# Alkaline Phosphatase Levels Before and After Nonsurgical Periodontal Therapy

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## ABSTRACT

**Introduction:** Chronic periodontitis is a multifactorial disease resulting in the inflammation and destruction of the supporting structures. Early detection of periodontal changes, prognosis and efficacy of treatment have been monitored by Alkaline Phosphatase Levels (ALP) levels.

Objective: This study was carried out to determine level of ALP in saliva and serum before and after periodontal therapy.

**Methods:** This pretest posttest study included 22 patients with generalised chronic periodontitis (GCP). The patients received nonsurgical periodontal therapy (NSPT). Saliva and serum levels of ALP were measured at baseline and after two months of periodontal therapy.

**Results:** Twenty-two patients of mean age 44 years were analysed. Participants had significantly better periodontal parameters after two months. Salivary ALP levels, which were high at baseline, decreased after periodontal treatment. A significant positive correlation (0.0001) was found between the salivary levels of ALP and periodontal inflammatory conditions. Gingival index was found directly proportional with salivary ALP level but not with serum ALP.

**Conclusion:** Periodontal therapy lowered the levels of ALP saliva in GCP patients with high ALP levels. Biochemical analysis of enzymes found in saliva may help in patients' evaluation to determine the control and progression of periodontal destruction and aid in a correct diagnosis, prognosis and, consequently, better treatment.

Keywords: Alkaline phosphatase; nonsurgical periodontal therapy; periodontal diseases; saliva; serum.

### INTRODUCTION

Chronic periodontitis is a multifactorial disease resulting in inflammation and destruction of supporting structures which may progress in unpredictable manner.<sup>1</sup> Periodontal disease concept has changed from slow, continuously progressing disease to random bursts attachment loss.<sup>2</sup>

Traditional periodontal diagnostic methods are efficient but current appraisal of disease status cannot be determined.<sup>3</sup> Biomarkers serve as indicators of biological health, pathogenic processes, environmental exposure, and pharmacologic responses to therapeutic intervention.<sup>4</sup> Alkaline phosphatase (ALP) is a membrane bound glycoprotein derived from leukocytes, osteoblasts, macrophages and fibroblasts.

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Abundance of polymorphonuclear leukocytes at site of periodontal inflammation serves as primary source for gingival crevicular fluid ALP. Early detection of periodontal changes, prognosis and efficacy of treatment can be monitored by ALP levels.<sup>5</sup>

The current clinical diagnostic parameters provide disease severity rather than disease activity.<sup>6</sup> Saliva sample which is easy to collect can substitute serum in diagnosis of oral conditions and ALP levels in saliva can be detected by using ultraviolet spectrometry.

Alkaline phosphatase is phenotype marker of bone turnover<sup>7</sup> which is yet to be considered as a predictive indicator for future periodontal tissue breakdown. The objective of this study was to determine ALP saliva and serum level before and after nonsurgical treatment.

## METHODS

The pretest posttest study was conducted at Department of Dental Surgery, Periodontology and Oral Implantology, National Academy of Medical Sciences (NAMS), Kathmandu, Nepal and consisted of 22 patients of generalised chronic periodontitis, who attended from April 2017 to June 2017. All the patients who had probing pocket depth  $\geq$  4 mm and

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clinical attachment level  $\geq 3$  mm in more than 30% of sites were included. Patient having at least 20 teeth of age 35-50 years were included. Ethical approval was taken from Institutional Review Board, NAMS and informed consents were taken from all the participants.

Participants suffering from any chronic inflammatory and infectious condition, pregnant and lactating mothers and post-menopausal females, those undergoing active periodontal therapy, current smokers and intake of antibiotics in last three months were excluded. Full mouth Gingival Index (Loe and Silness) and Plaque Index (Silness and Loe) were taken.

Gingival index and Plaque index (Silness and Loe 1964)8 of each tooth was examined. Five ml of whole saliva sample was collected by expectoration method in a sterile disposable plastic container by unstimulated passive drool. Saliva was collected according to a modification of the method described by Navazesh and Christensen (1982).9 Prior to saliva sample collection, the participant was instructed to refrain from intake of any food or beverage for two hours. Chewing gum was prohibited during this hour. The participant was instructed to minimise all facial movements, particularly movements of mouth. The participant was first asked to void mouth of saliva by swallowing to begin saliva collection and was then asked to lean slightly forward over tube. The subject was then instructed to keep his/her mouth slightly open and to allow saliva to drain into the biochemistry sample collection tube. At the end of the five minutes collection period, the participant was asked to collect any remaining saliva in his/her mouth and expectorate into the tube.

No brushing, no eating was allowed two hours before and no dental examinations were performed 48 hours before saliva collection. Though ALP activity remains stable for four hours in room temperature, the samples were stored at 4°C and sent to biochemistry lab. Venous blood was drawn in pathology laboratory for serum ALP level. After saliva collection was over, routine nonsurgical therapy including thorough scaling and root planing was carried out. After two months of nonsurgical therapy, baseline investigations were repeated in both saliva and serum.

**Principle of the assay**<sup>10</sup> - At pH 10.3 alkaline phosphatase catalyzes the hydrolysis of p-Nitro phenyl phosphate to yellow coloured p-Nitro phenol and phosphate. The change in absorbance measured by auto analyser at 405 nm wavelength is proportional to ALP activity in sample.

Data was collected using SPSS version 16. The Pearson correlation coefficient was calculated using correlation test

at 5% level of significance to see the correlation between saliva ALP level, Plaque Index and Gingival Index. P-value <0.05 was considered significant.

### RESULT

This clinical study was conducted to evaluate the levels of serum and salivary ALP in patients with generalised chronic periodontitis before and after nonsurgical periodontal therapy to determine periodontal disease activity. All clinical parameters were measured at baseline with saliva and blood samples collected on the same day (Table 1) and then 60 days after Phase I periodontal therapy. The collected samples were spectrometrically analysed for ALP levels.

ALP levels in saliva has been found significantly increased during active phase of disease followed by statistically significant reduction of ALP level after phase I therapy (Table 2). Baseline levels of ALP in saliva were higher in most of generalised chronic periodontitis cases, which is significantly reduced after periodontal therapy. The pattern of reduction of salivary ALP after scaling and root planing can be seen in Figure 2. The results from this study showed that serum ALP levels have no significant relationship with disease activity (Figure 1 and Table 3).

Difference between Plaque Index (PI), Gingival Index, Probing Pocket Depth (PPD), ALP serum level and ALP saliva values baseline and post treatment has been analysed (Table 4). Levels of saliva ALP change was positively correlated to probing pocket depth, plaque index and gingival index changes after periodontal therapy (Table 4). Baseline difference in levels of ALP in saliva was also statistically significant. ALP saliva values showed a significant decrease after NSPT at 60 days from (26  $\pm$  21.4) IU to (16.48  $\pm$  14.68) IU.

| Variables  | Minimum | Maximum | Mean± SD    |
|------------|---------|---------|-------------|
| ALP Saliva | 4       | 63      | 26.29±16.41 |
| ALP Serum  | 30      | 136     | 67.62±23.24 |

| Table 2. Saliva ALP levels before and after freduite |
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| Saliva ALP level | Mean ±SD          | p-value |  |
|------------------|-------------------|---------|--|
| Before treatment | 26±21.40          | 0.0001  |  |
| After treatment  | $16.48 \pm 14.47$ | 0.0001  |  |

## Table 3: Serum ALP level before and after periodontal therapy.

| Serum ALP level  | Mean        | p-value |  |
|------------------|-------------|---------|--|
| Before treatment | 67±13.79 SD | 0.1028  |  |
| After treatment  | 68±16.37 SD |         |  |

| Variables  | Pearson correlation (r) | p-value (p) |
|--|-------------------------|-------------|
| Change in Saliva ALP level versus change in Plaque Index   | 0.258                   | 0.2455      |
| Change in Saliva ALP level versus change in Gingival Index | 0.126                   | 0.5752      |
| Change in Saliva ALP level versus change in PPD            | 0.30                    | 0.1655      |
| Change in Serum ALP level versus change in Plaque Index    | -0.14                   | 0.5221      |
| Change in Serum ALP level versus change in Gingival Index  | -0.016                  | 0.9433      |
| Change in Serum ALP level versus change in PPD             | 0.03                    | 0.8774      |

 Table 4: Correlation of saliva ALP level, Serum ALP Level, Plaque Index and Gingival Index (r = Pearson correlation coefficient).



Figure 1: Inconsistent serum ALP level before and after treatment.



Figure 2: Consistently reduced saliva ALP level after treatment.

## DISCUSSION

Alkaline phosphatase is one of the enzymes produced by many cells within area of periodontium. Although it has been shown that the oral bacteria including some gram negative microorganisms which are typical for sub gingival plaque also produce this enzyme, Neutrophils are probably the main source of enzymes in the gingival sulcus.<sup>11</sup>

Serum ALP was not found to positively correlate with disease activity. The changes in the level of alkaline phosphatase in serum after NSPT was not statistically significant and it was also negatively correlated with clinical parameters like PI, GI and PPD. This could have been probably due to the fact that the local changes in periodontium may not have a direct effect on the levels of this enzyme in serum. Its changing concentration in serum depends on function of other organ systems like bone, kidney, liver, etc.<sup>12, 13</sup>

Various studies obtained results that have shown the activity of examined enzymes in saliva of the patients with periodontal disease was significantly higher with statistical significance of a high level (p < 0.001).<sup>14-24</sup>

Gibert (2003)<sup>25</sup> predicted ALP as an indicator for future loss of periodontium. It may serve as a marker in periodontal treatment planning and monitoring. Its level may also be useful as a potential bone turnover marker to establish the diagnosis and prognosis of periodontal disease. This study showed a positive and statistically significant correlation with ALP saliva level and not significant relation with serum ALP levels after phase I therapy which is in accordance to a study by Singh (2017).<sup>26</sup>

Jeyasree (2018) found the difference in salivary and serum ALP levels from baseline (79.55  $\pm$  6.40 and 97.62  $\pm$  4.17) to postoperative (49.47  $\pm$  5.11 and 85.40  $\pm$  4.10) to be statistically significant with P = 0.000 and 0.009 for saliva and serum, respectively.<sup>27</sup>

Because of the complex, multifaceted nature of periodontal disease, it is highly unlikely that a single biomarker will prove to be a stand-alone measure for periodontal disease diagnosis. More probable may be the development of an oral fluid-based diagnostic using a combination of host- and site-specific markers that accurately assess periodontal disease status.<sup>28</sup>

A new paradigm for periodontal diagnosis would ultimately affect improved clinical management of periodontal patients. Let us not be limited to conventional disease diagnosis techniques that lack the capacity to identify highly susceptible patients who are at risk for future breakdown.

#### CONCLUSION

Periodontal therapy led to reduction of ALP saliva in GCP patients who had high ALP levels. Biochemical analysis of

enzymes found in saliva may help patient's evaluation to assess periodontal disease status and to establish a correct diagnosis, prognosis and, consequently, better treatment. Such tool can be helpful in a developing country like Nepal where chair side diagnostic tool is expensive to perform as the prevalence of periodontal disease is high.<sup>29</sup>

Conflict of Interest: None

#### REFERENCES

- 1. Newman MG, Takei H, Klokkevold PR, Carranza FA. Carranza's Clinical Periodontology. 11th ed. Philadelphia: Elsevier Health Sciences; 2011.
- Socransky SS, Haffajee AD. The bacterial aetiology of destructive periodontal disease: current concepts. J Periodontol. 1992;63:322-31.
   Kinney JS, Ramseier CA, Giannobile WV. Oral fluid-based biomarkers of alveolar bone loss in periodontitis. An New York Acad Sci. 2007;1098:230.
- 4. Taba M, Kinney J, Kim AS, Giannobile WV. Diagnostic biomarkers for oral and periodontal diseases. Dental Clinics. 2005;49(3):551-71.
- 5. Chapple I, Garner I, Saxby M, Moscrop H, Matthews J. Prediction and diagnosis of attachment loss by enhanced chemiluminescent assay of crevicular fluid alkaline phosphatase levels. J Clin Periodontol. 1999;26(3):190-8.
- 6. Zhang L, Henson BS, Camargo PM, Wong DT. The clinical value of salivary biomarkers for periodontal disease. Periodontol 2000. 2009;51(1):25-37.
- 7. Berglundh T, Donati M. Aspects of adaptive host response in periodontitis. J Clin Periodontol. 2005;32:87-107.
- 8. Dabra S, Singh P. Evaluating the levels of salivary alkaline and acid phosphatase activities as biochemical markers for periodontal disease: A case series. J Dent Res. 2012;9(1):41-5.
- 9. Navazesh M, Christensen C. A comparison of whole mouth resting and stimulated salivary measurement procedures. J Dent Res. 1982;61(10):1158-62.
- 10. Breslow R, Katz I. Relative reactivities of p-nitrophenyl phosphate and phosphorothioate toward alkaline phosphatase and in aqueous hydrolysis. J Am Chem Soc. 1968;90(26):7376-7.
- 11. Ishikawa I, Cimasoni G. Alkaline phosphatase in human gingival fluid and its relation to periodontitis. Arch Oral Biol. 1970;15(12):1401-4.
- 12. Weiss MJ, Ray K, Henthorn PS, Lamb B, Kadesch T, Harris H. Structure of the human liver/bone/kidney alkaline phosphatase gene. J Biol Chem. 1988;263(24):12002-10.
- 13. Palcanis KG, Lariava IK, Wells BR, Suggs KA, Landis JR, Chadwick DE, et al. Elastase as an indicator of periodontal disease progression. J Periodontol. 1992;63(4):237-42.
- 14. Sophia K, Suresh S, Sudhakar U, Jayakumar P, Mathew D. Comparative Analysis of Salivary Alkaline Phosphatase in Post menopausal Women with and without Periodontitis. J Clin Diagn Res. 2017;11(1):Zc122-4.
- 15. Adesakin AM, Elizabeth DB, OI O, Modupe AO. Evaluating Salivary Alkaline Phosphatase Levels as a Biochemical Marker of Periodontal Disease in Periodontal Patients in a Tertiary Hospital in Nigeria. J Periodontol Implant Dent. 2016;8(1):19-23.
- 16. Patel RM, Varma S, Suragimath G, Zope S. Estimation and Comparison of Salivary Calcium, Phosphorous, Alkaline Phosphatase and pH Levels in Periodontal Health and Disease: A Cross-sectional Biochemical Study. J Clin and Diagn Res. 2016;10(7):ZC58-61.
- 17. AlMoharib HS, AlMubarak A, AlRowis R, Geevarghese A, Preethanath RS, Anil S. Oral Fluid Based Biomarkers in Periodontal Disease: Part 1. Saliva. J Int Oral Health. 2014;6(4):95-103.
- Todorovic T, Dozic I, Pavlica D, Markovic D, Brajovic G, Ivanovic M, et al. [Use of saliva as a diagnostic fluid in dentistry]. J Serbian Med Soc. 2005;133(7-8):372-8.
- 19. Patil PB, Patil BR. Saliva: A diagnostic biomarker of periodontal diseases. J Indian Soc Periodontol. 2011;15(4):310-7.
- 20. Kalburgi V, Jenifer HD, Warad S, Bhola S, Chaudhari HL. Comparison of salivary alkaline phosphatase levels among diabetics and nondiabetics with chronic periodontitis. J Oral Health Res. 2010;1(4):18.
- 21. Merza KS, Alaaraji SB, Abdullah BH. Comparative study on lactate dehydrogenase, alkaline phosphatase and immunoglobulins in serum and saliva of acute leukemia and oral squamous cell carcinoma patients. Iraqi J Sci. 2010;51(2):262-70.
- Agnihotram G, Singh TRM, Pamidimarri G, Jacob L, Rani S, Sravanthi. Study of clinical parameters in chronic periodontitis. 2010;1(3):1202-8.
   Desai S, Shinde H, Mudda J, Patil V. Levels of alkaline phosphatase (ALP) in saliva of patients with chronic periodontitis; A clinical and
- biochemical study. Internet J Dent Sci. 2008;8(1):1937-8238.
- 24. Todorovic T, Dozic I, Vicente-Barrero M, Ljuskovic B, Pejovic J, Marjanovic M, et al. Salivary enzymes and periodontal disease. Med Oral, Patol Oral Cir Bucal. 2006;11(2):E115-9.
- 25. Gibert P, Tramini P, Sieso V, Piva M. Alkaline phosphatase isozyme activity in serum from patients with chronic periodontitis. J Periodontal Res. 2003;38(4):362-5.
- 26. Singh N, Chandel S, Singh H, Agrawal A, Savitha A. Effect of scaling and root planing on the activity of ALP in GCF and serum of patients with gingivitis, chronic and aggressive periodontitis: A comparative study. J Oral Biol and Craniofacial Res. 2017;7(2):123-6.
- Jeyasree RM, Theyagarajan R, Sekhar V, Navakumar M, Mani E, Santhamurthy C. Evaluation of serum and salivary alkaline phosphatase levels in chronic periodontitis patients before and after nonsurgical periodontal therapy. J Indian Soc Periodontol. 2018;22(6):487-91.
- 28. Kaufman E, Lamster IB. Analysis of saliva for periodontal diagnosis: a review. J Clin Periodontol. 2000;27(7):453-65
- 29. Pradhan S, Bhat M. Assessment of periodontal status of rural Nepalese population using the community periodontal index. J Nepal Dent Assoc. 2009;10(2):97-104.