Saliva as a Diagnostic Aid in Periodontitis -A Biochemical Study

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Abstracts: Aim: To study the saliva as a diagnostic aid in gingivitis and periodontitis patients. Method: The study includes 60 subjects of both sexes aged between 15-55 years. 60 subjects were divided into 3 groups of 20 individuals each. Further, each group was divided equally into 10 males and 10 females. The three groups were: (1) Healthy patients who have no gingival or periodontal disease, (2) Gingivitis patients in whom the findings are confined only to gingiva and (3) Third group included periodontitis patients who have periodontal pockets ≥ 4 mm as well as clinical attachment loss. Unstimulated whole saliva samples were been collected and it was transported to the laboratory for biochemical analysis of alkaline phosphatase enzyme, aspartate aminotransferase enzyme and calcium ions, within 24 hours. Result: Results showed, the level of alkaline phosphatase enzyme, aspartate aminotransferase enzyme and calcium ions was found significantly higher in the periodontitis group as compared to the gingivitis and healthy groups. Conclusion: Saliva can be used as an aid in the diagnosis of periodontal disease by identifying the biochemical markers. More longitudinal studies examining the relationship of these identified biochemical markers to the natural history of periodontal disease is the obvious next step in the line of investigation. [Jaladh P NJIRM 2017; 8(2) :106-109]

Key Words: Saliva, diagnostic aid, alkaline phosphatase, aspartate aminotransferase.

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Introduction: Despite our understanding of the etiology and pathogenesis of periodontal disease, the diagnosis of the disease is still based on traditional methods including clinical signs, symptoms and radiographs. The diagnosis of active phases of periodontal diseases and the identification of patients at risk for active disease represents a challenge for both clinical investigators and clinicians. Occasionally, monitoring of microbial infection and analysis of host response in the gingival crevicular fluid are utilized in an attempt to identify individuals at risk for future periodontal breakdown. However till date no clinical or laboratory test is routinely employed in the monitoring of patients with periodontal disease. It has long been realized that a rapid and simple diagnostic test that can provide a reliable evaluation of periodontal disease and identify patients at risk for active disease, would be of value for both clinicians and patients.[1]

Saliva is readily available, can be easily collected and contains locally and systemically derived markers of periodontal disease and hence may offer the basis for a patient specific diagnostic test for periodontitis[2, 3]. A combination of two or more markers may provide a more accurate assessment of the periodontal patient. Salivary diagnostic tests because of their numerous advantages appear to hold promise for the future[4, 5, 6]

Hence in this present biochemical study an attempt is made to assess the saliva as a diagnostic aid in periodontitis.

Methods: The study includes 60 subjects of both sexes aged between 15-55 years have been selected from the Out Patient Department of Dentistry, Government Medical College, Bhavnagar, Gujarat. 60 subjects divided into 3 groups of 20 individuals each. Further, each group was been divided equally into 10 males and 10 females. The groups were as following:

Group I: Comprised of healthy patients who have no gingival or periodontal disease.

Group II: Comprised of gingivitis patients in whom the findings are confined only to gingiva and who do not have a periodontal pockets.

Group III: Comprised of periodontitis patients who have periodontal pockets ≥ 4 mm as well as clinical attachment loss.

A brief case history was taken and following clinical parameters were recorded:

- Gingival index (Loe and Silness 1963).
- Probing depth recorded by using graduated periodontal probe.
- Clinical attachment level.

Collection of Sample:

- Unstimulated whole saliva samples were been collected following a brief rinsing of the mouth with water.

The saliva samples were been collected from the lower vestibular sulcus by a plastic syringe, 2.5 ml sample was been collected and it was been transported to the laboratory for biochemical analysis within 24 hours.

Result: Results of Clinical Parameters:

(A) Gingival Index: The gingival index in group I showed a mean of 0.5270 with a standard deviation of 0.01658. In the group II gingival index had a mean of 1.2575 with a standard deviation of 0.03177, while in group III the mean gingival index was 1.5005 with a standard deviation of 0.0182.

(B) Probing Depth: The mean probing depth in group I was 1.4885 mm with standard deviation of 0.02412; in group II it was 1.994 mm with standard deviation of 0.02741 while in group III it was 4.4945 with standard deviation of 0.02781.

(C) Clinical Attachment Level: Clinical attachment level in group I showed a mean of 1.4915 mm with a standard deviation of 0.02519. In the group II clinical attachment level had a mean of 2.0035 mm with a standard deviation of 0.02925 while in group III the mean clinical attachment level was 5.4940 mm with a standard deviation of 0.03899.

Results of Biochemical Analysis:

(A) Alkaline Phosphatase Enzyme: [Table I] [Graph I]: The salivary alkaline phosphatase level in group I had a mean value of 1.205 U/L with a standard deviation of 0.248. In the group II the mean value was 1.440 U/L with a standard deviation of 0.239. In the group III the alkaline phosphatase enzyme had a mean value of 3.195 U/L with a standard deviation of 0.268.

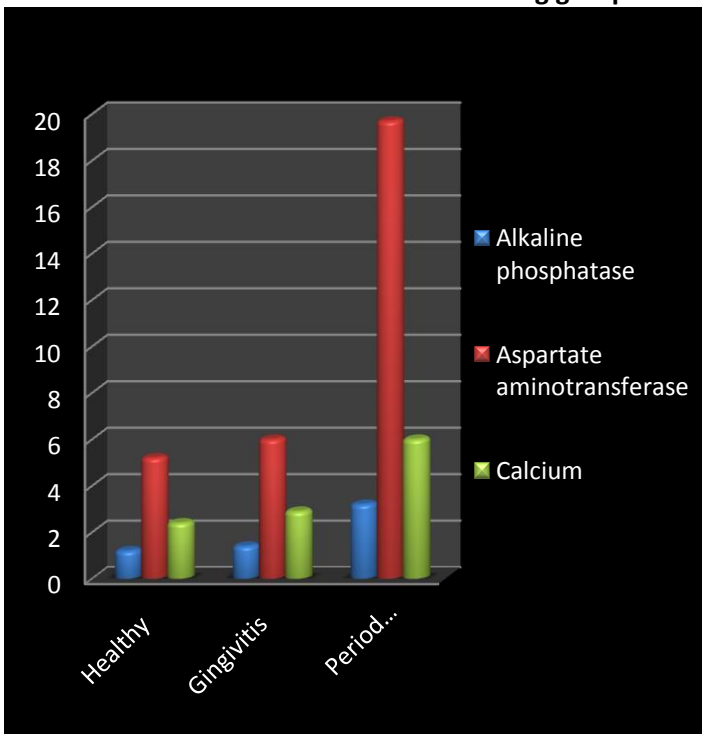
(B)Aspartate Aminotransferase Enzyme: [Table I] [Graph I]: The salivary aspartate aminotransferase in group I had a mean value of 5.205 U/L with a standard deviation of 1.385. In the group II the mean value was 6 U/L with a standard deviation of 2.106. In the group III the aspartate aminotransferase enzyme had a mean value of 19.725 U/L with a standard deviation of 2.199.

(C) Calcium: [Table I] [Graph I]: The salivary calcium in group I had a mean value of 2.435 mg/dl with a standard deviation of 0.708. In the group II the mean value was 2.890 mg/dl with a standard deviation of 0.689. In the group III the calcium had a mean value of 6.035 mg/dl with a standard deviation of 0.870.

Table:1 Comparision of Alkaline Phosphatase, Aspartate Aminotransferase and Calcium in Group I, Group II and Group III Groups (Anova)

	Group I	Group II	Group III	P-Value
Alkaline Phosphatase (U/L)	1.205 ± 0.248	1.440 ± 0.239	3.195 ± 0.268	0
Aspartate Aminotransferase (U/L)	5.205 ± 1.385	6.000 ± 2.106	19.725 ± 2.199	0
Calcium (mg/dl)	2.435 ± 0.708	2.890 ± 0.689	6.035 ± 0.870	0

Graph- 1: Alkaline phosphatase, Aspartate aminotransferase and Calcium levels among groups



Discussion: Alkaline phosphatase is an intracellular enzyme present in most tissues and organs, particularly in bones. In our study we found that the periodontitis group had the highest alkaline phosphatase levels 3.2 U/L with a standard deviation of 0.27. This was statistically highly significant (p=0.000). The increased activity in the saliva is probably the consequence of destructive processes in the alveolar bone in the advance stages of development of periodontal disease. The results are in accordance to the results obtained by Todorovic T,

Dozic I, Vicente-Barrero M et al. (2006)[7] who compared the enzyme activities in subjects with periodontitis and in healthy controls. Similar results were also obtained by Nakamura and Slots (1983)[8] who found that enzyme activity is higher in patients with periodontal disease. Totan A, Greabu M, Totan C et al.[9] also confirmed these findings in 2006. They found that periodontal destruction such as periodontal pockets; gingival bleeding and suppuration were related to higher alkaline phosphatase (ALP) levels. They concluded that the increase in salivary ALP activity in periodontitis demonstrated could be associated with alveolar bone loss, a key feature of periodontal disease. Nomura Y, Tamaki Y, Tanaka T et al. (2006)[10] also had similar findings when they tested the salivary enzyme activity in 187 subjects.

Similarly when aspartate aminotransferase (AST) levels were analyzed they were found to be highest in the periodontitis group. The increased activity of aspartate aminotransferase (AST) indicates pathological changes located in the soft tissues only, primarily in the gingiva which could coincide with the initial stage of periodontal disease[7]. Our findings corroborated the findings of Cesco R, Ito IY and Albuquerque Junior RF (2003)[11], they found that aspartate aminotransferase (AST) levels in saliva were higher in presenting code 4 of the CPITN than patients coded lower. Totan A, Greabu M, Totan C et al. (2006)[9] also found that salivary aspartate aminotransferase (AST) activity in patients with periodontal disease was significantly increased compared with controls. Nomura Y, Tamaki Y, Tanaka T et al. **(**2006)[10] also found similar results. They concluded that salivary aspartate aminotransferase (AST) were potentially useful for screening of periodontal disease. Todorovic T, Dozic I, Mario Vicente Barrero M et al. (2006)[7] also found statistically significant increases in the activity of aspartate aminotransferase (AST) in saliva from patients with periodontal disease in relation to control group. Based on these results, they concluded that activity of this enzyme in saliva, as biochemical markers for periodontal tissue damage, may be useful in diagnosis, prognosis and evaluation of therapy effects in periodontal disease.

The salivary enzymes can be considered as biochemical markers of the functional condition of periodontal tissues that provides new opportunities in making diagnosis.

The results showed that the subjects in the periodontitis group had the highest levels of salivary calcium, which was statistically significant. There was also a significant difference in the calcium levels between the gingivitis and healthy group with the higher level being in the gingivitis group. These results are consistent with the findings of Sewon L, Soderling E, Karjalainen S (1990)[12] who found that there was a higher calcium concentration in the saliva in the periodontitis subjects as compared to the periodontitis free subjects. They opined that periodontitis affected subjects had a higher re mineralization potential than individuals with no signs of periodontal disease. Similarly the results are also in accordance with those of Sewon and Karjalainen (1998)[13] who found that subjects with higher calcium levels had more bleeding on probing.

The results however were in contrast with the results obtained by Sewon L and Makela M (1990)[14] who found that higher calcium level was related to good dental health but there was no relation to periodontal bone destruction.

The calcium levels of saliva may also reflect the fluctuations in dietary calcium and general calcium turnover. In the present study the effect of diet could not be demonstrated. It seems that salivary calcium, due to its affinity for being readily taken up by plaque, is an important factor, with regard to onset of periodontitis[13]. The study showed a clear and a significant association between high salivary calcium levels and periodontal disease states.

Thus it can be suggested that the detection of increased alkaline phosphatase (ALP) enzyme, aspartate aminotransferase (AST) enzyme and calcium from saliva reflects the clinical status of the periodontal tissues. These tests appear to hold promise as useful test for periodontal diseases and can serve as adjuncts to conventional methods of diagnosis.

Conclusion: The observations of this study are as follows:

1. The level of alkaline phosphatase enzyme was found significantly higher in the periodontitis

group as compared to the gingivitis and healthy groups.
2. The level of aspartate aminotransferase enzyme was also found significantly higher in the periodontitis group as compared to the gingivitis and healthy groups.
3. Increasing calcium level was seen in all the groups. Periodontitis affected subjects had the highest mean calcium level. The increase in the calcium level in the periodontitis group was highly significant.

It can be concluded that the collection and analysis of saliva offers to resolve some but not all the technical concerns associated with gingival crevicular fluid based diagnostic tests. The test acceptance is likely to improve when clinicians view these tests as adjunct to patient care, i.e. the early identification of increased tissue inflammation and quantification of a therapeutic endpoint. A saliva based diagnostic test may also prove valuable in screening large populations.

Therefore saliva can be used as an aid in the diagnosis of periodontal disease by identifying the biochemical markers. It can not only serve as an important adjunct to clinical and radiographic diagnosis, but also as an aid for patient education.

Longer studies examining the relationship of these identified markers to the natural history of periodontal disease is the obvious next step in the line of investigation.

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