



RESEARCH ARTICLE

SALIVARY ALKALINE PHOSPHATASE ESTIMATION IN CHRONIC PERIODONTITIS IN SMOKERS AND NON-SMOKERS: A BIOCHEMICAL STUDY

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ARTICLE INFO

Article History:

Received 13<sup>th</sup> April, 2018  
Received in revised form  
19<sup>th</sup> May, 2018  
Accepted 28<sup>th</sup> June, 2018  
Published online 31<sup>st</sup> July, 2018

Key words:

Salivary Alkaline Phosphatase,  
Chronic Periodontitis,  
Smokers, Non-Smokers.

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DOI: <https://doi.org/10.24941/ijcr.30808.07.2018>

ABSTRACT

**Background:** Periodontal disease is a chronic disease of the oral cavity comprising a group of inflammatory conditions affecting the supporting structures of the dentition. Numerous markers in saliva have been proposed as a diagnostic tests for periodontal disease such as intracellular enzymes and alkaline phosphatase is one amongst them. The purpose of this study was to determine the salivary levels of alkaline phosphatase (ALP) activities in patients with periodontal disease in smokers and non-smokers, and to compare them with ALP in healthy individuals.

**Materials and Methods:** In this study, we examined the activities of salivary ALP in 75 patients ageing between 20-50 years. The experimental groups consisted of 50 Chronic periodontitis patients (25 with smoking and 25 without smoking history) and the control group had healthy subjects (25 samples). Unstimulated whole saliva collected from each subject was transferred to auto analyzer to measure the salivary alkaline phosphatase levels. The quantitative analysis of salivary alkaline phosphatase levels of samples obtained from subject groups was done.

**Results:** ALP level were higher in the subjects of chronic periodontitis in smokers habit when compared to non-smokers and healthy individuals

**Conclusion:** Salivary level of Alkaline Phosphatase was significantly elevated in patients with chronic periodontitis in smokers as compared to non-smokers and healthy individuals and can be considered as a biomarker for periodontal disease.

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Citation: Dr. Mamta Malik, Ravinder S Saini, Dr. Sanjeev Laller, Dr. Kanwalpreet Kaur, Dr. Pooja Sheokand and Dr. Nisha Dahiya 2018. "Salivary alkaline phosphatase estimation in chronic periodontitis in smokers and non-smokers: A biochemical study", *International Journal of Current Research*, 10, (07), 71825-71827.

INTRODUCTION

Alkaline phosphatase (ALP) also known as Orthophosphoric-monoester phosphohydrolase is a zinc-containing metalloenzyme; it is activated by Mg<sup>2+</sup> and other divalent ions. It has various isoforms, some being true isoenzymes, that is, encoded by different genes. It is widely distributed in the body, but is particularly associated with bone (osteoblasts), small intestine (mucosal cells), liver (cells of the biliary system), placenta and kidney (proximal convoluted tubules). (Coleman, 1992). Periodontal disease is a chronic bacterial disease of the oral cavity comprising a group of inflammatory conditions affecting the supporting structures of

teeth. Periodontitis is a multifactorial disease which is affected by both genetic and environmental factors. Diagnosis of periodontal disease is dependant mainly on clinical and radiographic parameters like gingival index, bleeding on probing, pocket depth and alveolar bone loss. These measures are useful in detecting evidence of past disease, and to verify periodontal health, but provide only scant clue about patients and sites at risk for future periodontal loss. Multiple markers in saliva have been proposed as a diagnostic tests for periodontal disease such as intracellular enzymes like alkaline phosphatase. If a periodontal tissue becomes weak and its cells become damaged, due to oedema or destruction of a cellular membrane, these intracellular enzymes are increasingly being released into

the gingival crevicular fluid and saliva where their activity can be measured (Newman, 2006; Agrawal, 2014 and Kumar, 2011). Tobacco smoking is a risk factor for many oral diseases, and many studies suggest periodontal health is also affected by smoking. Current literature suggests that smoking has direct relation with periodontal health and independent to oral hygiene index. Smoking plays a vital role as a risk factor for periodontitis and it was also proved that the potential risk reduces with stoppage of smoking habit. The response of an organism to the periodontal infection includes production of several enzymes and inflammation markers which can be analyzed both in serum as well as saliva. Literature have shown a correlation between high ALP levels and periodontitis (Newman, 2006 and Agrawal, 2014). So the present study was conducted with the aim to compare the salivary ALP level in healthy individual with patients having chronic periodontitis with and without the habit of smoking.

## MATERIALS AND METHODS

The present case control study was performed in the outpatient department of Oral Medicine, Diagnosis and Radiology Department of PDM Dental College and Research Institute, Bahadurgarh, India. The study was conducted after getting ethical clearance from institutional ethical committee. A written informed consent was taken from each subject. Total 75 subjects of age group 20-50 having at least 20 teeth were included in the study. Group-I included 25 healthy patients without periodontitis, Group-II included non-smokers with chronic periodontitis and Group-III included smokers with chronic periodontitis. Study samples taking medicines known to affect periodontal conditions or gingival secretion, having cardiac disease, hepatobiliary disease, diabetes, thyroid and parathyroid abnormalities, Viral, fungal or bacterial infection, having recent trauma or tooth extractions pregnant or lactating women, history of systemic antibiotic therapy within 6 months were excluded from study because the pathophysiology of periodontium gets affected in such conditions. A detailed case history of each subject was taken and for clinical examination minimum 18 teeth in each subject were examined for the depth of periodontal pocket with the help of William's periodontal probe. A minimum of one pocket with probing depth of 6 mm or above was considered as chronic periodontitis. 1 ml of whole saliva sample were collected in a sterile disposable plastic container with patients instructed not to eat 1 hour before collection of sample. Thereafter immediately salivary samples were sent to the laboratory, the saliva samples were centrifuged at 1000 rpm for 15 minutes, the quantitative analysis of alkaline phosphatase levels by using auto-analyzer was done (Kishore, 2014). The result obtained were tabulated and shown in figures after the statistical analysis for significance difference between groups was done.

## RESULTS

The obtained results from the study showed the potential value of salivary alkaline phosphatase level as indicator for periodontal disease status in smokers. The quantitative levels of salivary alkaline phosphatase may distinguish varying periodontal condition in smokers and non-smokers as compared to healthy individuals. The mean value of ALP levels in healthy individuals were found  $23.96 \pm 2.97$  as compared to Non-smokers with chronic periodontitis in which mean ALP levels were found  $64.40 \pm 5.71$  as shown in table-1 and Figure-1, and the results were found significant. Similarly the ALP

levels in smokers with chronic periodontitis were  $75.57 \pm 4.66$  which were found significant when compared with healthy individuals (Table-2 and Figure-1) and non-smokers with periodontitis group (Table-3 and Figure-1).

**Table 1. Shows comparison between healthy and Non-smokers**

GP	Mean	Std. Deviation	P value	Significance
Healthy	23.9676	2.97155	0.001	Significant
Non-smokers with PD	64.4048	5.71437		

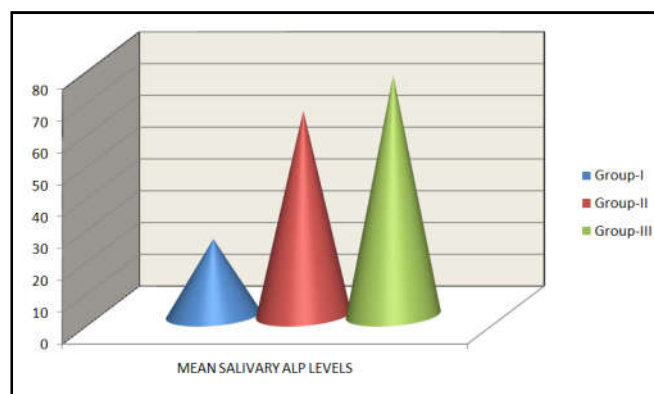
GP = Group, PD = Periodontitis

**Table 2. Shows comparison between healthy and smokers**

GP	Mean	Std. Deviation	P value	Significance
Healthy	23.9676	2.97155	0.001	Significant
Smokers with PD	75.5700	4.66648		

**Table 3. Shows comparison between Non-smokers with PD and Smokers with PD**

GP	Mean	Std. Deviation	P value	Significance
Non-Smokers with PD	64.4048	5.71	0.001	Significant
Smokers with PD	75.5700	4.66		



**Figure 1. Mean Salivary ALP levels in all study groups**

## DISCUSSION

The etiopathophysiology of periodontitis with features of loss of connective tissue and pocket formation due to loss of alveolar bone; is complex with multiple bacterial involvements and with the role of various biological substances. The diagnosis of periodontitis is mainly done with the clinical and radiographic finding but salivary enzymes also plays an important role in diagnosis along with future control of periodontal health. One among such enzyme is alkaline phosphatase which acts as indicator of progressed inflammation and periodontium cellular damage because more amount of ALP is released from the damaged cells of periodontium due to altered metabolic changes (as ALP is produced by PMNs, osteoblasts, macrophages, fibroblasts of periodontium and plaque bacteria within periodontal pocket). (Kumar, 2011 and Kishore, 2014). Earlier studies were also done where serum and gingival crevicular fluid (GCF) was used for estimation of ALP levels. But saliva is thought to be better as compared to above two, as estimation from serum is invasive and usually patients do not prefer needle prick and GCF though reflects much closer levels of ALP but the technique of collecting GCF is complicated and not easy and feasible in routine dental practice (Kishore, 2014). The severity and rate of progression and destruction of

periodontium is accelerated with the habit of tobacco smoking. Though the periodontal destruction is more due to smoking by affecting the normal host response in neutralization of infection, but the reduction in gingival bleeding and redness of gingival is seen due to vasoconstrictive effects of nicotine in tobacco which results in misdiagnosis of periodontitis. Thus salivary ALP could be more beneficial in diagnosis of periodontitis than clinical parameters (Agrawal, 2014).

Our study was conducted with the aim of evaluation and comparison between the salivary ALP levels in chronic generalized periodontitis with smokers, non smokers and healthy controls. Till date many studies are conducted for detection of levels of ALP in GCF and saliva and in serum. But studies comparing and correlating ALP levels in healthy and periodontitis smokers and non-smokers are very scant. ALP levels can be detected in saliva of patients with periodontitis. In our study there was a significant correlation of increased levels of salivary ALP in chronic periodontitis as compared to healthy patients. There was a highly significance correlation of high levels of serum ALP in chronic periodontitis with smokers and non-smokers (Agrawal, 2014; Lubaba, 2015 and Mohammad, 2015). Our results are in accordance with Lubaba A and colleagues who suggested that suppression of salivary Osteocalcin levels by smoking and slight increase in alkaline phosphatase in smokers groups, may explain the deleterious effects of smoking on periodontal health status. Also Mohammad K et al concluded that it is beneficial to use both GCF and salivary levels of ALP as diagnostic markers for periodontitis. Higher levels of ALP in GCF samples of smokers with periodontitis could explain the higher rate of alveolar bone destruction in smokers (Lubaba, 2015 and Mohammad, 2015).

Also Agrawal N and his colleagues demonstrated similar results with serum ALP levels and reported that ALP levels can be detected in serum of patients with periodontitis. There was a highly significant correlation of increased levels of serum ALP in chronic periodontitis as compared to healthy patients. There was a highly significance correlation of high levels of serum ALP in chronic periodontitis with smokers and non-smokers (Agrawal, 2014). Almost similar results were found by Kumar R, Sharma G, and Kishore P K and his colleagues who also found increased levels of salivary ALP levels in gingivitis and periodontitis as compared to healthy control group and were significantly and positively correlated with probing depth and gingival index (Kumar, 2011 and Kishore, 2014). Similar conclusions were made by Nakamura M and Slots J in their research work. Ishikawa and Cimasoni demonstrated positive correlation of alkaline phosphatase in periodontitis patient with increased pocket depth. Also Chapple IL, Chemiluminescent also made similar observations when they evaluated ALP levels in gingival crevicular fluid. Binder et al demonstrated that ALP concentration in GCF showed a positive relationship with attachment loss (Nakamura, 1983; Mandel, 1991; Chapple, 1996 and Binder, 1987).

Finally the results of our study dictated positive correlation between increased ALP levels in periodontitis patients as compared to healthy and non smokers. Increased ALP levels indicates advancing periodontal tissue injury and damage and can be helpful in early management of chronic periodontitis in smokers where the clinical parameters are obscure due to nicotine effect.

### Conclusion

Salivary Alkaline phosphatase levels are increased with the increase in inflammation and destruction of periodontal tissues. Thus their levels can be used as clinical biomarkers in diagnosis and management of periodontal disease in patients with and without smoking habit.

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