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Comparative evaluation of alpha amylase levels in serum and saliva of healthy individuals and subjects with chronic periodontitis

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Abstract
Introduction: Amylase is a key protein secreted in saliva and serum as a part of oral host immune response in periodontal disease. However very few documents are available on serum and salivary alpha amylase levels correlating the periodontal status in same patients. Hence to evaluate and compare the levels of alpha amylase in serum and saliva between chronic periodontitis and healthy patients.
Material and Methods: In this cross-sectional study, serum and saliva was collected from 40 patients between 25-60 years of age. They were divided into two groups Both the groups were evaluated for serum and salivary alpha amylase and clinical parameters were recorded
Results: The result of this study showed that salivary alpha amylase was significantly higher in chronic periodontitis group as compared to healthy control group. Serum alpha amylase was equally prevalent in both the groups.
Conclusion: There was an increase in salivary alpha amylase levels in chronic periodontitis patients as compared to the healthy group but the serum alpha amylase level was constant in both the groups. Thus present results have documented that salivary alpha amylase can be used as a biomarker to link new emerging diagnostic technique with novel therapeutic approach.

Keywords: alpha amylase, chronic periodontitis, saliva, serum, biomarker

Introduction
Periodontal diseases reflect the interplay between a pathogenic bacterial biofilm, present on the root surface/periodontal pocket and host-derived inflammatory cells and molecules from periodontal tissue. This process results in the loss of connective tissue and bone support and is a major cause of tooth loss in adults. [5]Saliva is composed of a complex mixture of secretary products (organic and inorganic) primarily secreted by salivary glands. It is an important resource for evaluating physiological and pathological conditions [23]. Salivary alpha amylase is a highly abundant protein in saliva [24]. In the normal conditions salivary amylase is present at low concentration in whole saliva forming an efficient molecular defense netork of the oral cavity. However, during inflammatory conditions, salivary glands may respond to these inflammatory conditions by increasing the synthesis of salivary amylase as a result of which the protective potential of saliva is enhanced. This reason explains that saliva in chronic periodontitis patients shows increase amylase concentration [25]. Over 2, 000 proteins have been identified in the saliva, which approximately 2530% of them are shared with the serum. The serum amylase concentration reflects the balance between the rates of amylase entry into and removal from the blood. Elevated amylase levels can result either from an increased rate of entry of amylase into the circulation and/or a decreased metabolic clearance of this enzyme. [1]The aim of the study was to compare the levels of alpha amylase in serum and saliva between chronic periodontitis group and healthy control group and its correlation with clinical parameters associated with health and disease.

Materials and Methods
The study protocol was approved by the institutional ethicl committee with the institutional ethical clearance no. EC201705. A total of 40 patients were selected from the out patient department of Periodontology, Peoples College of Dental Sciences and Research Centre, Bhanpur, Bhopal.

Pts was divided into two groups

Group A: 20 systemically healthy patients with chronic periodontitis.

Group B: 20 periodontally and systemically healthy patients (controls).

Inclusion criteria and exclusion criteria

Subjects with moderate to severe periodontitis and without periodontitis, without any systemic diseases aged between 25-60 years of age and with a probing pocket depth of ≥5mm were included in the study. Subjects with any systemic diseases, on antibiotic therapy, smokers and subjects already undergone periodontal therapy were excluded from the study.

Study desgin

The nature and purpose of the study was explained to the patients and an informed consent was obtained from patients and clinical parametres like plaque index, gingival gingival index, simplified oral hygiene index, probing pocket depth, gingival recession/enlargement, clinical attachment level index, simplified oral hygiene index, probing pocket depth, gingival recession/enlargement, clinical attachment level were recorded. Approximately 2ml of unstimulated saliva was collected in a sterile disposable plastic container by spitting method and serum was collected by witdrawing 5 ml of venous blood from the antecuboital vein. After collection, serum and saliva samples were send to the laboratory for further biochemical analysis.

Statistical analysis

The data obtained was subjected to statistical analysis with the consult of a statistician. The data so obtained was compiled systematically. A master table was prepared and the total data was subdivided and distributed meaningfully and presented as individual tables along with graphs. Data were analyzed using chi –square test, student t-test and analysis of variance (ANOVA), Pearson correlation coefficient, and multiple regression analysis with (SPSS Version 21;IBM Corporation, Armonk, New York, USA). The values are expressed as mean± SD. A. P value of < 0. 05 was considered to be significant. P value of < 0. 01 was considered to be highly significant and P value of < 0. 001 was considered very highly significant.

Results

The mean ginigival index score, plaque index score, simplified oral hygiene index score, mean probing pocket depth, mean clinical attachment level, mean gingival recession (p<0. 001) were highly significant in chronic periodontitis group as compared to healthy control group. Mean salivary alpha amylase level (p <0. 001) was highly significant in chronic periodontitis group as compared to healthy control group while there was no significant difference in serum alpha amylase level (P>0. 05) between chronic periodontitis group and healthy control group. In chronic periodontitis and healthy control group there was a weak correlation between serum and salivary alpha amylase levels and hence the result of our study showed that the necessary increase in salivary alpha amylase level will not lead to the increase in serum alpha amylase levels both in chronic generalized patients and healthy controls.

Table 1: Clinical parameter score and level of serum and salivary alpha amylase level in chronic periodontitis group

Variable	Mean	Sd	Range	T value	P value
Serum α amylase	66. 50	15. 860	30-92	0. 099	0. 922
Salivary α amylase	205200. 00	260195. 985	5600-764000	3. 526	0. 001
Gingival index	2. 0330	0. 52683	0. 96-2. 69	12. 099	0. 001
Plaque index	1. 7935	0. 41928	0. 94-2. 58	12. 413	0. 001
Simplified oral hygiene index	0. 7685	0. 19378	0. 39-1. 07	13. 314	0. 001
Probing pocket depth	5. 4250	0. 38432	4. 70-6. 04	28. 644	0. 001
Clinical attachment level	5. 9390	0. 52240	4. 88-6. 93	27. 127	0. 001
Gingival Recession	0. 5875	0. 15522	0. 29-0. 85	16. 927	0. 001

Table 2: Clinical parameter score and level of serum and salivary alpha amylase level in healthy control group

Variable	Mean	Sd	Range	T value	P value
Serum α amylase	66. 05	12. 738	37-86	0. 099	0. 922
Salivary α amylase	46. 35	10. 230	30-69	3. 526	0. 001
Gingival index	0. 5315	0. 17458	0. 22-0. 77	12. 099	0. 001
Plaque index	0. 5450	0. 16292	0. 30-0. 80	12. 413	0. 001
Simplified oral hygiene index	0. 1400	0. 08379	0. 03-0. 35	13. 314	0. 001
Probing pocket depth	2. 2270	0. 31875	1. 53-2. 67	28. 644	0. 001
Clinical attachment level	2. 2270	0. 31875	1. 53-2. 67	27. 127	0. 001
Gingival Recession	0. 0000	0. 0000	0. 0-0. 0	16. 927	0. 001

Table 3: Comparison of pearson correlation coefficient and significance P value in serum and salivary alpha amylase levels of chronic periodontitis patients and healthy control group.

	Pearson's Correlation Coefficient	Significance P value	Inference
Chronic periodontitis(Serum and salivary alpha amylase)	0. 211	0. 372	Weak Correlation
Healthy control group (Serum and salivary alpha amylase)	0. 210	0. 375	Weak Correlation

Discussion

Periodontal diseases are chronic inflammatory disorders in which numerous inflammatory and immune mediators are released in response to bacteria and bacterial products. Patients with periodontal disease have differences in the protein composition of whole saliva as it is an important component of host oral immune defense [2]. The enzyme alpha amylase is one of the key protein which accounts for 60% of all proteins produced by the salivary gland. In our study, the level of serum and salivary alpha amylase in healthy

individuals and subjects with chronic periodontitis was compared in 40 patients Various periodontal clinical parameters used in the study include gingival index (GI), plaque index (PI), Simplified oral hygiene index (OHI-S), Probing pocket depth, Gingival recession, Clinical attachment level, and biochemical parameters used include serum and salivary alpha amylase level. The results of our study depict that the gingival index score was more in chronic periodontitis group compared to healthy control group which is at par with the observations made by Ahmadi F *et al* (2017) [1] determine the relationship between serum and salivary alpha amylase levels in chronic periodontitis group and healthy group in which clinical parameters like gingival index, clinical attachment loss, were used as a complete periodontal examination. The plaque index, simplified oral hygiene index and the clinical attachment level score was more in chronic periodontitis group as compared to healthy control group which is at par with the observation made by Neha T (2018) [23] conducted a cross-sectional study in total 45 subjects and all groups were evaluated for salivary alpha amylase and other clinical parameters i. e. plaque index, simplified oral hygiene index, periodontal probing pocket depth, clinical attachment level at baseline and 6 weeks after scaling. The mean probing pocket depth score was more in chronic periodontitis group as compared to the healthy control group which is at par with the observations made by Sanchez GA, Miozza (2013) [5] in which the result of this study showed that salivary mucin and amylase showed a positive correlation with probing pocket depth and clinical attachment level before periodontal treatment and after treatment. The gingival recession was more in chronic periodontitis group compared to healthy control group. The results of our study also indicates that salivary levels of alpha amylase levels were significantly higher in chronic periodontitis group but the serum level were equivalent in both the groups. Hence the increase in salivary alpha amylase levels will not necessarily lead to increase in serum alpha amylase levels which is at par with the observations made by Ahmadi F *et al* (2017) [1] who compared the serum and salivary alpha amylase levels between chronic periodontitis group and healthy control group and concluded that salivary alpha amylase was significantly higher in chronic periodontitis group compared to the control group but serum alpha amylase was not statistically significant in chronic periodontitis group as compared to healthy control group.

Summary and conclusion

A study was conducted on 40 patients (20 systemically healthy patients with chronic periodontitis and 20 controls) from the outpatient department of Periodontology in Peoples college of Dental Science and Research centre with the aims and objectives to evaluate the levels of serum and salivary alpha amylase levels in patients with and without chronic periodontitis as well to evaluate whether serum and salivary alpha amylase concentrations correlate with clinical parameters such as plaque index, gingival index, simplified oral hygiene index, probing pocket depth, gingival recession/enlargement, clinical attachment level, associate with periodontal disease and health. The results of our study depicted that mean gingival index, plaque index, simplified oral hygiene index score, probing pocket depth, clinical attachment level, gingival recession was more in chronic periodontitis group as compared to healthy control group. serum alpha amylase levels are equal in chronic periodontitis patients and healthy individuals. Salivary alpha amylase levels is elevated in chronic periodontitis patients as compared to healthy individuals and the increase in salivary alpha amylase levels will not necessarily lead to increase in serum alpha amylase levels and hence there was a weak correlation between serum and salivary alpha amylase level in healthy individuals and in subjects with chronic periodontitis. Salivary alpha amylase level increases as the severity of periodontal disease increases. Level of salivary alpha amylase was found to be correlated with the clinical parameters recorded in the study. In conclusion, present results have documented that salivary alpha amylase can be used as a biomarker to link new emerging diagnostic technique with novel therapeutic approach.

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