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# QUANTITATIVE EVALUATION OF SALIVARY CALCIUM, PHOSPHOROUS, PROTEIN AND pH IN HEALTH AND DISEASED PERIODONTIUM

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**ABSTRACT**

*Saliva contains a variety of host defense factors. It influences calculus formation and periodontal disease. This study was conducted to estimate the role of salivary factors such as calcium, phosphorus, protein and pH in progression of periodontal disease. Depending upon severity of periodontal diseases subjects were divided into IV groups of 25 subjects in each group. Results obtained were subjected to statistical analysis by application of 't' test. The findings suggested that salivary calcium values in all groups were statistically significant. The salivary pH in group IV was found to be statistically significant in the present study. A positive correlation between the level of calcium and the severity of disease was observed. It is therefore, suggested that monitoring for change in salivary composition might be useful tool to establish periodontal health status.*

***KEY WORDS :*** *saliva, Periodontal Health, Calcium, Phosphorus, Protein, pH*

## INTRODUCTION

Oral cavity is a moist environment with a film of fluid called saliva. The saliva is a complex fluid containing a variety of mucosal host defense factors from the different salivary glands and the crevicular fluid[1]. Compelling reasons exist to use saliva as a diagnostic fluid. It meets the demands for inexpensive, noninvasive and easy-to-use diagnostic methods[2]. As a clinical tool, saliva has many advantages over serum, including ease of collection, storing and shipping, and it can be obtained at low cost in sufficient quantities for analysis[3]. For patients, the noninvasive collection techniques dramatically reduce anxiety and discomfort and simplify procurement of repeated samples for monitoring over time. Saliva also is easier to handle for diagnostic procedures because it does not clot, thus lessening the manipulations required[4]. Saliva exerts a major influence on plaque initiation, maturation, and metabolism. Salivary flow and composition influences calculus formation, periodontal disease. The inorganic components of plaque are calcium, phosphorous and other minerals. As the mineral content increases, the plaque mass become calcified to form calculus[5]. The purpose of this study was to estimate the salivary concentration of calcium, phosphorus, protein and pH of human saliva in progression of periodontal disease.

### Materials and methods

A Clinico-biochemical study was designed and conducted at Department of Periodontics, College of Dental Surgery, K. M. C., Manipal. The study included 100 subjects, both male and female of age ranging between 15 -30 years. They had normal occlusion (Cl. I) and no habits like smoking, pan and tobacco chewing, thumb sucking and mouth breathing and were free from systemic disease.
Total sample were divided into IV groups of 25 subjects in each group.

| | |
|---|---|
| **Group I** | 25 subjects in whom the gingiva and occlusion were normal. Measures were taken for these subjects to maintain very good oral hygiene |
| **Group II** | 25 subjects in whom occlusion was normal but with Chronic marginal gingivitis. |
| **Group III** | 25 subjects in whom occlusion was normal but with Chronic marginal gingivitis and localized chronic periodontitis. |
| **Group IV** | 25 subjects with chronic generalized periodontitis |

The health status of gingival and periodontium was assessed by using mouth mirror and Williams Periodontal probe in natural light. Clinical examination to determine the Periodontal status

includes the assessment of Plaque index (Silness and Loe 1964)[6] and Periodontal index (Russell's 1956) [7].

About 5-10 ml of resting saliva was collected and biochemical analysis was carried out for calcium by precipitation (Trinder, 1960)[8], centrifuge tubes 12 to 15 ml capacity are used. Inorganic phosphorus in saliva (Fiske and Subbarao, 1925)[9] and proteins by (Biuret method, 1953)[10] and estimation of salivary pH using single electrode pH meter (digital).

**STASTICAL ANALYSIS:** Results were tabulated ( Table 1 and Table 2.) and were analyzed by 't' test of significance. Group II, III & IV were compared to the control i. e. group I. The level of significance "p" value at 95% confidence interval was calibrated as: Nonsignificant (NS): $p > 0.05$, Significant (S): $0.01 < p < 0.05$ and highly significant (HS): $p < 0.001$.

**Results:**

**Group I**: The mean value for the total salivary calcium level was found to be 4.3 mg/ 100 ml and mean salivary phosphorous level was 18.49 mg/ 100ml of saliva. The mean protein level was 263.05 mg/ 100ml and the pH of saliva was 7.15.

**Group II**: The mean calcium level for this group was 4.6 mg/ 100 ml. the salivary phosphorous level was estimated to be 17.74 mg/ 100 ml. the protein level of saliva was 281.32 mg/ 100 ml and the pH level of saliva was 7.15.

**Group III**: The salivary calcium level was found to be 4.1 mg/ 100 ml and phosphorous content of saliva was 19.26 mg/ 100 ml. mean protein value 367.12mg/100 ml and mean pH value as 7.27.

Group IV: Mean salivary calcium level was 4.02 mg/ 100 ml, phosphorous level was estimated to be 19.4 mg/ 100 ml. the mean value of salivary protein was found to be 406.8 mg / 100 ml and the mean value of saliva in these subjects was 7.5.

Results were analyzed by 't' test of significance. Group II, III & IV were compared to the control i. e. group I.

The findings showed that salivary calcium values in all the groups were statistically significant. Salivary phosphorus and salivary protein values were not statistically significant in all the groups. The pH value of group IV was found to be statistically significant in the present study.

## Discussion

Mouth is a biological system. Saliva and crevicular fluid play a decisive role in the prevention of periodontal disease and indeed paradoxically in the induction of periodontal pathology. Thus both of these factors have been the object of much study. The saliva that basically forms the environment of the oral cavity is the resting or pooled saliva. For this reason unstimulated whole saliva was collected from the subjects.

Some relationship between the inorganic constituent like **Ca** and **P** in saliva and periodontal health is suspected, the exact mechanism is still unclear. Ashley and Wilson, 1978 also postulated that a possible relationship between Ca and P in saliva periodontal disease exists, but this must be studied in the mixed saliva where of course a number of inter reactions are possible. [11]

While caries tends to occur at an early age and tends to regress in later years, the reverse is true of periodontal disease. This relationship has been the object of study by Waerhaug et al(1952). Periodontal pathology being a cumulative process, tissue changes would be minimal, in younger age groups. However, participants below 15 was excluded due to possible effect of physiological metabolic growth changes on Ca and $Po_4$ levels of saliva[12].

Malocclusion, mouth breathing and local pathologic factors conductive to induction of periodontal disease were also eliminated. Into this category also fell patients with systemic disease which could influence saliva and the tissue resistance.

Chaterjee and Kleinberg opine that the level of calcium and P in saliva could be of clinical value as regards to periodontal disease[13].

Calcium levels in saliva appeared to decrease correspondingly to an increase in the degree of gingival and periodontal inflammation. The level obtained in group I of this study is consistent with those of Becks[14] and Weinwright, and Kamat N.V[15]. However, this is not in agreement with the findings of Sewon, calcium level was significantly higher in periodontitis affected subjects[16]. It has been reported increased calcium level associated with increased periodontal inflammation[17]. As reported by Kamat the gingival calcium level decreases as the disease progresses from gingivitis to periodontitis. Also the variation could be due to factor like age, diet, absorption and excretion of Ca variation as documented by Ramakrishna[18]. Lower levels of calcium in adolescence and young adults, than in older age group has been described by

**Table 1: Showing the mean and standard deviation of normal group (control- normal gingiva)**

| variable | Group I (Normal) | Group II (Chronic marginal gingivitis) | Group III (Chronic marginal gingivitis and localized periodontitis) | Group IV (Chronic marginal gingivitis and generalized periodontitis) |
|---|---|---|---|---|
| | All values in Mean± SD | | | |
| **Ca** | 4.3 ±0.26 | 4.6±0.31 | 4.1±0.23 | 4.02± 0.19 |
| **P** | 18.49±2.56 | 17.79±0.31 | 19.26±1.53 | 19.4± 0.36 |
| **Protein** | 263.05±183.25 | 281.32±187.35 | 367.12±256.02 | 406.80±308.12 |
| **pH** | 7.15± 0.32 | 7.15±0.22 | 7.27±0.26 | 7.5±0.25 |

**Table 2: Showing statistically "t"-test value and the probability level**.

| Source | Ca | P | Protein | pH |
|---|---|---|---|---|
| **I vs II** | 3.66<br>P < .001* | 0.81<br>P > .05 | 0.34<br>P > .05 | 0<br>P > .05 |
| **I vs III** | 2.86<br>P < .01* | 1.27<br>P > .05 | 1.62<br>P > .05 | 1.15<br>P < .05 |
| **I vs IV** | 4.30<br>P < .001* | 1.72<br>P > .05 | 1.95<br>P > .05 | 4.22<br>P < .001* |

**** Statistically significant***

Chatterjee and Kleinberg who contributed rapid growth as in adolescence and hence there is low salivary output[13].

Salivary P level increases with the increase in severity of periodontal disease. But P level variation was found to be of no statistically significant. The findings of P level in this study are in agreement with those of Ericson[19] De Jorge and Carvallo, Kamath[20].

Role of nutrition in periodontal disease is well documented;protein is an important factor in maintaining the integrity of the supporting tissues of teeth. Experimental research on animals has proved that the protein deprivation leads to degeneration of connective tissue of the periodontal ligament. In the present study it was found that the salivary protein level increases with the increase in the disease process of periodontium. Studies have shown elevation of total Protein in saliva in Periodontitis, elevated protein can be used as a marker of inflammation in the periodontium[21].

Salivary pH values shows no statistical significance in our study. The present study corroborates with the findings of Green and Loe et al (1965)[22]. As reported by Mukerji the rise in pH causes precipitation of calcium phosphate salts in plaque resulting in calculus formation.

The existence of direct relationship between Ca and P level of plaque and saliva is possible. It is unlikely that Ca and phosphate ions diffuse into plaque from the saliva (Jenkins, 1975, Ashley and Wilson)[23]. Probably these ions are incorporated during the formation of plaque together with salivary protein[24]. The precipitation of Ca and P may be influenced by the change in pH during the formation of plaque (Chatterjee and Kleinberg, 1973).
The exact nature of the inorganic constituents of saliva in the induction and maintenance of periodontal disease is far from being understood. Here definitive study investigating the mechanism of incorporation of Ca and P ions in the plaque from saliva, along with a greater understanding of biochemical mechanisms in gingival inflammation is essential.

**CONCLUSION:**

The study was undertaken to examine the relationship of salivary calcium, phosphorous, protein and pH changes with regard to periodontal disease. A positive correlation between the level of calcium and the severity of disease was observed. It

is therefore, suggested that monitoring for change in salivary composition might be useful tool to establish periodontal health status.

A study more closer to the seat of periodontal disease such as within plaque, or on the calcification of plaque, is necessary to composition and to establish the nature of their exact role in the induction and the progression of periodontal disease

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