Higher Intakes of Fruits and Vegetables, \( \beta \)-Carotene, Vitamin C, \( \alpha \)-Tocopherol, EPA, and DHA Are Positively Associated with Periodontal Healing after Nonsurgical Periodontal Therapy in Nonsmokers but Not in Smokers\(^1\text{-}^3 \)

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**Abstract**

**Background:** Periodontitis is a chronic inflammatory disease and a significant risk factor for tooth loss. Although a link between diet and periodontal health exists, the relation between diet and healing after periodontal therapy has yet to be investigated.

**Objective:** The objective was to determine whether higher intakes of fruits and vegetables or nutrients with antioxidant or anti-inflammatory activity are associated with greater healing, measured as reduced probing depth (PD), after scaling and root planing (SRP), a cost-effective treatment to manage periodontal disease and prevent tooth loss.

**Methods:** Patients (63 nonsmokers, 23 smokers) with chronic generalized periodontitis who were undergoing SRP participated. Healing was evaluated based on PD, assessed at baseline and 8–16 wk after SRP. Intakes of fruits, vegetables, \( \beta \)-carotene, vitamin C, \( \alpha \)-tocopherol, \( \alpha \)-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) were estimated using the Block 2005 food frequency questionnaire and a supplement questionnaire. Serum 25-hydroxyvitamin D concentrations were also measured. PD (% sites >3 mm) was modeled in multiple linear regression and analysis of covariance by tertile of intake and adjusted for age, sex, body mass index (BMI), baseline PD, examiner, gingival bleeding, and study duration.

**Results:** In nonsmokers, PD was associated with fruit and vegetable, \( \beta \)-carotene, vitamin C, \( \alpha \)-tocopherol, EPA, and DHA intakes \((P < 0.05) \). PD was not significantly associated with ALA intake or serum 25-hydroxyvitamin D concentration. Significant associations that included supplements (\( \beta \)-carotene, vitamin C, \( \alpha \)-tocopherol) were attenuated or lost, depending on the statistical model used. There were no significant associations within the group of smokers.

**Conclusions:** Dietary intakes of fruits and vegetables, \( \beta \)-carotene, vitamin C, \( \alpha \)-tocopherol, EPA, and DHA are associated with reduced PD after SRP in nonsmokers, but not smokers, with chronic generalized periodontitis. These findings may lead to the development of dietary strategies to optimize healing after periodontal procedures. This trial was registered at clinicaltrials.gov as NCT02291835.

**Keywords:** anti-inflammatory, antioxidants, bone, healing, oral health, periodontal health, tooth retention

**Introduction**

Periodontal disease is a chronic inflammatory condition that affects over 42% of the adult population in the United States and 65% of the population older than 65 y \((1) \). Similarly, in Canada, periodontal disease affects 20% of the adult population and 30% of the population older than 65 y \((2) \), indicating a high burden of periodontal disease in both countries. Periodontal disease results in the destruction of the tissues that support the teeth, including the gingivae, periodontal ligament, and alveolar bone. Although initiated by bacterial pathogens, periodontal disease mainly occurs due to an overactive immune response in susceptible individuals \((3) \). Smoking is a significant risk factor for periodontal disease and may account for approximately half the cases in the United States \((4) \). Smokers experience further...
burden due to the decreased effectiveness of periodontal therapy (5). Periodontal disease is characterized by increased inflammation and reactive oxygen species (ROS)6 locally in the periodontal tissue but also at the systemic level (6, 7). Periodontal disease, therefore, increases the risk for other chronic diseases with an inflammatory component, including diabetes (8) and cardiovascular disease (9). If left untreated, periodontal disease ultimately results in loss of teeth, which has a number of significant health consequences. Tooth loss is associated with poorer nutritional status (10, 11), poorer quality of life (12) and increased risk of death from chronic diseases such as cancer (13) and cardiovascular disease (14).

Nonsurgical periodontal therapy consisting of deep scaling and root planing (SRP) is a first-line and cost-effective treatment to manage periodontal infections (15). SRP is a mechanical process that removes bacterial pathogens that have invaded into the periodontal tissues, thus removing the etiological agent of the disease. As tissue destruction subsides, healing can take place and some reattachment of teeth can occur (16). The retention of natural teeth depends on the success of periodontal therapy; therefore, there is a need to develop strategies to increase its effectiveness. Because periodontal healing depends on the resolution of inflammation and ROS, dietary strategies may prove effective in optimizing periodontal healing after SRP.

The association between nutrition and periodontal health has been investigated in many studies. Greater intakes of foods and nutrients with antioxidant and anti-inflammatory activity have consistently been linked to a reduced risk of developing periodontal disease. Higher intakes of vitamin C (17–22) β-carotene (18, 20), vitamin E (19, 20), fruits and vegetables (20), and omega-3 FAs (23, 24) have each been associated with greater periodontal health (reviewed in 25). Some (26–28) but not all (29) studies have shown that higher serum 25-hydroxyvitamin D [25(OH)D] concentrations are associated with better periodontal health. Studies investigating diet and periodontal healing have since emerged. For example, I study has demonstrated an association between vitamin D sufficiency and improved healing after periodontal surgery (30). Preliminary clinical trials using dietary supplements including fish oil (31) and encapsulated fruit and vegetable concentrate (32) have also shown to be effective in optimizing periodontal outcomes after SRP. However, there still remain relatively few studies that have investigated the association between diet and periodontal healing. The objective of this study was to determine whether greater intakes of fruits and vegetables or specific nutrients with antioxidant/anti-inflammatory activity, including β-carotene, vitamin C, α-tocopherol, vitamin D, α-linolenic acid (ALA), EPA, and DHA, are associated with improved periodontal healing after SRP. We hypothesized that higher intakes of fruits and vegetables, β-carotene, vitamin C, α-tocopherol, ALA, EPA, and DHA and higher serum 25(OH)D concentrations would be associated with greater reductions in PD after SRP but that this relation would be attenuated in smokers.

Methods

Study population and design. The study took place at a reconstructive periodontics and implant surgery clinic (Fonthill, Ontario, Canada) between January 2013 and July 2014. Before study enrollment, patients attended a consultation visit in which a comprehensive baseline dental examination including medical/dental history and complete periodontal charting was completed. All patients whose treatment plan included deep SRP were invited to participate in the study. To be included in the study, patients had to have a PD of 4 mm or greater in at least 30% of probed sites, which is the criteria for chronic generalized periodontitis set by the American Academy of Periodontology (33). Both nonsmokers and smokers were recruited for the study with the intention of performing stratified analyses because smokers have compromised healing after SRP (5). Full-mouth SRP was then performed by one of four hygienists using hand and ultrasonic instruments as necessary, and oral hygiene instructions were provided to the patients. At the appointment in which SRP was performed, participants had their serum 25(OH)D concentrations measured and were provided with an FFQ and dietary supplement use questionnaire to be completed at home and returned at their follow-up appointment. Patients then returned ~8–16 wk later for their follow-up appointment, which included a complete periodontal examination. Only patients who followed up within 16 wk of SRP were then included in the study, to avoid missing relapse of periodontitis as compromised healing. The human research ethics board at Brock University in St. Catharines, Ontario, Canada approved the study protocol, and all participants provided written informed consent.

Periodontal examination. Baseline and follow-up periodontal examinations included measurements of PD and bleeding on probing (BOP). PD was selected as our main outcome because it is routinely used to evaluate the response to periodontal therapy. BOP was used as a measure of acute inflammation and oral hygiene care. PD was measured using a periodontal probe as the distance from the gingival margin to the bottom of the periodontal pocket at six sites per tooth (buccal, mesial buccal, distal buccal, lingual, mesial lingual, and distal lingual) on all teeth present. Overall PD was then calculated as the % of sites with PD > 3 mm. Sites on teeth designated for extraction were not included in the calculation. BOP was assessed by visual inspection after probing and is expressed as percentages of sites that bled when probed. The baseline examinations were all performed by the same periodontist and occurred 1–19 wk before SRP, with the exception of the consultation of one patient, which occurred 57 wk before SRP. The hygienist, who then performed the SRP, also performed the follow-up periodontal examination. Before the study, the hygienists were calibrated to apply 25 N of pressure when probing by repeated probing simulations against an electronic scale.

Dietary assessment. To estimate usual intakes of nutrients and foods, participants completed the 2005 Block FFQ, which has been previously validated against multiple diet records (34). The FFQ queries 110 food items with an additional series of adjustment questions to provide greater accuracy in assessing carbohydrate and fat intake. Frequency (never, a few times a year, once per month, 2–3 times per month, once per week, 2 times per week, 3–4 times per week, 5–6 times per week, every day) and portion size were asked about for each food, and portion-size pictures were provided to enhance accuracy of quantification. Because the FFQ estimates intakes during the past year, an assumption was that usual intakes during the past year reflect intakes during the study period. Nutrient and food intake estimates were calculated using a database of nutrient values derived from the Canadian Nutrient File. One serving of fruits and vegetables was counted as 125 ml fresh, frozen, or canned fruit or vegetable (including potatoes) or 100% juice; or 250 ml leafy raw vegetables or salad; or 1 piece of fruit. Completeness was checked by ensuring that larger sections of the FFQ (>2 consecutive items) were not missed. Missing values were interpreted as no intake. The reported energy intake (REI) was comparable to that of other studies using the BLOCK FFQ in a Canadian population (35). All of the participants’ total energy intakes fell within the following energy limits: >600 kcal/d and <4200 kcal/d for males and >450 kcal/d and <3600 kcal/d for females; with a total of 10 participants who had an REI of <1000 kcal/d. To account for the possible effects of underreporting on observed associations, participants were classified as under-reporters or accurate reporters by comparing their REI to their estimated resting metabolic

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6 Abbreviations used: ALA, α-linolenic acid; BOP, bleeding on probing; PD, probing depth; REI, reported energy intake; ROS, reactive oxygen species; SRP, scaling and root planing; 25(OH)D, 25-hydroxyvitamin D.
rate (RMR), which was calculated using age, height, and weight. Fifty participants (59%) had a REI less than their RMR and thirty-five participants (41%) had a REI greater than their RMR. Regression analyses were repeated, examining under-reporters and accurate reporters separately. Regression coefficients were generally smaller in under-reporters, indicating that error from under-reporting was not leading to false significant associations because under-reporters slightly weakened the associations observed in this study. Therefore, participants with REI less than RMR were included in the analyses to preserve sample size.

All nutrient and food intakes were then energy adjusted and standardized to a 2000 kcal diet using the residual method (36). Supplemental nutrient intakes were assessed using a dietary supplement questionnaire. Participants were provided a list of supplements and asked to report which ones were taken, the brand, the dose, frequency, and the length of time that they have been taking it. The nutritional composition of all supplements (including multivitamins) was recorded. To differentiate between natural and synthetic forms of \( \alpha \)-tocopherol from dietary supplements, intake in mg was calculated from the amount in international units listed on the manufacturers label as follows: 1 mg \( \alpha \)-tocopherol = 1.49 IU dl-\( \alpha \)-tocopherol (natural, RRR form) or 2.22 IU dl-\( \alpha \)-tocopherol (synthetic, all \( \alpha \)-tocopherol form).

For dietary \( \alpha \)-tocopherol intake estimates, it was assumed that only the natural form of \( \alpha \)-tocopherol was present in foods because there are currently no foods to which micronutrient addition regulations in Canada allow for the addition of synthetic vitamin E (except for meal replacements). Total intakes for nutrients were calculated as the sum of energy-adjusted dietary intakes plus supplemental intakes (supplemental intakes were not energy adjusted).

Assessment of vitamin D status. During the appointment in which SRP was performed, participants met with a study nurse who collected a venous blood sample. Blood was allowed to clot for 30 minutes at room temperature before centrifugation, and serum samples were stored at -20°C until analysis. Frozen serum samples were delivered to LifeLabs Medical Laboratory Services (St. Catharines, ON), where serum 25(OH)D concentrations were measured using the LIAISON chemiluminescent immunoassay (DiaSorin Inc.). The LIAISON 25-Hydroxyvitamin D method is traceable to the National Institute of Standards and Technology (NIST) Standard Reference Material (SRM) 972a and NIST and University of Ghent reference methods, through the National Institutes of Health Office of Dietary Supplements Vitamin D Standardization Program (VDSP). Internal quality control materials included the LIAISON 25 OH vitamin D control set and LIAISON Calibration Verifiers. The laboratory participates in the Vitamin D External Quality Assessment Scheme (DEQAS), as well as programs with the College of American Pathologists (CAP) and the Institute for Quality Management in Healthcare (IQM4).

Assessment of covariates. Self-reported age, sex, health conditions, medications used, and smoking status (never, former, or current) were recorded from the patient’s medical history form. When participants met with the study nurse, they had their height and body weight measured (Health-O-Meter Professional; Sunbeam Products, Inc.) to calculate their body mass index (BMI; in kg/m²).

Statistical analysis. Participant characteristics, nutrient intakes, and supplement use were compared between nonsmokers and smokers using an independent sample \( t \) test for continuous variables and chi-square testing for categorical variables. When the expected cell count was <5 for categorical variables, the Fisher exact test was used in place of the chi-square test. The relation between diet and periodontal healing was investigated using multiple linear regression. Periodontal healing was evaluated using the percentage of sites with a PD \( \geq 3 \) mm as a continuous dependent variable with adjustment for baseline PD (percentage of sites \( \geq 3 \) mm) in the model. Dietary intakes and total intakes (where applicable), of fruits + vegetables, \( \beta \)-carotene, vitamin C, \( \alpha \)-tocopherol, ALA, EPA + DHA, as well as serum 25(OH)D concentrations were each separately used as continuous predictors. The sum of EPA and DHA was used as a predictor because intakes of EPA and DHA were very highly correlated (\( r = 0.99 \)). Dietary intakes of EPA, DHA, and total intakes of vitamin C, \( \alpha \)-tocopherol, EPA, and DHA were log transformed to improve the fit of the model. Then, to determine the clinical effect size associated with a defined level of intake, the same predictors were classified into tertiles of equal size. Differences between tertiles were evaluated using ANCOVA. All models were adjusted for baseline PD, hygienist, age, sex, BMI, and BOP (a marker of oral hygiene care), along with the number of days between treatment and follow-up. To our knowledge, there were no apparent sources of bias. To comply with assumptions of regression, all models were evaluated for normality of residuals and multicollinearity. Data were analyzed with SPSS version 20 (SPSS Inc.); statistical significance was defined as \( P < 0.05 \).

Results

The recruitment and final sample size flow chart is shown in Figure 1. Of the 129 patients recruited for the study, 17 were lost to follow-up, 11 did not follow-up within four months, and 3 had missing data because they did not complete the FFQ, leaving 98 patients who completed the study. Of those, 12 did not meet the criteria for chronic generalized periodontitis at baseline (at least 30% of sites with PD \( \geq 4 \) mm) and were excluded from the analysis, leaving a final sample size of 63 nonsmokers and 23 smokers. There were no statistically significant differences in patient characteristics, baseline clinical measures, or the serum 25(OH)D concentration between those included and those excluded from analyses (Supplemental Table 1). Participant characteristics, nutrient intakes, and supplement use are shown in Table 1. Compared with nonsmokers, smokers were younger, more likely to be of normal weight (BMI \( = 18.5-24.0 \)), less likely to be overweight (BMI \( = 25.0-29.9 \)), and had a greater percentage of sites with PD \( \geq 3 \) mm at follow-up (\( P < 0.05 \)). Smokers also had lower intakes of dietary EPA and DHA and had lower serum 25(OH)D concentrations, compared with nonsmokers (\( P < 0.05 \)).

To determine the effects of selected covariates, follow-up PD was modeled in linear regression using age, sex, BMI, baseline PD, BOP, hygienist, and the number of days between treatment and follow-up as predictors (Table 2). Of these, baseline PD (\( P = 0.001 \)) and BOP (\( P = 0.023 \)) were positively associated with follow-up PD, and patients who saw hygienist 2 had fewer sites of \( >3 \) mm at follow-up (\( P = 0.012 \)).

The regression and ANCOVA analyses for nonsmokers, as well as dietary intake ranges for each tertile, are summarized in Table 3. Using multiple linear regression, fruit + vegetable intake was not significantly associated with the percentage of

![FIGURE 1](https://academic.oup.com/jn/article-abstract/145/11/2512/4585757)
Dietary intakes
Dietary supplement use,
Follow-up clinical measures
Baseline clinical measures
BMI, n (%)       M |
| (n = 63) | (n = 23) | P value |
| M       | 33 (52) | 10 (44) | 0.47 |
| F       | 30 (48) | 13 (57) | 0.47 |

BMI, n (%)
Normal, 18.5–24.9 | 9 (14) | 9 (39) | 0.014 |
Overweight, 25.0–29.9 | 30 (48) | 5 (22) | 0.041 |
Obese, ≥30.0 | 24 (38) | 8 (35) | 0.89 |

Smoking status, n (%) 6
Never smoker | 30 (48) | — | — |
Former smoker | 33 (52) | — | — |

Diabetes, n (%) 6
6 (10) | 2 (9) | 1.00 |

Baseline clinical measures
Teeth, n 6
25 ± 4 (12–32) | 25 ± 4 (12–28) | 0.81 |
PD, % sites >3 mm 6
63 ± 20 (34–99) | 68 ± 21 (33–99) | 0.25 |
BOP, % of sites 6
50 ± 29 (0–100) | 48 ± 33 (7–100) | 0.77 |
Plaque score, % of teeth 6
73 ± 29 (10–100) | 70 ± 33 (0–100) | 0.72 |
Follow-up time, d 6
76 ± 16 (50–116) | 80 ± 16 (56–117) | 0.26 |

Follow-up clinical measures
PD, % sites >3 mm 6
9 ± 6 (0–25) | 13 ± 10 (2–44) | 0.028 |
BOP, % of sites 6
4 ± 1 (0–23) | 5 ± 6 (0–34) | 0.36 |
Plaque score, % teeth 6
32 ± 23 (0–100) | 40 ± 31 (0–100) | 0.28 |

Dietary intakes
FV 2 servings/d 6
6.4 ± 2.9 (1.5–16.8) | 5.2 ± 2.9 (1.8–12.6) | 0.08 |
β-Carotene, mg/d 6
6.8 ± 4.4 (0.1–26.9) | 5.3 ± 6.3 (0.3–31.5) | 0.27 |
Vitamin C, mg/d 6
131 ± 45 (24–241) | 117 ± 68 (40–311) | 0.26 |
α-Tocopherol, mg/d 6
6.7 ± 1.9 (2.5–12.4) | 7.5 ± 3.4 (2.8–20.6) | 0.20 |
ALA, g/d 6
1.4 ± 0.4 (0.7–2.9) | 1.2 ± 0.3 (0.6–1.8) | 0.07 |
EPA + DHA, mg/d 6
275 ± 279 (0–155) | 137 ± 99 (25–338) | 0.024 |

Dietary supplement use, n (%) 6
None 24 (38) | 12 (52) | 0.24 |
Any type 39 (62) | 11 (48) | 0.24 |
Multivitamin 20 (32) | 5 (22) | 0.37 |
Vitamin C 10 (16) | 3 (13) | 1.00 |
Vitamin D 19 (30) | 3 (13) | 0.11 |
Vitamin E 2 (3) | 1 (4) | 1.00 |
Fish oil 10 (16) | 1 (4) | 0.28 |
ω-3/ω-6/ω-9 3
2 (3) | 0 (0) | 1.00 |

Serum 25(OH)D, nmol/L 6
64 ± 24 (13–129) | 51 ± 18 (29–90) | 0.020 |

1 All values are means ± SDs (range) for continuous variables and counts (%) for categorical variables. ALA, α-linolenic acid; BOP, bleeding on probing; FV, fruits and vegetables; PD, probing depth; 25(OH)D, 25-hydroxyvitamin D.
2 One serving is 125 mL fresh, frozen, or canned fruit or vegetable or 100% juice; or 250 mL leafy raw vegetables or salad; or one piece of fruit.
3 Supplement includes a combination of ω-3, ω-6, and ω-9 FAs.

Diet and periodontal healing
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TABLE 2 Eﬀects of covariates on regression of follow-up PD in nonsmoking adults undergoing nonsurgical periodontal therapy 6

<table>
<thead>
<tr>
<th>Predictor</th>
<th>β (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>0.14 (−0.09, 0.38)</td>
<td>0.23</td>
</tr>
<tr>
<td>Sex</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>M</td>
<td>0.01 (−0.23, 0.25)</td>
<td>0.91</td>
</tr>
<tr>
<td>F</td>
<td>−0.04 (−0.26, 0.21)</td>
<td>0.77</td>
</tr>
<tr>
<td>BMI, kg/m2</td>
<td>−0.04 (−0.26, 0.21)</td>
<td>0.77</td>
</tr>
<tr>
<td>Follow-up BOP, % sites &gt;3 mm</td>
<td>0.47 (0.21, 0.72)</td>
<td>0.001</td>
</tr>
<tr>
<td>Time to follow-up, d</td>
<td>−0.01 (−0.25, 0.22)</td>
<td>0.91</td>
</tr>
<tr>
<td>Hygienist</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>Hygienist 1</td>
<td>−0.38 (−0.68, −0.09)</td>
<td>0.012</td>
</tr>
<tr>
<td>Hygienist 2</td>
<td>−0.22 (−0.52, 0.07)</td>
<td>0.13</td>
</tr>
<tr>
<td>Hygienist 3</td>
<td>−0.19 (−0.48, 0.10)</td>
<td>0.19</td>
</tr>
</tbody>
</table>

1 BOP, bleeding on probing; PD, probing depth.

Discussion
In the present study, our objective was to determine whether greater intakes of fruits and vegetables or specific nutrients with antioxidant/anti-inflammatory activity, including β-carotene, vitamin C, α-tocopherol, vitamin D, ALA, EPA, and DHA, are associated with improved periodontal healing after SRP. Our data showed that patients that were nonsmokers with greater intakes of fruits and vegetables, β-carotene, vitamin C, α-tocopherol, EPA, and DHA had a lower percentage of sites with PD >3 mm after SRP, indicating that they experienced a greater degree of periodontal healing compared with patients with lower intakes. Furthermore, because our findings were adjusted for BMI and BOP and did not include smokers, we showed that these associations were independent of major risk factors for periodontal disease, which include maintenance of a healthy body weight (37), poor oral hygiene (38), and smoking (4). It is also likely that these associations are generalizable to other populations of patients with chronic periodontitis undergoing SRP because these associations were found within a heterogeneous
population with minimal inclusion criteria (i.e., nonsmokers with chronic generalized periodontitis). We suspect that associations between diet and periodontal healing were not detected in smokers because smoking is an overwhelming risk factor for periodontal disease (4) and compromised oral wound healing (5). In addition, studies in humans show that smokers have lower circulating levels of \( \beta \)-carotene (39), vitamin C (40), and \( \alpha \)-tocopherol (41), making it possible that associations were not observed due to depletion of antioxidants in smokers as a result of oxidative turnover. Whether smokers require higher antioxidant intake for periodontal healing is a question that warrants further investigation in a larger sample of smokers with periodontitis.

In this study, reductions in PD were seen among patients with greater fruit and vegetable intake. Significant improvements were seen even in the middle tertile of intake, with at least 5.1 – 6.7 servings/d (125 mL/d) of fruits and vegetables per day. These amounts are slightly lower than current dietary guidelines in Canada, which recommend 7–8 servings/d for women and 8–10 servings/d for men (42). However, even modest increases in fruit and vegetable intakes (especially in those with very low intakes) may be beneficial for periodontal healing. It is important to recognize, however, that FFQs tend to overestimate food intakes (43) and therefore, intake levels derived from this study and comparisons to the RDA should be interpreted with caution. This finding is consistent with the findings of a clinical trial by Chapple et al (32), who provided periodontal patients undergoing SRP a supplement containing fruit and vegetable juice powder concentrates or placebo and observed greater reductions in mean PD in the treatment group two months after SRP. The association between fruit and vegetable intake and periodontal healing may

### TABLE 3

Regression of follow-up PD by dietary intakes, total intakes (dietary + supplemental), or serum 25(OH)D concentrations in nonsmoking adults with periodontitis undergoing SRP

<table>
<thead>
<tr>
<th><strong>Linear Regression</strong></th>
<th><strong>ANCOVA</strong></th>
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</thead>
<tbody>
<tr>
<td><strong>b (95% CI) P value</strong></td>
<td><strong>Tertile (n = 21/tertile) % Sites &gt;3mm (95% CI) P value</strong></td>
</tr>
<tr>
<td><strong>Fruits and vegetables</strong></td>
<td>-0.20 (−0.45, 0.05) 0.11</td>
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<tr>
<td></td>
<td>5.1–6.7 serving/d 8 (6, 10)*</td>
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<td></td>
<td>6.8–18.8 serving/d 7 (4, 9)*</td>
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<tr>
<td><strong>Total ( \beta )-carotene</strong></td>
<td>-0.28 (−0.52, −0.03) 0.031</td>
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<tr>
<td></td>
<td>6.99–15.00 mg/d 9 (6, 11)</td>
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<tr>
<td><strong>Dietary vitamin C</strong></td>
<td>-0.36 (−0.58, −0.14) 0.002</td>
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<td></td>
<td>108–149 mg/d 9 (7, 12)</td>
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<tr>
<td><strong>Total vitamin C</strong></td>
<td>-0.29 (−0.53, −0.04) 0.022</td>
</tr>
<tr>
<td></td>
<td>141–204 mg/d 8 (5, 10)*</td>
</tr>
<tr>
<td><strong>Dietary ( \alpha )-tocopherol</strong></td>
<td>-0.24 (−0.50, 0.01) 0.06</td>
</tr>
<tr>
<td></td>
<td>5.9–7.1 mg/d 8 (5, 10)*</td>
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<td></td>
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<tr>
<td><strong>Total ( \alpha )-tocopherol</strong></td>
<td>-0.06 (−0.39, 0.18) 0.61</td>
</tr>
<tr>
<td></td>
<td>6.6–12.4 mg/d 8 (5, 11)</td>
</tr>
<tr>
<td><strong>Serum 25(OH)D</strong></td>
<td>-0.11 (−0.34, 0.13) 0.37</td>
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<tr>
<td></td>
<td>54–68 nmol/L 9 (6, 11)</td>
</tr>
<tr>
<td><strong>Dietary ALA</strong></td>
<td>-0.10 (−0.34, 0.13) 0.42</td>
</tr>
<tr>
<td></td>
<td>1.2–1.5 g/d 8 (5, 10)</td>
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<td></td>
<td></td>
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<tr>
<td><strong>Dietary EPA+DHA</strong></td>
<td>-0.14 (−0.38, 0.10) 0.23</td>
</tr>
<tr>
<td></td>
<td>137–225 mg/d 9 (6, 11)</td>
</tr>
<tr>
<td><strong>Total EPA+DHA</strong></td>
<td>-0.28 (−0.52, −0.04) 0.025</td>
</tr>
<tr>
<td></td>
<td>149–249 mg/d 8 (6, 10)*</td>
</tr>
</tbody>
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1 All models were adjusted for age, sex, BMI, baseline PD, hygienist, follow-up BOP, and time to follow-up. *, **Different from lowest tertile: * P < 0.05; ** P < 0.01. ALA, \( \alpha \)-linoleic acid; BOP, bleeding on probing; PD, probing depth; SRP, scaling and root planing; 25(OH)D, 25-hydroxyvitamin D.

2 Regression of PD (% sites >3 mm) using intakes or serum 25(OH)D concentration as continuous predictors. Standardized regression coefficients (\( b \)) shown.

3 Comparison of PD (% sites >3 mm) between tertiles of intake or tertile of serum 25(OH)D using ANCOVA. Estimated marginal means are shown.

4 One serving is 125 mL fresh, frozen, or canned fruit or vegetable or 100% juice; or 250 mL leafy raw vegetables or salad; or one piece of fruit.
partly be due to greater intakes of dietary antioxidants. In this study, higher dietary intakes of β-carotene (≥7.07 mg/d), vitamin C (≥150 mg/d), and α-tocopherol (≥5.9 mg/d) were associated with greater PD reductions. Levels of reactive oxygen metabolites have been correlated with clinical parameters of periodontal disease (44), generating a potential role for dietary antioxidants in managing periodontitis. However, associations between PD and antioxidant intakes were consistently attenuated when contributions from dietary supplements were included in our analyses. This finding seems to suggest that single antioxidants alone do not provide the same benefit compared with whole-food sources. A partial explanation is that antioxidants such as vitamin C and vitamin E function in concert to eliminate reactive oxygen metabolites and lipid peroxidation (44). In addition, diets rich in fruits and vegetables not only provide antioxidants but are a source of over 5000 plant-based phytochemicals (45), and health benefits are likely due to synergistic effects rather than any individual nutrient (46). Similarly, we suspect that optimization of periodontal healing with diet may be better approached using whole foods, including antioxidant-rich foods, rather than purified compounds in supplements.

Vitamin C intake was the nutrient most strongly correlated with periodontal healing. Low vitamin C intake or low circulating ascorbic acid concentrations have consistently been associated with increased risk for periodontal disease (17–22). This has been attributed to the antioxidant activity of vitamin C and the fact that it is essential for collagen biosynthesis (47) and, therefore, wound healing. The RDA for vitamin C is 75 mg/d for females and 90 mg/d for males. Despite this, patients with intake of >150 mg/d had improved response to sanative therapy, suggesting that intake of vitamin C beyond the RDA may be beneficial in periodontal treatment. In a clinical trial, patients undergoing SRP were given 2000 mg/d of supplemental vitamin C for 4 wk with no difference in periodontal outcomes (48). This study, however, had only a small sample of n = 15 and did not account for baseline vitamin C status. Thus, based on the strong associations observed in our study, the use of vitamin C as an adjunct to SRP warrants further investigation.

Because a previous study (30) showed greater reductions in mean PD after open-flap SRP when serum 25(OH)D concentrations were ≥50 nmol/L, we expected to see an association between 25(OH)D concentrations and mean PD after SRP. However, that was not observed. Discrepancies between their study results and ours may be due to the fact that the previous study had patients with more severe periodontal disease and that those patients had a more invasive procedure performed. It is also possible that the associations with vitamin D were driven by smoking status because there was a disproportionate number of smokers in the low-25(OH)D group. In contrast, we analyzed smoking status separately and found no evidence that short-term periodontal healing is affected by vitamin D status in smokers or nonsmokers. Vitamin D intake, however, has been linked to maintenance of periodontal health after initial periodontal treatment (49) and therefore, studies of longer duration may be needed to determine the benefits that vitamin D may have for patients with periodontal disease.

ω-3 FAs may also play important roles in periodontal healing. In this study, greater intakes of EPA and DHA but not ALA were associated with a lower mean PD after SRP. This finding is consistent with epidemiological evidence that shows associations between the long-chain ω-3 FA DHA, and periodontal disease but not the shorter-chain ω-3 FA ALA (23). In this study, FA supplements that include EPA and DHA appeared to benefit patients because dietary intakes alone were not significantly associated with PD. In a recent clinical trial, patients undergoing SRP taking a supplement with 300 mg/d of EPA + DHA had a lower mean PD, compared with those taking placebo (31). In vitro, resolvin D1, a downstream metabolite of DHA, has been shown to optimize healing of periodontal biopsies through downregulation of inflammatory mediators (50). Therefore, the consumption of fish or the use of ω-3 FAs in conjunction with SRP should be further investigated.

There are limitations in this study. We had a relatively small sample size (n = 63 nonsmokers and n = 23 smokers). However, having repeat measurements at the individual level and adjustment for baseline periodontal status has provided additional power relative to a cross-sectional design. Also, because the ultimate goal is to develop clinically useful dietary strategies, we were not interested in small effect sizes requiring large populations to detect. Other limitations include the use of a self-reported dietary-assessment method and having only a one-time assessment of dietary intakes because it is possible that patients changed their dietary behaviors after periodontal treatment. FFQs have also shown to overestimate dietary intakes (43) and thus, a 3-d food record may have been more suitable for our sample size. In investigating the relation between vitamin E and periodontal healing, we focused only on the α-tocopherol form of vitamin E, which is a potential limitation of this work because other forms of vitamin E, such as γ-tocopherol, have been shown to exert anti-inflammatory effects, especially when combined with α-tocopherol (51). Our study also included multiple comparisons, which increases the possibility of type I error. Some of the clinical limitations are the use of multiple examiners and not having baseline PD measurements taken at the time of treatment. However, it should be noted that these limitations would have likely resulted in attenuation of the significant associations detected in this study. Lastly, due to the observational nature of the study, the possibility remains that covariates that were unaccounted for may have contributed to the observed associations.

In conclusion, these findings suggest that higher dietary intakes of fruits and vegetables, β-carotene, vitamin C, α-tocopherol, EPA, and DHA are associated with greater reductions in PD after SRP in nonsmokers with periodontal disease. Dietary intervention studies should be developed to determine whether a causal relation exists between these dietary components and optimal periodontal healing.

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