Prashaanthi.N et al /J. Pharm. Sci. & Res. Vol. 8(7), 2016, 623-626

ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in

A Study on Association of Salivary Calcium and Phosphate in Oral Health

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Abstract:

Aim and objective:
The aim of the study is to associate salivary calcium and phosphate in oral health.

Background:
Salivary glandular secretion which constantly bathes the teeth and oral mucosa. Presence of saliva is vital for healthy oral tissue. Positive correlation has been shown in salivary calcium and phosphate and oral health.

Reason:
To associate role of salivary calcium and phosphate in patients with intact teeth.

Result:
The association of salivary calcium and phosphate in periodontitis was assessed.

Keywords: salivary calcium, inorganic phosphate, periodontitis, remineralisation.

INTRODUCTION:

Saliva is the glandular secretion , which constantly bathes the teeth and oral mucosa. The presence of saliva is vital for the maintenance of healthy oral mucosa. (1). Saliva has protective properties and contains a variety of antimicrobial components and growth factors. Saliva acts as a lubricant and aids in the digestion of food.(8) The presence of saliva keeps the oral mucosa and the teeth in a healthy condition. Severe reduction of saliva results in a deterioration in oral health and also has an impact on the quality of life for the sufferer. (9) . An important component of saliva are its proteins, such as the glycoproteins which adsorb onto tooth structure to form the protective pellicle layer, and the phosphoproteins which regulate calcium saturation of the saliva.(10)

Saliva as a host-associated factor plays essential role in preserving the integrity of oral structure that can be summarised into four aspects: diluting and eliminating sugars and substances, buffering capacity, balancing between demineralisation and remineralisation and antimicrobial activity.(6) Calcium is an important ion present in the body. The balance between demineralisation ,remineralising process depends on concentration of the salivary parameters-calcium, phosphate and pH(2). The phosphate ions and some electrolytes play a important role in buffer mechanism. Saliva plays a part in reducing the acids in plaque , contains specific buffer mechanism such as bi carbonates, phosphates and some proteins system with not only have as a buffer effect but eliminates certain bacterial components that requires very low pH to survive. The phosphate buffer plays an essential role when salivary flow is low. (3).According to the mineral precipitation theory for calculus formation, calcification will occur when pH, calcium, and phosphate concentrations are high enough to allow the precipitation of a calcium phosphate salt.(4)

The dissolution of hard tissues of tooth in the state of calcium and phosphorus occurs in the oral cavity in the presence of saliva. The ionic concentration of calcium and phosphate in saliva helps maintain an equilibrium between dissolution and remineralisation of enamel (5).

Saliva plays a fundamental role in maintaining the physical-chemical integrity of tooth enamel by modulating remineralization and demineralization . The main factors controlling the stability of enamel hydroxyapatite are the active concentrations free of calcium, phosphate, and fluoride in solution and the salivary pH.(7) The abnormal secretions present in cystic fibrosis (CF) caused clinicians to explore the usefulness of saliva for the diagnosis of the disease. Most studies agree that saliva of CF patients contains increased calcium levels (Mandel et al., 1967; Blomfield et al., 1976; Mangos and Donnelly, 1981). Elevated levels of calcium and proteins in submandibular saliva from CF patients were found, and resulted in a calcium-protein aggregation which caused turbidity of saliva (Boat et al., 1974). The elevated calcium and phosphate levels in the saliva of chil- dren diagnosed with CF may explain the fact that these children demonstrate a higher occurrence of calculus as compared with healthy controls (Wotman et al., 1973). (8) We have shown positive correlations between high salivary calcium content and periodontitis (Sewon et al, 1990b), and between high ,salivary calcium level and the number of intact teeth (Se-won & Makela 1990), Though contrary findings also occur (,\tnanio et al, 1980, Skier & Mandel 1980, Kinane et al, 1991), we have earlier also found that subjects with periodontitis have more intact teeth and more intact molars than subjects who ate free of the disease (Sewon et ai, 19SSl, Therefore, our present concept IS that periodontitis affected subjects have a higher intraoral mineralization potential.(9) Changes in salivary composition and flow

rates may compromise the integrity of the soft and hard tissues in the oral cavity, because saliva functions include food and bacteria clearance, mastication and digestion, lubrication, antimicrobial defence, and buffering effect [6,7]. Saliva is composed of water , organic and inorganic molecules, but a large intra- and inter-subject variability in composition is reported. (10) salivary calcium and phosphate concentrations increase with age showing peak values around menopause. Therefore we suggest that menopause is reflected in saliva as elevated levels of calcium and phosphate (11)

Saliva also plays an important role in maintaining the integrity of dental tissues due to the presence of calcium, phosphorous and other inorganic ions as this environment is known to facilitate remineralization of incipient lesions or demineralized zones of enamel. Thus calcium and phosphorous in saliva forms a natural defence mechanism against dissolution of teeth.(12)

Positive correlations have been shown between high salivary calcium content and periodontitis and between high salivary calcium content and number of intact teeth. It was also found that subjects with periodontitis have more intact teeth and more intact molars than subjects who are free of the disease.Therefore, the present concept is that periodontitis affected subjects have higher intraoral mineralization potential.[1]

This study was designed to estimate and compare inorganic salivary calcium, phosphate of periodontally healthy subjects and patients with periodontitis.

MATERIALS AND METHOD:

A total of 30 subjects in the age group of 40-50 years visiting Saveetha dental college were assessed. Out of 30 subjects, 15 subjects affected with periodontitis and 15 subjects as a control group were included in this study.

Collection of the Salivary Sample :

Patients were advised that a very small amount of saliva will ooze into their mouth in un-stimulated state and that the objective of the test was to measure the rate of flow of this secretion. Saliva was collected at least 1 1/2 hr after eating. Un- stimulated whole saliva was collected by making the patient to sit in upright position at rest, bow their head and try not to move during the test. Immediately before the test begun, they were instructed to swallow any residual saliva that may be in their mouth. The saliva was allowed to accumulate for 2 min and then expectorated into the collecting vessel. If insufficient saliva was obtained then test may be conducted for a longer period of time often for 5 min[1]. The procedure was done after getting consent from the patients.

Estimation of Inorganic Salivary Calcium By Calorimetric Method:

Calcium in saliva was estimated as described by Arsenazo III method using kit supplied by Agappe. Calcium ions (Ca2+) reacts with Arsenazo III (2,2'-[1,8-Dihydroxy-3,6-disulphonaphthylene-2,7-bisazo]- bisbenzenear-sonic acid) and forms an intense purple coloured complex.

Magnesium does not significantly interfere in calcium determination using Arsenazo III. In this method the absorbance of the Ca-Arsenazo III complex is measured bichromatically at 660/700 nm. The resulting increase in absorbance of the reaction mixture is directly proportional to the calcium concentration in the sample.

Ca2+ + Arsenazo III = Ca-Arsenazo III complex (purple)

Estimation of Inorganic Salivary Phosphate By Calorimetric Method:

Salivary phosphate levels were assessed by Ammonium molybdate end method.

Inorganic phosphorus reacts with ammonium molybdate in an acidic medium to form a phosphomolybdate complex which absorbs light at 340nm. The absorbance at this wavelength is directly proportional to the amount of inorganic phosphorus present in the sample.

Ammonium molybdate + Inorganic Phosphorus = Phosphomolybdate Complex.

Statistics:

The obtained results were statistically assessed.

T-test:

Group Statistics

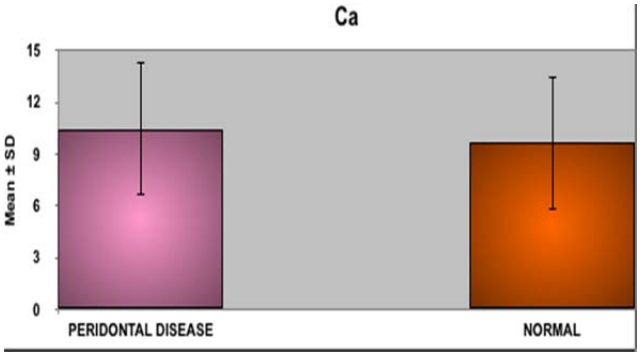
	Group	N	Mean	Std. Deviation	Std. Error Mean
Ca	Normal	15	9.660	.7278	.1879
	Periodontitis	15	10.462	3.8767	1.0010
P	Normal	15	3.560	.4453	.1150
	Periodontitis	15	4.260	.7317	.1889

Independent Samples Test

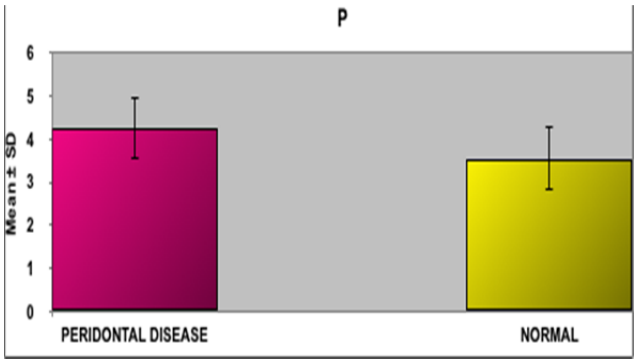
		Levene's Test for Equality of Variances		t-test for Equality of Means						
									95% Confidence Interval of the Difference	
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	Lower	Upper
Ca	Equal variances assumed	7.736	.010	-.787	28	.438	-.8020	1.0185	-2.8882	1.2842
	Equal variances not assumed			-.787	14.986	.443	-.8020	1.0185	-2.9730	1.3690
P	Equal variances assumed	3.329	.079	-3.165	28	.004	-.7000	.2212	-1.1530	-.2470
	Equal variances not assumed			-3.165	23.119	.004	-.7000	.2212	-1.1574	-.2426

RESULTS AND DISCUSSION:

Salivary Calcium	Mean	SD
Periodontal disease	10.46	3.87
Normal	9.66	0.72



Salivary Phosphorous	Mean	SD
Periodontal disease	4.26	0.73
Normal	3.56	0.44



In this study , 15 subjects affected with periodontitis and 15 subjects as control group were compared. In these two groups , unstimulated saliva was collected and analysed for inorganic calcium and phosphate.

The comparison of mean of salivary inorganic calcium and phosphate of periodontitis [group statistics] revealed that the mean of salivary inorganic calcium as 9.660+_ 0.7278 for control group and 10.462+_ 3.8767 for periodontitis group. The observed mean between the two groups was statistically significant. The mean of salivary inorganic phosphate is 3.560+_0.4453 for control group and 4.260+_0.7317 for periodontitis group. The observed mean between the two groups was statistically significant.

Since 1990, Sewσn et al. with their series of studies have shown that oral mineralization potential of saliva plays an important role in periodontal health and disease. Mineralization favoring factors are known to maintain the integrity of enamel surfaces and intraoral mineralization capacity has been a matter of scientific interest for decades. [1]

In our study , salivary calcium levels and phosphate levels of patients with periodontitis were highly significant than healthy patients. They are in accordance with the study of Sewon et al. There is positive co-relation between high salivary calcium levels with periodontitis.

Therefore, the present concept is that periodontitis affected subjects have higher intraoral mineralization potential. Sewón et al. 1990, have also demonstrated that several mineralization favoring factors are prominent in periodontitis-affected subjects when compared with those who are periodontitis-free.[1]

The result of this study shows that the patients with high salivary calcium and phosphate are more prone to get periodontitis and on the other hand they are resistant to caries. Healthy patients with normal salivary calcium and phosphate levels or decrease in salivary calcium and phosphate levels are prone to get dental caries as their plaque has the affinity to demineralise the enamel.

CONCLUSION :

This study highlights the potential for a relationship between levels of inorganic calcium and phosphate in diseases like periodontitis which affects humans and needs more investigation. Conclusions that can be drawn from this study are that the individuals who have increased salivary calcium and phosphate are at a higher risk for developing periodontitis. In future , the study of the salivary biomarkers can be done and co-related with periodontitis.

ABBREVIATIONS:

CF- cystic fibrosis

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