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Original Article

RELATION BETWEEN SALIVARY AND SERUM VITAMIN C LEVELS AND DENTAL CARIES EXPERIENCE IN ADULTS - A BIOCHEMICAL STUDY

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Abstract :
The aim of this study was to estimate the vitamin C levels in saliva and serum of caries free and caries active adults and to correlate the vitamin C level with DMFT index (D=decayed, m=missing, f=filled, t=teeth) index. The present study included eighty healthy adults who were divided into four groups; Control, Group- I, Group II and Group- III with a DMFT index 0, <3, <10 and > 10 respectively. Saliva and serum samples were collected from all the four groups. The vitamin C of saliva and serum was estimated by dinitro phenyl hydrazine (DNPH) method. One-way ANOVA was used to compare the vitamin C levels of saliva and serum. Only differences with 'p'value <0.05 were considered statistically significant. Saliva and serum vitamin C level decreases with increase in caries activity and is statistically significant suggesting the powerful antioxidant property of vitamin C.

Keywords: Vitamin C, Saliva, Serum, Dental caries

Introduction :

Diagnostic procedures most commonly used in laboratory involve the analyses of the cellular and chemical constituents of blood. Saliva offers some distinctive advantages when used for diagnosis of disease. Human saliva is a fluid with many biological functions essential for the maintenance of oral health. In past scientists were more interested in studying the biological functions of saliva in the mouth than in trying to assess its possible role as an indicator of systemic or oral disease[1]. The potential of saliva for diagnosis purpose has attracted more attentions in recent years[2]. Saliva is armed with various defense mechanisms, such as the immunological and enzymatic defense systems[3]. According to recent data it mirrors general health condition thus reflecting various systemic changes in the body[3,4]. Locally produced proteins along with some other molecules from the systemic circulation are found in saliva. On the other hand, various amounts of blood, serum derived molecules, gingival crevicular fluid, electrolytes, epithelial cells, microorganisms and some minor substances are also found in whole saliva[5,6,7].

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Dental caries is a complex multifunctional disease, as multiple factors influence the initiation and progression of the disease. Among the factors that have been related to greater cariogenic activity are inadequate dental hygiene and care. Changes in salivary components are in connection with caries formation and it may be used for recognizing risk in patients and to maintain prevention[8].

Antioxidants are present in all body fluids and tissues and protect against endogenously-formed free radicals[9]. Salivary antioxidants system consist of enzymatic (superoxide dismutase and peroxidase) and non enzymatic (uric acid) components[3]. Vitamin C is able to scavenge free radical of both reactive oxygen group (super oxide and

hydro peroxyl) and reactive nitrogen group (nitrogen dioxide and peroxynitrite)[10].Vitamin C may be capable of regenerating other antioxidants like vitamin E therefore prevents oxidative damages[11].

The aim of the present study was to determine the correlation between salivary and serum vitamin C concentration of adults in correspondence to DMFT index.

Materials and Methods :

This study was approved by the Committee for Ethics in Research, Dental College, Nitte University, Karnataka, India. 12,500 healthy adult patients coming to the OPD of Department of Conservative Dentistry and Endodontics, A.B.Shetty Memorial Institute of Dental Sciences under the age group of 25-50 years between December 2010- June 2011 were randomly selected. The patients fulfilling the inclusion criteria were free from systemic or local disease which affects salivary secretions and their caries status was assessed according to World Health Organization "W.H.O. recommendations 1997" to calculate dental caries index[12]. Patients with periodontal disease, hypertension, diabetes, radiotherapy, chemotherapy, systemic disease of the vital organs and history of long term medications were excluded from the study.

Out of these, 80 healthy adults were selected for the study and divided into groups as caries free consisting of 20 individuals and caries active group consisting of 60 individuals. The caries active group was further divided into three subgroups based on the DMFT score as follows, Group I (DMFT<3), Group II (DMFT<10) and Group III (DMFT>10), each group consisting of 20 individuals.

A detailed case history of the patient was recorded, informed consent read and duly signed by each patient.

The smooth and occlusal surfaces of teeth were cleaned with soft bristle brush, dried and examined. DMFT score calculated.

Collection of saliva:

Collection of saliva was done in the noon time before food to maintain the uniformity of the composition. Unstimulated saliva was collected from a patient who is not involved in any masticatory function in the last two hours prior to saliva collection and is seated in an ordinary chair and not on any dental / operatory chair to avoid anxiety.

5ml of saliva was collected from the patient, centrifuged and the supernatant obtained was stored at 4°C for subsequent analysis.

Collection of Serum:

5ml venous blood was withdrawn from the patient, centrifuged and serum obtained was stored at 4°C for subsequent analysis.

Estimation of Vitamin C by DiNitroPhenylHydrazine (DNPH) method[13]

Vitamin C or ascorbic acid being a good reducing agent can undergoes reversible conversion to its oxidised form dehydroascorbic acid. Both these forms couple with 2,4-dinitrophenyl hydrazine to yield an ozazone which gives yellow color with sulphuric acid. Copper in the dinitrophenyl hydrazine reagent acts as a catalyst, and the intensity of the colour is read at 520nm.

Statistical Analysis :

One-way ANOVA was used to correlate vitamin C level in saliva and serum and DMFT index. Results are presented as mean ± standard deviation value. 'p' value of 0.05 or less was considered significant.

Result :

The mean levels of vitamin C in saliva of control group was 1.243±0.239 and that of group I, II and III were 0.985± 0.151, 0.853±0.309 and 0.721± 0.151. 'P' value was statistically significant (P<0.05).

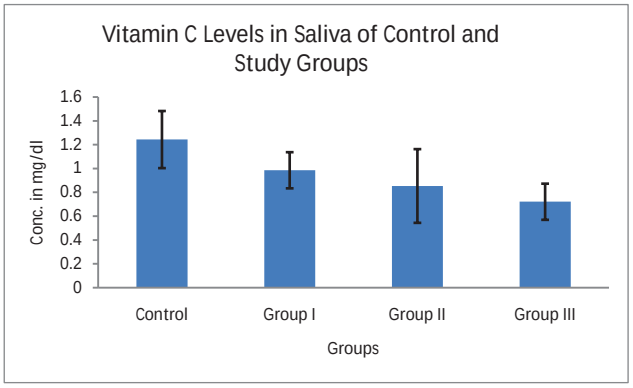
Table 1: Correlation between salivary and serum vitamin C levels in caries free and caries active adults.

Parame-ters	Caries Free Group Mean±SD (µM/L)	Caries Active Group			'P' value
		Group I Mean±SD (µM/L)	Group II Mean±SD (µM/L)	Group III Mean±SD (µM/L)	
Saliva	1.24±0.24	0.98±0.15	0.85±0.30	0.72± 0.15	P<0.0001
Serum	1.38±0.24	1.27±0.17	1.15±0.21	1.08±0.22	P<0.0004

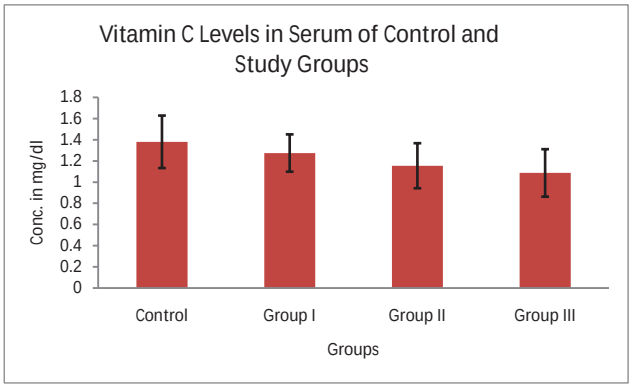
'p'<0.05 is statistically significant. Statistical comparisons were performed by one-way ANOVA. Data are expressed as mean ±SD

Keywords: Vitamin C, Saliva, Serum, Dental caries

Graph 1: Comparision saliva and serum vitamin C levels in caries free and caries active adults.



Graph 2: Comparision of serum vitamin C levels in caries free and caries active adults.



The mean level of vitamin C in serum of control group was 1.380±0.247 and that of group I, II and III were 1.274± 0.176, 1.154± 0.212 and 1.086±0.224. 'P' value was statistically significant (P<0.05).

Discussion :

The rich variety of molecules present in the salivary secretions renders saliva an attractive possible source of disease biomarkers. Over the last few years salivary research workers have been developing salivary diagnostic tools to monitor both oral and systemic disease. Saliva has many apparent advantages over serum as a medium for clinical diagnosis. Recent reports have identified several potentially useful biomarkers in saliva[14,15].

Salivary antioxidant system was found to reduce the susceptibility to dental caries[16]. The specific role of antioxidants is to neutralize rampaging free radical and thus reducing its capacity to damage. They act as radical scavenger, hydrogen donor, electron donor, peroxide decomposer, singlet oxygen quencher, and synergist[17]. Large numbers of free radicals are produced during the process of dental decay. The numbers of free radicals vary directly with the degree of activity of the caries.

Antioxidants might adversely affect the oxidative carbohydrate metabolism within dental plaque thereby reducing bacterial activity and growth and consequently dental caries severity [15]. Vitamin C acts as a powerful water-soluble antioxidant[18]. Furthermore, it inhibits lipid peroxidation by reduction of tocopheroxyl radical to tocopherol and so protects the cell membrane from external oxidants[19]. Water soluble antioxidant nutrients (reduced vitamin C) are initially consumed, followed by lipid soluble antioxidants (alpha tocopherol) in quenching the free radicals. Also, it has been reported that vitamin C regenerates vitamin E by nonenzymic mechanism[19].

Vitamin C (ascorbic acid) is an essential nutrient that serves to maintain the integrity of teeth. A prolonged deficiency of vitamin C results in tooth loss[20]. The relationship between the serum level of vitamin c and number of carious lesions in the mouth is poorly understood. The study suggests that saliva and serum vitamin C level decreases with increase in caries activity and is statistically significant. The result is in accordance with our previous study[21]. Variation in serum vitamin C levels between the study groups is because the dietary vitamin c intake was not controlled in groups. The decrease in vitamin C levels between the control and study group may be related to its antioxidant property of neutralizing the free radicals.

Conclusion :

Vitamin C plays an important role in maintaining the integrity of the teeth and also as an non-enzymatic antioxidant defense system. From the study it is evident that serum and salivary vitamin C level decreases with increase in caries activity suggesting the powerful antioxidant property of vitamin C.

Keywords: Vitamin C, Saliva, Serum, Dental caries

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