# Original Article EVALUATION OF SALIVARY ENZYMES IN POST MENOPAUSAL WOMEN WITH AND WITHOUT PERIODONTITIS

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#### Abstract:

Aim : To compare the levels of Lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) in post-menopausal women with and without periodontitis

Methodology : A cross-sectional pilot study was conducted. A total of 50 postmenopausal women were recruited and categorized into two groups based on their periodontal status. Their salivary samples were collected and subjected to Lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) estimation in the laboratory

Results : The activity of LDH and ALP were significantly higher in the post-menopausal women with periodontitis than those without periodontitis.

Conclusion : The present study demonstrated post-menopausal women may have exaggerated inflammatory response to dental plaque.

Keywords : Lactate dehydrogenase (LDH), Alkaline phosphatase (ALP), post-menopause, periodontitis, saliva

#### Introduction:

Periodontitis is an infectious disease of microbial origin, the progression of which is, however, mediated through host related factors. These host factors, may be, further modulated by various systemic factors, the female sex hormones, being one of them<sup>1,2</sup>. The effects of circulating estradiol (E2) on the periodontal tissues homeostasis have previously been well documented<sup>3</sup>.

Menopause is permanent cessation of menstrual cycle after 12 consecutive months of amenorrhea and is also characterized with decreasing levels of estradiol (E2) as the



principal circulating estrogen<sup>4</sup>. Estrogen deficiency enhances the rate of breakdown of the connective tissue components of the gingiva by stimulating synthesis of m a t r i x metalloproteinases, nitrous oxide, and several cytokines implicated in bone resorption, especially in response to bacterial infection. Thus, it has been proposed that alteration in the levels of sex hormones may exacerbate periodontal tissue breakdown by altering host response<sup>5,6</sup>.

Lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) are intracellular cytoplasmic enzymes that previously been used as markers for early diagnosis of periodontal disease. The extracellular presence of LDH is thought to be related to cell necrosis and tissue breakdown while alkaline phosphatase is indicative of bone turnover and an increase in its levels indicates bone resorption<sup>7-9</sup>.

In recent years, saliva has been recognised as a bio-fluid that can be used to evaluate for markers of various disease processes due its ease of availability and non-invasive means of collection. The composition of this oral fluid is influenced by constituents derived not only from the major and minor salivary glands, but also from gingival crevicular fluid (GCF), serum, bacteria, desquamated epithelial cells





#### etc<sup>9</sup>.

The purpose of the present study was to investigate salivary levels of lactate dehydrogenase and alkaline phosphatase in postmenopausal women with and without periodontitis.

#### Methodology:

Ethical clearance was obtained from the institution Ethics committee. This cross-sectional study was conducted in the department of Periodontics, A. B. Shetty Memorial Institute of Dental Sciences, Mangalore and Department of Obstetrics and Gynecology, K. S. Hegde Hospital, Nitte University, Deralakatte, Mangalore. The total of 72 postmenopausal women were screened while 50 patients among them were selected for the study based on following selection criteria.

Inclusion criteria: a) Individuals age between 45-55 years b) Naturally attained menopause c) 20-28 natural teeth to present

Exclusion criteria: a) Patient on any medications b) Patient with any other systemic diseases.

A detailed medical and dental history was recorded. An informed written consent was obtained from selected patients who were categorized into two groups, Group-A postmenopausal women with healthy periodontium and Group-B postmenopausal women with periodontitis based on periodontal health status.

## Periodontal examination:

The periodontal status was assessed by measuring severity of gingival inflammation by using Löe and Silness Gingival index , and periodontal pocket was measured at mesio-facial, mid facial, disto-facial, disto-lingual,mid lingual and mesio-lingual areas in millimeters using Michigan 'O' probe with William's graduation.

The loss of attachment (LOA) was calculated based on the position of marginal gingiva to cement-enamel junction.

#### Collection of saliva:

Unstimulated saliva was collected in a sterile container and

stored at -20°C, later these samples were brought to room temperature and then subjected for estimation of LDH and ALP levels.

### Enzyme estimation:

LDH and ALP activity were measured at 37° C in laboratory by using Aspen Laboratory kit and Star 21+ semi auto analyzer. Pyruvate and 4-nitrophenylphosphate were used as substrate to determine LDH and ALP levels.

#### Statistical analysis:

The obtained values were tabulated and analyzed by using Independent Student't' test. Correlation among groups interpreted using Pearson correlation coefficient and the p<0.05 was considered to be significant.

### Results:

Table 1 Independent Student's 't' value obtained between mean values of LDH and ALP levels between two groups was found to be very significant[p<0.01] .Table 2 Correlation coefficient at 0.05 level [2 tailed test] between LDH and loss of attachment [LOA] was significant and also there was significant differences found between gingival index and pocket depth.

#### Discussion:

Estrogen deficiency is reported to increase a woman's risk for developing various acute and chronic diseases. The possible association between menopause and periodontal disease progression has been discussed in many studies. However, the results have been inconclusive, with some studies suggesting that menopause related systemic conditions may induce increased alveolar bone resorption and periodontal attachment loss, while others have suggested no such relationship. The present study sought to evaluate the influence of the post-menopausal state on the periodontal tissue by evaluated the salivary levels of LDH and ALP in post-menopausal women with and without periodontal disease.

Most of previous literature that focused on LDH as a diagnosis marker for periodontal disease has been carried out in samples of gingival crevicular fluid (GCF). Studies that have evaluated periodontal disease activity and LDH



Table 1 : Mean and standard deviation of age, LDH and ALP for both the study groups.

Group A N=25			Group B N=25		Remarks
	Mean	Std	Mean	Std	
		Deviation		Deviation	
Age	52.64	8.103	50.48	7.7	Not
					significant
ALP level	32.04	5.35	46.79	7.93	Highly
(IU/L)					significant*
LDH level	229.24	81.21	278.34	89.5	Highly
(IU/L)					significant*

\*p value < 0.01

Table 2: Correlation of LDH and ALP levels in saliva with gingival index, loss of attachment and pocket depth for group B patients.

		Gingival	Loss of	Pocket			
		index	Attachment	depth			
LDH Level	Pearson						
	Correlation	0.457*	0.593*	0.348			
ALP Level	Pearson						
	Correlation	0.105	0.277	0.228			
*P.voluo < 0.01							

\*P value<0.01

Graph 1 : LDH levels in Group A and Group B







levels has shown a weak correlation between the two. It has been proposed that LDH is only indicative of the inflammatory process occurring in the periodontal tissues Kinney et al<sup>9</sup>.

The existing literature about the levels of activity of LDH in saliva is scarce. These studies have demonstrated significantly higher levels of salivary LDH activity in individuals with periodontal disease than those obtained in patients with a healthy periodontium. De La Pen<sup>~</sup> a et al<sup>10</sup>. Previous evidence suggests that the main source of LDH in whole saliva is the oral epithelium and not the salivary glands.

Alkaline Phosphatase (ALP) found in whole saliva originate from salivary secretions, the GCF and disposed bacterial cells from dental biofilms and mucosal surfaces. Earlier studies have demonstrated a significant positive correlation between salivary ALP and periodontal disease severity and inflammation.

In postmenopausal women , studies have shown that plasma ALP and LDH activities were significantly elevated when compared to that of the pre-menopausal women and this was thought be due to hormonal influence<sup>11,12</sup>.

In the present study, salivary levels of LDH and ALP showed significant increase in postmenopausal women with periodontitis as compared with postmenopausal women without periodontitis. There was also a significant correlation between LDH and ALP with clinical parameters.

Though the levels of LDH and ALP showed a significant difference between the two groups it was within normal values suggesting that the systemic status of the patient may have a limited role in the etiopathogenensis of periodontal disease.

As a predictive indicator for future periodontal breakdown, both LDH and ALP has not been supported by research findings and therefore may best serve as a marker in periodontal inflammatory state and thus, facilitate treatment planning and monitoring. Kinney et al 2007<sup>9</sup>.

Within the limits of the present study, our present data, suggests that menopausal state may only have a limited influence on the severity of periodontal disease, but may have influenced an exaggerated inflammatory response to plaque<sup>13</sup>.



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