Biochemical Evaluation of the Effect of Green Tea on Nitric Oxide Radical in Patients with Chronic Periodontitis

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ABSTRACT
Statement of the Problem: Periodontal disease is a periodontal tissue inflammation caused by gram-negative and other bacteria. The release of various cytokins and mediators, including nitric oxide, in the inflammation area usually has detrimental effects on the periodontium, potentiating the inflammatory process. In order to prevent this side effect, investigators have used anti-oxidants such as green tea or nitric oxide synthase inhibitors as a remedy.

Purpose: In this study the effect of green tea on salivary nitric oxide radical was investigated in patients with chronic periodontitis.

Materials and Methods: Two groups of patients (n=10), suffering from chronic periodontitis as control and case groups, were evaluated. The case group used green tea for a period of four weeks. The control group received no intervention. At the end of this period the salivary total nitrite concentration as an indicator of nitric oxide was measured by ELISA technique based on Griess reaction. The results were compared statistically by paired-t-test at P<0.05.

Results: Green tea reduced the salivary concentrations of nitric oxide significantly (P=0.000) in the case group in comparison with the control group which receieved no treatment.

Conclusion: The results indicated that green tea may be considered a useful herb for treatment of chronic periodontitis.

Keywords: Green tea, Nitric oxide, Periodontitis.

INTRODUCTION
Chronic periodontal inflammation is known as periodontitis which is mainly triggered and accentuated by bacterial causes. This immune process and any other inflammatory cascade are affected by humoral and cellular immune elements, among which nitric oxide (NO) can be mentioned. This biochemical substance which is synthesized from L. arginine by inducible nitric oxide synthase (iNOS) plays a protective role in infectious inflammatory diseases. The production rate of this enzyme (iNOS) and hence the release of nitric oxide by immune-competent cells, such as macrophages, is accerlated during infection. A link has been reported between this excess production of nitric oxide and periodontal disease and tissue damage seen in this pathology. In addition, the usefulness of salivary nitric oxide as a biochemical marker of periodontal disease has been proposed. Based on the reports proposing an important role for nitric oxide in etiopathogenesis of periodontal diseases some investigators employed iNOS inhibitors and anti-oxidants in order to treat periodontitis. Green tea is one of the anti-oxidants used. As there are conflicting reports in relation to salivary concentrations of NO in periodontitis this study was undertaken in order to evaluate use of green tea as a remedy in periodontitis.

MATERIALS AND METHODS
A group of twenty patients with severe chronic periodontitis, confirmed by clinical parameters obtained through observations, participated in this study. The protocol of
the investigation was approved by the Ethics Committee of the university. Inclusion criteria
1-Clinical attachment loss of ≥5 mm, measured using Williams periodontal probe
2-Bleeding on probing
Exclusion criteria
1-Subjects who required antibiotic or anti-inflammatory drug therapy
2-History of any systemic diseases
3-Subjects who were pregnant and pre-eclamptic
4-Subjects with a history of smoking and tobacco use
5-Subjects with vitamin supplements
6-Subjects who regularly used mouthwashes
The patients were randomly divided into two groups of ten (n=10). One group was considered as the control (no use of green tea) and the other used green tea (1.75 g of tea leaves in 200 mL of water) for a period of four weeks and was considered as the case group. At the beginning of the experiment and before any intervention a sample of 10 mL of unstimulated salivary samples were collected, centrifuged and the supernatant was frozen at -20°C and stored as base-line samples. After four weeks of treatment of the case group with green tea, again the same salivary samples were collected from the control and case groups.

The NO was measured in the above salivary samples employing Elisa Kit. Based on Griess reaction, the results were analyzed and compared using paired-t-test. Statistical significance was defined at P<0.05.

RESULTS
Total salivary nitrite was measured as nitric oxide index by Griess reaction. There were no significant differences in salivary nitric oxide concentrations between the two groups (A&C) at the beginning of the experiment (P=0.0753).
Significant differences were observed in the salivary nitric oxide concentrations between the control group at the beginning of the experiment (C) and the case and control groups after intervention (B) (P=0.001).
NO salivary concentrations were significantly different in the case group (A) before intervention and the control group (D) at the end of the experiment (P=0.000).
A significant difference was observed between NO salivary concentrations in the control group at the beginning (C) and at the end (D) of the experiment (P=0.000).
Significant differences (P=0.000) were observed in NO salivary concentrations in the case group before (A) and after (B) the intervention (Table 1).

Table 1: Comparison of NO salivary concentrations in the case and control groups at the beginning and at the end of the experiment

<table>
<thead>
<tr>
<th>Nitrite</th>
<th>N</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>Std. Error Mean</th>
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<tbody>
<tr>
<td>Nitrite</td>
<td>A</td>
<td>9</td>
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<td>1.14602</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>9</td>
<td>2.4522</td>
<td>1.14808</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>8</td>
<td>5.1575</td>
<td>1.53026</td>
</tr>
<tr>
<td></td>
<td>D</td>
<td>8</td>
<td>9.8338</td>
<td>1.80885</td>
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</tbody>
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REFERENCES
8- Paquette DW, Williams RC. Modulation of host inflammatory mediators as a treatment strategy for periodontal diseases. Periodontol


