Sialochemistry: Intracellular enzymes & fluoride in periodontitis

Ghalaut Pankaj¹, Kaur Ramanjit²*, Singh Ragini³, Veena S.Ghalaut², Bansal Piyush², Manjubala²

¹Dept. of Periodontics, Govt. Dental College Rohtak, (INDIA)
²Dept. of Biochemistry, PGIMS, Rohtak, (INDIA)
³Dept. of Pathology, PGIMS, Rohtak, (INDIA)

E-mail: Ramanjit.kaur@gmail.com

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ABSTRACT

Sialochemistry provides important information in making a diagnosis and explaining the pathogenesis in a variety of oral and systemic conditions. Study of changes in the enzymatic activity in saliva due to pathological and metabolic changes in the gingiva and periodontium in response of an organism to the periodontal infection may help in accurate assessment of the pathology and provide a non-invasive and convenient diagnostic tool. While fluoride has been considered a protective element it may itself be involved in pathogenesis or its levels may serve as a marker of periodontal pathology.

Intracellular enzymes and fluoride levels were estimated in 30 healthy adult (volunteers) of both sexes of age group 18 – 40 years and 30 persons, of both sexes, aged 18 – 40 years, with periodontal disease. A significant increase in activity of LDH, AST, ALT and ALP was observed in saliva from the patients with periodontal disease in relation to the control group. Fluoride levels were also significantly raised in saliva of the patients with periodontal disease in comparison to the control group.

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INTRODUCTION

Periodontal diseases, including gingivitis and periodontitis, are inflammatory diseases of periodontium. Periodontal infections frequently involve bacteria that discharge hydrogen sulfide, ammonia, amines, toxins, and inflammation-causing enzymes that can cause loss of tissue and teeth. Periodontitis is characterized by inflamed, red gums and deepening pockets (> 3mm in gingivitis &> 5mm in periodontitis) between the tooth root and the gum tissue, as well as loss of bone in the jaw. Advanced periodontal disease can be diagnosed by noticeable loosening of the teeth, gum recession with the tooth root exposed, new spaces forming between the teeth, food being trapped between teeth and where gums have receded and constant bad taste in the mouth[1]. Sialochemistry provides important information in making a diagnosis and explaining the pathogenesis in a variety of clinical conditions, which include a whole spectra ranging from diseases of the oral cavity, salivary glands, systemic diseases and clinical situations in which the chemistry and salivary flow helpful in diag-
nosis or monitoring patient progress[2]. Saliva is admixture of parotid, submandibular, sublingual and minor buccal gland secretions. Several important enzymes are released from stromal, epithelial, inflammatory or bacterial cells which are associated with cell injury or cell death[3]. Changes in the enzymatic activity can be due to metabolic changes in the gingiva and periodontium in response of an organism to the periodontal infection. Prompt diagnosis and a clear picture of pathogenesis of the periodontal disease can be achieved by the analysis of these enzymes in salivary secretions[4,5].

The increased release of intracellular enzymes from the damaged cells of periodontal tissues which are particularly relevant are aspartate and alanine transaminase (ALT & AST), lactate dehydrogenase (LDH), gamma-glutamyltransferase (GGT), creatinine kinase (CK), alkaline phosphatase (ALP), and acid phosphatase (ACP)[6]. While fluoride is considered an protective against dental caries its exact status in periodontal diseases is not clearly understood. In this paper we have examined the activity of LDH, AST, ALT and fluoride in saliva from patients with periodontal disease (experimental group) and in saliva of the healthy tested persons (control group).

**MATERIALS AND METHODS**

The study was carried out in the Department of Biochemistry and Government Dental college, University of Health Sciences, Rohtak. 30 healthy adult volunteers of both sexes of age group 18 - 40 years and 30 persons, of both sexes, aged 18 - 40 years, with periodontal disease were included. Patients with a probing depth >5 mm, bleeding on probing and alveolar bone loss >40% were included.

3.0 mL of un-stimulated mixed saliva was collected in a sterile test tube before breakfast, 3 minutes after a single mouth rinse with 15.0 mL of distilled water to wash out exfoliated cells. Samples were then centrifuged at 10000 rpm for 10 min. Samples were processed on the same day of collection.

Salivary enzyme activities were measured on a Thermoking (Konelab) automated analyzer using analysis kits from Randox and Seimens. Kinetic methods was used for the determination of AST, ALT, ALP and LDH activity. Fluoride was measured using Orion Ion Selective Electrode from thermo- scientific. Samples and standards were diluted in a ratio of 1:4 with TISAB - II (Total Ionic Strength Adjustment Buffer) buffer. Standard curve was prepared and had a mean slope of 52.3 mV / 10X change in fluoride concentration in ppm. The instrument was set to report values in ppm[7]. Statistical analysis was done using microsoft excel and Student’s t-test was applied to compare the study and control group.

**RESULTS**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>Cases</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT (IU/L)</td>
<td>8.65±3.45</td>
<td>76.44±13.58</td>
<td>&lt; 0.001**</td>
</tr>
<tr>
<td>AST (IU/L)</td>
<td>19.53±7.73</td>
<td>189.3±39.7</td>
<td>&lt; 0.001**</td>
</tr>
<tr>
<td>ALP (IU/L)</td>
<td>10.48±5.36</td>
<td>51.68±10.33</td>
<td>&lt; 0.001**</td>
</tr>
<tr>
<td>LDH (IU/L)</td>
<td>89.33±16.45</td>
<td>983.5±158.5</td>
<td>&lt; 0.001**</td>
</tr>
<tr>
<td>Fluoride (ppm)</td>
<td>0.298±0.067</td>
<td>0.467±0.088</td>
<td>&lt; 0.05*</td>
</tr>
</tbody>
</table>

** Highly significant; * Significant

A significant increase in activity of LDH, AST, ALT and ALP was observed in saliva from the patients with periodontal disease in relation to the control group. Fluoride levels were also significantly raised in saliva of the patients with periodontal disease in comparison to the control group.

**DISCUSSION**

Periodontal disease diagnosis is primarily made on the basis of gingival index (GI), bleeding on probing (BOP), probing depth (PD) and alveolar bone loss which is a radiographic parameter, which provides evidence in detecting past disease and verifying periodontal health. Evaluation of many systemic disorders is based on diagnostic laboratory tests in serum. Similarly, many markers in saliva reflect the pathologic changes in the cells of periodontal tissues, particularly intracellular enzymes (AST, ALT, CK, LDH, GGT, ALP, ACP). The activity of these enzymes is ascertained in saliva of normal healthy individuals within normal limits[4]. The destructive process in the alveolar bone in advanced stages of the development of periodontal disease due to damage, edema or destruction of cell membrane of the periodontal tissue causes increased release of these intracellular enzymes into the gingival cervical fluid and saliva. These enzymes can thus serve as markers of functional condition of the periodontal tissue. Similar
results have been observed in the studies on activity of these enzymes in saliva in relation to periodontal diseases. The increased activity of ALT, AST and LDH indicates the pathological changes are primarily located in soft tissues; however increased ALP indicates advanced periodontal disease affecting alveolar bone.\(^\text{8,9}\)

Saliva which is often regarded as the bloodstream of tooth has been discussed lately as important biochemical material, in which these biochemical markers of the functional condition of periodontal tissues reflect pathological changes in cells of periodontal tissues. Simplicity and non-invasiveness of the salivary diagnostic tests leads to greater acceptance by patients.\(^\text{10,11}\)

Plaque bacteria lowers the pH in presence of sucrose which causes the release of the ‘storage’ fluoride which is tightly bound to bacterial cells, epithelial cells or the inorganic constituents, resulting in increase in the salivary fluid levels.

**CONCLUSIONS**

The results of the study emphasize the use of salivary enzymes and salivary fluoride as objective and simple markers of the pathological state of the periodontal tissues which opens new horizons in diagnostic efficacy.

**REFERENCES**