expect to observe greater effects on gingival inflammation in a long-term clinical study of the role of APM on the gingival defense response and saliva antioxidant status.

## CONCLUSIONS

Within the limitations of this study, the results indicate that regular application of a dentifrice containing an antioxidant significantly reduces gingivitis, redness, and gingival bleeding. The relationship

between signs of gingivitis and salivary antioxidant levels should be investigated in more detail.

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# Emerging Regenerative Approaches for Periodontal Reconstruction: A Consensus Report From the AAP Regeneration Workshop

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**Background:** Historically, periodontal regeneration has focused predominantly on bone substitutes and/or barrier membrane application to provide for defect fill and/or selected cell repopulation of the lesion. More recently, a number of technologies have evolved that can be viewed as emerging therapeutic approaches for periodontal regeneration, and these technologies were considered in the review paper and by the consensus group. The goal of this consensus report on emerging regenerative approaches for periodontal hard and soft tissue reconstruction was to develop a consensus document based on the accompanying review paper and on additional materials submitted before and at the consensus group session.

**Methods:** The review paper was sent to all the consensus group participants in advance of the consensus conference. In addition and also before the conference, individual consensus group members submitted additional material for consideration by the group. At the conference, each consensus group participant introduced themselves and provided disclosure of any potential conflicts of interest. The review paper was briefly presented by two of the authors and discussed by the consensus group. A discussion of each of the following topics then occurred based on the content of the review: a general summary of the topic, implications for patient-reported outcomes, and suggested research priorities for the future. As each topic was discussed based on the review article, supplemental information was then added that the consensus group agreed on. Last, an updated reference list was created.

**Results:** The application of protein and peptide therapy, cell-based therapy, genetic therapy, application of scaffolds, bone anabolics, and lasers were found to be emerging technologies for periodontal regeneration. Other approaches included the following: 1) therapies directed at the resolution of inflammation; 2) therapies that took into account the influence of the microbiome; 3) therapies involving the local regulation of phosphate and pyrophosphate metabolism; and 4) approaches directed at harnessing current therapies used for other purposes. The results indicate that, with most emerging technologies, the specific mechanisms of action are not well understood nor are the specific target cells identified. Patient-related outcomes were typically not addressed in the literature. Numerous recommendations can be made for future research priorities for both basic science and clinical application of emerging therapies. The need to emphasize the importance of regeneration of a functional periodontal organ system was noted. The predictability and efficacy of outcomes, as well as safety concerns and the cost-to-benefit ratio were also identified as key factors for emerging technologies.

**Conclusions:** A number of technologies appear viable as emerging regenerative approaches for periodontal hard and soft tissue regeneration and are expanding the potential of reconstructing the entire periodontal organ system. The cost-to-benefit ratio and safety issues are important considerations for any new emerging therapies.

**Clinical Recommendation:** At this time, there is insufficient evidence on emerging periodontal regenerative technologies to warrant definitive clinical recommendations. *J Periodontol 2015;86(Suppl.):S153-S156.*

**KEY WORDS**

**Alveolar bone grafting; bone matrix; guided tissue regeneration, periodontal; periodontitis; tissue engineering.**

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