Periodontal Therapy Reduces Plasma Levels of Interleukin-6, C-Reactive Protein, and Fibrinogen in Patients With Severe Periodontitis and Refractory Arterial Hypertension

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Background: Recent epidemiologic studies suggest that inflammation is the link between periodontal diseases and cardiovascular complications. This study aimed to evaluate the effects of non-surgical periodontal treatment on plasma levels of inflammatory markers (interleukin [IL]-6, C-reactive protein [CRP], and fibrinogen) in patients with severe periodontitis and refractory arterial hypertension.

Methods: Twenty-two patients were examined and randomly divided into two groups. The test group was composed of 11 patients (mean age, 48.9 ± 3.9 years) who received periodontal treatment, whereas the control group had 11 patients (mean age, 49.7 ± 6.0 years) whose treatment was delayed for 3 months. Demographic and clinical periodontal data were collected, and blood tests were performed to measure the levels of IL-6, CRP, and fibrinogen at baseline and 3 months later.

Results: The clinical results showed that the mean percentages of sites with bleeding on probing, probing depth (PD) 4 to 5 mm, PD ≥6 mm, clinical attachment loss (CAL) 4 to 5 mm, and CAL ≥6 mm were significantly reduced in the test group 3 months after periodontal treatment. There were no significant differences between the data at baseline and 3 months in the control group. Periodontal treatment significantly reduced the blood levels of fibrinogen, CRP, and IL-6 in the test group.

Conclusion: Non-surgical periodontal therapy was effective in improving periodontal clinical data and in reducing the plasma levels of IL-6, CRP, and fibrinogen in hypertensive patients with severe periodontitis. J Periodontol 2009;80:786-791.

KEY WORDS
Albumins; C-reactive protein; cardiovascular disease; fibrinogen; periodontitis.

Cardiovascular disease (CVD) is the leading cause of death in Western industrialized countries. Classic modifiable risk factors, such as smoking, diabetes, hyperlipidemia, and arterial hypertension, are present in 50% of the patients with CVD, and the treatment of these conditions may reduce morbidity and mortality. Hypertension is the product of a dynamic interaction between diverse genetic, physiologic, environmental, and psychosocial factors. Patients whose blood pressure remains persistently high despite the appropriate use of at least three classes of antihypertensive drugs, including a diuretic, are diagnosed as having refractory hypertension. Inflammation may be involved in the initiation and development of hypertension by the action of proinflammatory mediators. Inflammation was associated with endothelial dysfunction and increased arterial stiffness. Arterial stiffness is an independent predictor of mortality in patients with hypertension, and the reduction of arterial stiffness should be a goal in the treatment of hypertension.

Some systemic inflammatory markers may indicate the severity of inflammation, and their levels have been associated with CVDs, including hypertension.
Fibrinogen and C-reactive protein (CRP), products of the acute-phase reaction, were demonstrated to be independent cardiovascular risk factors for CVDs. Elevated CRP has been associated with an increased risk for CVDs, atherosclerosis, and the presence and development of hypertension. Moreover, elevated CRP levels are associated with artery endothelial dysfunction and arterial stiffness. Periodontal treatment improved endothelial dysfunction in recent studies. Kim et al. suggested that CRP could be a useful marker of arterial stiffness in treated hypertensive patients and a possible target for arterial inflammation in hypertension. Fibrinogen was recognized as an independent coronary heart disease risk factor and an inflammatory marker. Together with CRP, fibrinogen was considered predictive for future cardiovascular risk. Interleukin (IL)-6 is a major initiator of the acute-phase response by hepatocytes, a primary determinant of CRP production, and is associated with incident acute coronary events. IL-6 is the major initiator of the acute-phase response by hepatocytes and a primary determinant of hepatic CRP production. IL-6 is associated with incident acute coronary events and hypertension risk. Periodontal therapy reduced the plasma levels of IL-6 and improved endothelial function.

Periodontitis is a chronic low-grade inflammatory disease of the tooth-supporting tissues that may lead to tooth loss and may increase blood levels of inflammatory markers, including IL-6, CRP, and fibrinogen. However, there are controversial data about the effects of periodontal treatment on inflammatory markers.

Therefore, the aim of the present study was to evaluate the effects of non-surgical periodontal therapy on serum levels of plasma inflammatory markers (IL-6, CRP, and fibrinogen) in patients with severe periodontitis and refractory arterial hypertension.

**MATERIALS AND METHODS**

Twenty-two patients with severe primary non-responsive (refractory) arterial hypertension, enrolled for treatment at the Hypertension Department, National Institute of Cardiology, were selected for the study from June to December 2007. Refractory hypertension is diagnosed when blood pressure levels remain >140/90 mm Hg while a patient is engaged in a treatment program and uses three or more classes of antihypertensive drugs, including a diuretic. The patients were under the supervision of a cardiologist and were medicated with beta-blockers, angiotensin-converting enzyme inhibitors, and a diuretic. The patients were treated in the cardiology department for 5 to 8 years. Periodontal examination included plaque index (PI); bleeding on probing (BOP), recorded as present or absent; probing depth (PD); and clinical attachment level (CAL). PD and CAL were evaluated on six sites/tooth (mesio-buccal, buccal, disto-buccal, mesio-lingual, lingual, and disto-lingual), whereas PI and BOP were evaluated on four sites/tooth (mesial, buccal, distal, and lingual). The examination was performed by the same calibrated operator (kappa = 0.91) using a controlled-pressure periodontal probe (20 to 25 g/F).

The inclusion criteria were ≥12 teeth, at least four sites with PD ≥4 mm and five sites with CAL ≥6 mm, and compliance with treatment for hypertension. The exclusion criteria included pregnancy, previous periodontal therapy, systemic conditions that contraindicated periodontal therapy or that might affect the progression or treatment of periodontitis (e.g., diabetes mellitus or history of infective endocarditis), and the use of medications (e.g., antibiotics or anti-inflammatory drugs). Patients were consecutively recruited and randomly assigned, by means of a draw, to the test or control group.

The test group was composed of 11 patients (five men and six women; age range, 43 to 56 years; mean age, 48.9 ± 3.9 years), whereas the control group had 11 patients (six men and five women; age range, 39 to 62 years; mean age, 49.7 ± 6.0 years). There were two smokers (>10 cigarettes per day) in each group. On the morning of the periodontal appointment, blood samples were collected and immediately processed, masked by the laboratory staff from the hospital at the National Institute of Cardiology, Rio de Janeiro, RJ, Brazil, to quantify the plasma levels of IL-6, CRP, and fibrinogen. Patients from the test group received non-surgical periodontal therapy, whereas patients in the control group had their treatment delayed for 3 months. Periodontal therapy included oral hygiene instructions and supra- and subgingival scaling, which were performed using a sonic instrument and Gracey curets and followed the American Academy of Periodontology recommendations.

There was no limit to the appointments; however, in general, patients were seen four to six times over 2 weeks. The control group was recalled 3 months after the baseline visit; the test group was recalled 3 months after the last appointment for periodontal therapy, for reassessment of the periodontal parameters, and to repeat the blood test.

The study respected the principles of the Declaration of Helsinki, was revised and approved by the Hospital’s Ethical Board, and all patients gave their written consent to participate in the study.

**Statistical Methods**

A post hoc calculation showed that with a sample size of nine patients in each group, there was 80% power to
detect, at a 0.05 level, a 50% reduction in the mean percentage of sites with PD and CAL ≥6 mm. The t test was used for normally distributed data, whereas the Mann-Whitney test was used to compare non-normally distributed data at baseline. The χ² test was used to compare categoric data at baseline. To verify differences between clinical and laboratory values at baseline and 3 months after periodontal therapy, the paired t test and the Wilcoxon rank test were used to compare normally and non-normally distributed data, respectively. All statistical analyses were carried out with a statistical program,** with a significance level of 5% (P < 0.05).

RESULTS
There were no significant differences between the test and control groups in terms of age, number of teeth, gender, race, mean body mass index (BMI), systolic and diastolic blood pressures, percentage of smokers, percentage of persons from a low socioeconomic class, or percentage of patients with generalized chronic periodontitis (Table 1).

At baseline, there were no significant differences in the clinical periodontal data between the test and control groups. The mean percentage of sites with BOP, PD 4 to 5 mm, PD ≥6 mm, CAL 4 to 5 mm, and CAL ≥6 mm was significantly reduced in the test group 3 months after periodontal treatment. There was no difference in the mean percentage of sites with dental plaque (Table 2).

There was no difference between the groups at baseline for any of the laboratory parameters studied. In the test group, there was a significant reduction in the levels of IL-6 (P = 0.03), fibrinogen (P = 0.03), and CRP (P = 0.005) 3 months after periodontal therapy. In the control group, the levels of IL-6 (P = 0.01) and CRP (P = 0.01) had increased significantly after 3 months without periodontal therapy. No significant differences were observed with regard to fibrinogen (P = 0.75) levels in the control group (Table 3).

DISCUSSION
Our results showed that periodontal therapy significantly reduced the plasma levels of CRP, IL-6, and fibrinogen in patients with arterial hypertension. To our knowledge, this is the first study to show reduction of the inflammatory markers CRP, IL-6, and fibrinogen after periodontal treatment, at the same time observation (i.e., at 3 months). Our data showing a reduction in CRP is in line with other studies.29,31,35-38 Conversely, Ide et al.32 and Yamazaki et al.39 did not show significant reductions in CRP levels. Significant reductions in the plasma levels of IL-6 were observed by D’Aiuoto et al.31, Elter et al.20 and Higashi et al.38 but not by Ide et al.32 and Yamazaki et al.39 Fibrinogen levels were not decreased in the studies by Mattila et al.,35 Ide et al.,32 and Montebugnoli et al.37. The interval between blood collection varied between 1 and 6 months, which may have affected the results for the different markers. CRP was not reduced 6 weeks after periodontal therapy in the study by Ide et al.32 although Mattila et al.35 reported a reduction in CRP levels in this same interval. Iwamoto et al.36 collected blood 1 month after periodontal therapy and observed significant reductions in CRP, whereas Elter et al.20 reported a trend in the reduction of serum levels of CRP during this interval. D’Aiuoto et al.31 and Higashi et al.38 observed reductions in CRP only after 6 months. IL-6 levels were significantly reduced 1,20 2, and 6 months31,38 after periodontal treatment. However, Ide et al.32 and Yamazaki et al.39 reported no significant reductions in IL-6 levels 6 weeks and 3 months after periodontal treatment, respectively. Studies32,35,37 that evaluated fibrinogen levels collected blood samples 6 weeks and 3 months after periodontal treatment and did not find significant changes.

In the present study, we collected blood samples 3 months after periodontal therapy because this is considered an ideal time to evaluate periodontal healing after non-surgical periodontal treatment.40 Significant reductions in CRP, IL-6, and fibrinogen plasma

** SPSS 11.0, SPSS, Chicago, IL.

Table 1.
Distribution of Variables in the Test and Control Groups

<table>
<thead>
<tr>
<th>Variable</th>
<th>Test Group (n = 11)</th>
<th>Control Group (n = 11)</th>
<th>P Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years; mean [SD])</td>
<td>48.9 (3.9)</td>
<td>49.7 (6.0)</td>
<td>0.71</td>
</tr>
<tr>
<td>Male (%)</td>
<td>55.5</td>
<td>63.6</td>
<td>1.0</td>
</tr>
<tr>
<td>Black (%)</td>
<td>28</td>
<td>28</td>
<td>1.0</td>
</tr>
<tr>
<td>Smokers (%)</td>
<td>18.2</td>
<td>18.2</td>
<td>1.0</td>
</tr>
<tr>
<td>Low socioeconomic class (%)</td>
<td>100</td>
<td>100</td>
<td>1.0</td>
</tr>
<tr>
<td>Generalized chronic periodontitis (%)</td>
<td>72.7</td>
<td>72.7</td>
<td>1.0</td>
</tr>
<tr>
<td>Systolic BP (mm Hg; mean [SD])</td>
<td>176.4 (25.4)</td>
<td>188.2 (38.2)</td>
<td>0.41</td>
</tr>
<tr>
<td>Diastolic BP (mm Hg; mean [SD])</td>
<td>113.6 (16.3)</td>
<td>118.2 (24.0)</td>
<td>0.61</td>
</tr>
<tr>
<td>BMI (kg/m²; mean [SD])</td>
<td>31.0 (5.7)</td>
<td>29.6 (5.6)</td>
<td>0.56</td>
</tr>
<tr>
<td>Teeth (n; mean [SD])</td>
<td>20.2 (6.9)</td>
<td>18.6 (5.6)</td>
<td>0.55</td>
</tr>
</tbody>
</table>

BP = blood pressure.
* There were no significant differences between test and control groups at baseline.
levels were observed. The baseline values of inflammatory markers, susceptibility of the patients, selection criteria of the studied population, and differences in the severity of periodontal disease may explain some of the discrepancies in the results compared to previous studies.

**Table 2.**
Clinical Variables (% of sites; mean ± SD) in the Test and Control Groups at Baseline and 3 Months

<table>
<thead>
<tr>
<th>Variable</th>
<th>Test Group (n = 11)</th>
<th>Control Group (n = 11)</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline 3 Months</td>
<td>Baseline 3 Months</td>
<td>P1</td>
<td>P2</td>
</tr>
<tr>
<td>Visible plaque</td>
<td>59.4 (26.6) 49.4 (17.6)</td>
<td>65.8 (24.4) 60.8 (29.1)</td>
<td>0.260</td>
<td>0.59</td>
</tr>
<tr>
<td>BOP</td>
<td>45.7 (18.2) 19.4 (11.4)</td>
<td>42.7 (15.1) 40.3 (12.2)</td>
<td>0.002</td>
<td>0.44</td>
</tr>
<tr>
<td>PD 4 to 5 mm</td>
<td>19.5 (9.1) 4.9 (3.2)</td>
<td>19.2 (8.2) 17.8 (9.5)</td>
<td>0.000</td>
<td>0.18</td>
</tr>
<tr>
<td>PD ≥6 mm</td>
<td>8.1 (9.1) 1.2 (0.9)</td>
<td>5.6 (4.2) 5.6 (4.1)</td>
<td>0.025</td>
<td>0.96</td>
</tr>
<tr>
<td>CAL 4 to 5 mm</td>
<td>23.7 (8.9) 18.4 (10.1)</td>
<td>28.4 (11.9) 25 (9.5)</td>
<td>0.014</td>
<td>0.14</td>
</tr>
<tr>
<td>CAL ≥6 mm</td>
<td>24.5 (22.4) 11.2 (14.9)</td>
<td>18.9 (15.8) 19 (18.1)</td>
<td>0.002</td>
<td>0.86</td>
</tr>
</tbody>
</table>

P1 = difference between baseline and 3 months after periodontal therapy for test group; P2 = difference between baseline and 3 months for control group; P3 = difference between test and control groups at baseline.

**Table 3.**
Plasma Levels of CRP, Fibrinogen, and IL-6 at Baseline and 3 Months

<table>
<thead>
<tr>
<th>Patient</th>
<th>CRP (mg/dl)</th>
<th>Fibrinogen (mg/dl)</th>
<th>IL-6 (pg/ml)</th>
<th>CRP (mg/dl)</th>
<th>Fibrinogen (mg/dl)</th>
<th>IL-6 (pg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline 3 Months</td>
<td>Baseline 3 Months</td>
<td>Baseline 3 Months</td>
<td>Patient</td>
<td>Baseline 3 Months</td>
<td>Baseline 3 Months</td>
</tr>
<tr>
<td>1</td>
<td>1.49 1.35</td>
<td>376 360</td>
<td>234 87.6</td>
<td>0.71 2.99</td>
<td>331 490</td>
<td>5.8 6.8</td>
</tr>
<tr>
<td>2</td>
<td>0.38 0.15</td>
<td>494 376</td>
<td>2.1 1.0</td>
<td>0.09 0.1</td>
<td>512 490</td>
<td>9.0 9.2</td>
</tr>
<tr>
<td>3</td>
<td>0.70 0.30</td>
<td>318 312</td>
<td>1.0 1.0</td>
<td>0.03 0.04</td>
<td>253 254</td>
<td>1.0 1.0</td>
</tr>
<tr>
<td>4</td>
<td>0.49 0.06</td>
<td>252 258</td>
<td>1.0 1.0</td>
<td>0.03 0.07</td>
<td>283 276</td>
<td>1.0 1.0</td>
</tr>
<tr>
<td>5</td>
<td>2.95 1.05</td>
<td>385 372</td>
<td>5.1 2.0</td>
<td>1.07 1.78</td>
<td>372 360</td>
<td>1.0 7.6</td>
</tr>
<tr>
<td>6</td>
<td>1.07 1.19</td>
<td>497 380</td>
<td>4.1 1.0</td>
<td>0.82 1.49</td>
<td>380 376</td>
<td>54.4 234</td>
</tr>
<tr>
<td>7</td>
<td>0.51 0.61</td>
<td>423 396</td>
<td>5.9 7.4</td>
<td>0.61 0.49</td>
<td>276 253</td>
<td>1.0 1.0</td>
</tr>
<tr>
<td>8</td>
<td>0.36 0.25</td>
<td>376 231</td>
<td>3.3 1.0</td>
<td>0.98 2.95</td>
<td>372 385</td>
<td>1.0 5.1</td>
</tr>
<tr>
<td>9</td>
<td>1.36 1.39</td>
<td>364 332</td>
<td>2.5 1.0</td>
<td>0.68 1.07</td>
<td>372 497</td>
<td>1.0 4.1</td>
</tr>
<tr>
<td>10</td>
<td>2.81 0.39</td>
<td>505 490</td>
<td>2.4 1.0</td>
<td>0.15 0.23</td>
<td>372 396</td>
<td>1.0 1.0</td>
</tr>
<tr>
<td>11</td>
<td>2.99 2.93</td>
<td>490 322</td>
<td>6.8 6.1</td>
<td>0.68 1.36</td>
<td>376 394</td>
<td>1.0 2.5</td>
</tr>
<tr>
<td>Mean (SD)</td>
<td>1.4 (0.8)*</td>
<td>407 (82) (70)t</td>
<td>24.4 (26.8)</td>
<td>0.5 (0.4) (1.1)t</td>
<td>354 (70) (90)</td>
<td>7.0 (15.9) (69.4)t</td>
</tr>
</tbody>
</table>

* Significantly lower than baseline value (P = 0.005).
† Significantly lower than baseline value (P = 0.03).
‡ Significantly higher than baseline value (P = 0.01).

All clinical parameters improved after non-surgical periodontal therapy except for the percentage of sites with visible dental plaque, probably because oral hygiene instructions were only provided at the first visit. However, the mean percentage of sites with BOP, PD, and CAL improved after 3 months of periodontal
treatment, indicating that the treatment performed was effective.

Periodontitis is a low-grade chronic inflammatory disease that affects 35% to 60% of the adult population; severe forms may affect 10% to 15% of the population. Its prevention and treatment may have an impact on the control of chronic diseases related to inflammation, such as hypertension. The causes of refractory hypertension include biologic factors (e.g., obesity), inaccurate blood pressure measurement, non-compliance with prescribed medications, and inadequate treatment. These causes may not be important in the present study because the patients were under regular medical control and had been engaged in a maintenance program for ≥5 years. Despite taking medications, they had high blood pressure levels. The long-term benefits of reducing inflammatory markers and acute-phase proteins in hypertensive patients are unknown. The permanent control of periodontal inflammation may allow for better control of pressoric levels, a reduction in left ventricle mass, and a slowdown of the atherosclerotic process, leading to a reduction in the patient’s cardiovascular risk.

Limitations of this study include a relatively small sample size, which may affect the reproducibility of the results, and the short observation period (3 months). Prospective studies with a larger sample size and longer observation periods are required to evaluate the benefits of reducing these inflammatory markers as a way of decreasing the global cardiovascular risk for patients with arterial hypertension.

CONCLUSION

Non-surgical periodontal therapy was effective in improving periodontal clinical data and reducing the plasma levels of IL-6, CRP, and fibrinogen in hypertensive patients with severe periodontitis.

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REFERENCES

17. Amar S, Gokce N, Morgan S, Loukideli M, Van Dyke TE, Vita JA. Periodontal disease is associated with brachial artery endothelial dysfunction and systemic

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