

ASSESSMENT OF SALIVARY ALKALINE PHOSPHATASE IN PATIENTS WITH HEALTHY GINGIVA ON REDUCED PERIODONTIUM VERSUS PERIODONTITIS

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Abstract

Background: Alveolar bone loss is a significant factor in the breakdown of tissues in periodontal disease. The condition compromises the structural integrity of alveolar bone by altering its microenvironment. Numerous molecular activities, such as Alkaline Phosphatase (ALP), control the process.

Aims: This study aims to assess the level of salivary Alkaline Phosphatase with healthy gingiva on the reduced periodontium and compare them to their level in periodontitis patients.

Patients and Methods: Seventy five participants, both male and female, who took part in this study had their salivary ALP assessed. There were three groups formed from the participants: the first group clinically healthy periodontium control group (n=15), second group consisted of generalized periodontitis stage II and III (n=30), and third group healthy gingiva on reduced periodontium (n=30). Each group systemically healthy. The entire unstimulated salivary samples were taken, and all subjects underwent a periodontal evaluation, which included the assessment of clinical periodontal parameters (PLI, GI, BOP, PPD, and CAL). The concentration of ALP in saliva is measured using the ELISA method.

Results: A moderate positive significant correlation was found between ALP with CAL in periodontitis and positive weak significant correlation between ALP and CAL in excessive brushing group, as well as; the result revealed that The mean of salivary ALP was significantly lower in the control group (20.33 IU/L) than both the periodontitis and the excessive brushing group, while between periodontitis (72.50 IU/L) and excessive brushing (68.73 IU/L) there was no significant difference.

Conclusion: Healthy gingiva on reduced periodontium caused by excessive brushing is associated with high salivary levels of ALP.

Keywords: ALP, Biomarkers, Reduced periodontium, Saliva

Introduction

According to the definition of periodontal health as a condition free of inflammatory periodontal disease, which enables a person to operate normally and prevents negative mental or physical effects from a present or past

disease, It may comprise individuals who have a history of effectively treating gingivitis, periodontitis, or other periodontal disorders who have been able to preserve their dentition without showing any evidence of clinical gingival inflammation¹.

In the presence of reduced clinical attachment and bone levels, clinical gingival health on a reduced periodontium is indicated by the lack of bleeding on probing, erythema, edema, and patient symptoms.

There is a difference in the risk for periodontal disease progression between a case of gingival health on a reduced periodontium in a patient with stable periodontitis and a case of periodontal health on a reduced periodontium in a patient who does not have periodontitis (recession, crown lengthening) ¹.

The movement of the marginal gingiva to an apical level, utilizing the cemento-enamel junction (CEJ) as a point of reference, is known as gingival regression (GR), losing the periodontal attachment system and exposing the root surface. Numerous variables, which can be categorized into three categories: anatomical (dehiscence of the alveolar bone and incorrect positioning of the teeth), physiological (orthodontic motions), and pathological (including traumatic brushing), might be implicated in generating or aggravating the GR ².

Patients' main aesthetic concerns are related to gingival recession, which puts them at greater risk of developing root caries and sensitivity ³.

Inflammation of the teeth's supporting tissues, increasing loss of attachment, and bone loss are all symptoms of the periodontitis^{4&5}. With a global frequency of 11.2%, it is the sixth most prevalent disease ⁶.

Alveolar bone loss is a significant factor in the breakdown of tissues in periodontal disease. Alveolar bone's structural integrity is compromised as a result of the disease's alteration of its microenvironment. The process is influenced by a number of molecular events, such as (ALP). ALP is one of the intracellular degradation enzymes that has drawn the greatest interest as a potential indicator of ongoing periodontal destruction⁷. ALP plays a crucial role in the periodontium's proper turnover of the periodontal ligament, alveolar bone development, and root cementum maintenance⁷. Numerous cells, including neutrophils, fibroblasts, osteoblasts, and osteoclasts, generate ALP. ALP works to create the pH at which a molecule has a net neutral charge in the bone matrix, which causes hydroxyapatite to crystallize⁸. ALP may

serve as a measure of bone turnover, reflecting the severity and progression of periodontal disease ⁹.

Patients and Methods:

The ethics committee of the University of Baghdad's College of Dentistry gave their approval to the protocol on April 17, 2022 (Reference number: 522, Project number: 52262). Each subject was told of the study's objectives and procedures, and they had the option of participating in the study or not. The study's human sample comprised of (75) male and female study participants who were seeking periodontal care at the university of Baghdad's department of periodontology, college of dentistry (between February 2022 to June 2022).

Three groups were created from the study subjects:

1-Control group I (healthy periodontium): Consists of fifteen (15) systemically healthy and with healthy periodontium, this group presents baseline data for salivary ALP.

2_ Study group II (generalized periodontitis stage II and III): interproximal bone loss involving the coronal and middle third of the root, consisting of thirty (30).

3_Study group III (healthy gingiva on reduced periodontium): bone loss involving a coronal and middle third of the root consists of thirty (30) patients. Reduced periodontium due to excessive tooth brushing only (stiff tooth brush and horizontal brushing technique)

According to Tenovuo (1994)(10), entire saliva was collected after the initial periodontal examination. To collect 5 ml of saliva, the individual drooled passively into a 10 ml centrifuge tube. The sample was then put in a cooler box to be centrifuged later. Following the collection of saliva, a thorough periodontal examination was conducted to document the clinical periodontal parameters (PLI, BOP, PPD, and CAL). The salivary sample is then centrifuged at 3000 rpm for 20 minutes, conserved in a plane tube, and kept in a freezer at -20°C in order to be tested later with an ELISA kit to determine the salivary levels of ALP.

Using the ELISA technique, a kit made by BIONT, Catalog No: YLA165, China, was used to perform the biochemical analysis of salivary ALP.

Statistical Package for Social Science was used for data description, analysis, and presentation (SPSS version - 22, Chicago, Illinois, USA).

Results:

The mean of salivary ALP was significantly lower in the control group than both the periodontitis and the excessive brushing group, while between periodontitis

and excessive brushing there was no significant difference as shown in Table I and Table II. Levene p value=0.000 Sig.

Table 1 : Descriptive and statistical test of ALP among groups.

Groups	Mean ±SD	±SE	Minimum	Maximum	F	P value
Control	20.33±5.41	1.40	11.50	29.44	55.38	0.00
Periodontitis	72.50±17.30	3.16	48.24	105.70		
Excessive brushing	68.73±19.27	3.52	33.48	103.45		

Table II: Multiple Comparisons of ALP among groups using Games-Howell posthoc

(I) Groups	(J) Groups	Mean Difference	p value
Control	Periodontitis	-52.17	0.00
	Excessive brushing	-48.40	0.00
Periodontitis	Excessive brushing	3.77	0.70

Regarding Pearson’s correlation coefficient, Table III demonstrated that there was

non significant correlation between % PLI and ALP

Table III: Correlation between, ALP with(PLI and BOP)

Groups		ALP	
		r	p
Control	% PLI	0.24	0.39
	% BOP	0.17	0.55
Periodontitis	% PLI	0.13	0.50
	% BOP	0.07	0.71
Excessive brushing	% PLI	0.21	0.27
	% BOP	0.02	0.93

Table IV demonstrated that there was moderate positive significant correlation between ALP with CAL in periodontitis, while correlations between ALP and CAL in excessive brushing group was positive weak significant correlation.

Table IV also demonstrated that there was weak positive not significant correlation between PPD and ALP in periodontitis group.

Table IV: Correlation between ALP with (PPD and CAL)

Groups		CAL		PPD	
		r	p	r	p
Periodontitis	ALP	0.65	0.00	0.02	0.93
Excessive brushing	ALP	0.46	0.01		

Discussion:

Osteoclast recruitment to the periodontal tissues occurred in the excessive brushing group as a result of force- and time-dependent loading of 20 to 40 N,

Therefore, it is plausible to suppose that bone cells may respond as a result of mechanotransduction caused by vigorous and frequent tooth brushing¹¹.

The biochemical analysis of ALP results revealed a highly significant difference between the control and study groups. ALP had the lowest mean value in the control group, but the mean value had elevated in periodontitis and reduced periodontium groups. This may be explained by the fact that alkaline phosphatase enzyme is an intracellular enzyme found in most tissues and organs, particularly in bones. They may have enhanced activity as a result of alveolar bone-damaging processes. ALP, a phenotypic marker of bone turnover rate and one of several biomarkers of periodontal disease activity, has been discovered to be raised in a number of bone illnesses¹².

According to certain research, ALP activity was noticeably enhanced during periodontal disease, and following periodontal therapy, it returned to levels seen in healthy individuals¹³. These intracellular enzymes are increasingly discharged into the GCF and saliva, where their activity can be monitored, when a periodontal tissue gets ill or has cells that are injured owing to edema or the destruction of a cellular membrane, or of a cell as a whole¹³. ALP is a significant periodontal enzyme that is essential for the process of bone resorption. Increased ALP activity in saliva may be a sign of periodontal disease, especially when there is bone loss¹⁴. The increased ALP activity shows that the alveolar bone had been damaged by the pathological destructive process, which signifies that periodontal disease had greatly progressed¹⁴. Cells like fibroblasts, osteoblasts, and osteoclasts can also create ALP. The cell membrane of these cells will

break during the periodontitis active stage, releasing the intracellular contents outside and increasing the secretion of ALP in the periodontal patient's saliva and gingival crevicular fluid¹⁵. Increased ALP activity may be a result of damaging processes occurring in the alveolar bone during advanced stages of periodontal disease¹⁶. These findings are consistent with prior research' findings that indicated that patients with chronic periodontitis exhibit increased salivary ALP activity compared to gingivitis or healthy people¹⁷⁻¹⁹. but disagree with Daltaban et al. 2006 (20) discovered no appreciable variations in the ALP concentrations between the periodontitis and control groups. Concerning the Pearson's correlation coefficient between ALP and clinical periodontal parameters in study groups, there was moderate positive significant correlation between ALP with CAL in periodontitis group. The cause of this outcome is a membrane-bound glycoprotein that is produced by a variety of cells in the periodontium, including polymorphonuclear leukocytes, macrophages, fibroblasts, and osteoblasts¹². The normal immune reaction to alveolar bone loss causes the ALP concentration to positively correlate with CAL. Our results agreed with Ray et al 2007²¹ concluded that ALP and CAL had a significant correlation. The small sample size resulting from the exclusion criteria was a limitation of this study. Although this study's results were noteworthy, further research should be done with a larger sample size and site-specific techniques for more accurate findings.

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Availability of Data and Material:

The corresponding author is prompt to supply datasets generated during and/or analyzed during the current study on wise request

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