# Gingival health in relation to salivary vitamin C and total protein among dental students

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## ABSTRACT

Background: The protective roles of vitamin C and total proteins in gingival inflammation were reported by several studies. The aim of this study was to measure the concentration of salivary vitamin C, total protein and their relation to gingival health among dental students.

Materials and methods: The sample consisted of 67 dental students (33 males and 34 females) from College of Dentistry, University of Baghdad. Sillness and Löe (1964) was used for recording of dental plaque, while the gingival index (GI) was measured according to Löe and Sillness criteria (1963). Stimulated salivary samples were collected and chemically analyzed in Poisoning Center/Surgical Specialty Hospital by using colorimetric method to measure the salivary vitamin C and total protein. SPSS version 18 was used for analyzing data.

Results: A higher percentage of dental students were found with mild type of gingivitis. Higher mean value of salivary vitamin C was reported among dental students with mild type of gingival index compared to those with moderate type, while the opposite picture was noticed for total protein, differences were statistically significant (P< 0.05).

Conclusion: Significant associations between salivary vitamin C, total protein and gingivitis were found in the present study. The protective rule of salivary vitamin C and total protein may offer a route to improve oral healthcare.

Key Words: Gingival disease, vitamin C, total protein, periodontal disease, healthy individuals. (J Bagh Coll Dentistry 2014; 26(3):156-159).

**الخلاصة**

سجلت دراسات عديدة الدور الوقائي للفيتامين ج والبروتينات الكلية في التهاب اللثة. الهدف من هذه الدراسة هو لقياس تركيز فيتامين ج والبروتين الكلي في اللعاب وعلاقتهما بصحة اللثة لدى طلبة طب الاسنان.

المواد والطرق: تكونت العينة من سبعة وستين طالب من طلبة كلية طب الاسنان/جامعة بغداد (ثلاثة وثلاثون ذكر واربعة وثلاثون انثى). استخدم مؤشر دليل اللويحة الجرثومية (Sillness and Löe, 1964) ، بينما استخدم مؤشر اللثة تبعا لتصنيف (Löe and Sillness, 1963). تم جمع عينات من اللعاب المحفز وحللت كيميائيا في مختبر السموم/مستشفى الجراحات التخصصية باستخدام (colorimetric method) لقياس فيتامين (ج) والبروتين الكلي. حللت البيانات احصائيا باستخدام SPSS 18.

النتائج: وجد ان النسبة العالية من طلبة طب الاسنان مصابين بالتهاب اللثة الخفيف، كما سجل لديهم ارتفاع قيمة متوسط فيتامين (ج) في اللعاب مقارنة الى الذين لديهم التهاب اللثة المتوسط. بينما لوحظ العكس للبروتين الكلي، مع فروق معنوية احصائيا.

الخلاصة: وجدت هذه الدراسة علاقات معنوية واضحة بين فيتامين (ج)، البروتين الكلي والتهاب اللثة. ان الدور الوقائي لفيتامين (ج) والبروتين الكلي ربما يكون الطريق الى العناية بصحة الفم.

## INTRODUCTION

Periodontal disease is associated with increased oxidative modification of salivary DNA, lipids, and proteins [1]. Gingivitis and periodontitis are oral diseases that are characterized by chronic inflammation. Salivary protein and albumin concentrations were determined as markers for plasma protein leakage, occurring as a consequence of the inflammatory process [2].

Saliva possesses a wide range of antioxidants including uric acid, vitamin C, proteins, reduced glutathione, oxidized glutathione, and others [3]. Such antioxidants work in concert, and total antioxidant capacity may be the most relevant parameter for assessing the defense capabilities [4]. Periodontitis has been recognized as a risk factor for certain systemic diseases where low-grade inflammation within the peripheral circulation is associated with the etiology or progression of the disease. These manifestations of increased oxidative stress provide potential mechanisms whereby periodontal inflammation may impact upon systemic inflammatory status [5]. Vitamin C has long been a candidate for modulating periodontal disease. Studies of effects of vitamin C on extracellular matrix and immunologic and inflammatory responses provide a rationale for hypothesizing that vitamin C is a risk factor for periodontal disease [6]. Once the teeth are fully formed, vitamin deficiencies which cause gum damage, i.e. lack of vitamin C, will cause tooth loss [7]. No Iraqi study was noticed among dental students that searched the relation between salivary vitamin C, total proteins and gingival disease, therefore, this study was conducted.

## MATERIALS AND METHODS

The sample was chosen from College of Dentistry, University of Baghdad comprised of 67 dental students (33 males and 34 females) aged 22 years old. Plane mouth mirror and periodontal probe were used. Dental plaque (PI) recorded [8] by the criteria described by Sillness and Löe. The gingival index (GI) of Löe and Sillness used [9] for the assessment of gingival health, the whole teeth was examined and four surfaces of each tooth were scored.

Each student was asked to chew a piece of Arabic gum (0.5-0.7 gm) for 5 minutes for collection of whole stimulated saliva in a sterile capped bottle using a standardized method [10].

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Level of vitamin C (mg/dl) in saliva was determined photometrically with 2, 4-dinitrophenyl hydrazine (DNPH) to form red bis-hydrazone [11]. Total protein (mg/dl) determined by colorimetric method. A ready kit was used by Labkit, Nau J. Protein react in acid solution with pirogallol red and molybdate to form a colored complex. The intensity of the color formed is proportional to the protein concentration in the sample [12].

The values of this study were subjected to statistical analysis by SPSS version 18 (Statistical Package for Social Sciences) to specify the statistical differences between the groups. Mean and SD and the parametric statistical tests of significance were used. The independent samples t-test was used to test the statistical significance of difference in mean between groups of study. The linear correlation between two quantitative variables is measured by Spearman's rank linear correlation coefficient, while multiple linear regression was used to assess independent effect of explanatory variables on dependent quantitative variable. The confidence limit was accepted at 95%.

# RESULTS

Table (1) shows the distribution of the total sample by gender. The sample was consisted of males and females as a higher percentage of females were noticed compared to males.

**Table 1: The distribution of total Sample (Dental students) according to gender**

| Gender | No. | % |
|---|---|---|
| **Male** | 33 | 49.25 |
| **Female** | 34 | 50.75 |
| **Total** | 67 | 100 |

Table (2) demonstrates the mean values of plaque index, gingival index, salivary vitamin C and total protein (mg/dl) of total sample. The mild type of plaque and gingival indices was represented a minimum score while the maximum score was recorded of moderate type.

**Table 2: Oral indices and salivary variables (Mean±SD) of the total sample (Dental students).**

| Indices and Variables | No. | Min. | Max. | Mean | ±SD |
|---|---|---|---|---|---|
| **PlI** | 67 | 0.20 | 1.92 | 0.62 | 0.49 |
| **GI** | 67 | 0.14 | 1.90 | 0.59 | 0.49 |
| **Vitamin C (mg/dl)** | 67 | 0.05 | 0.08 | 0.061 | 0.007 |
| **Total protein (mg/dl)** | 67 | 0.18 | 0.29 | 0.261 | 0.018 |

Table (3) represents the mean values of salivary vitamin C and total protein following the severity of gingival index among dental students. The severe type of gingival index was not found among dental students, only mild and moderate types were noticed. Higher mean value of salivary vitamin C was recorded among dental students with mild type of gingival index compared with students with moderate gingivitis, differences were statistically significant (t= 2.342, df= 65, P< 0.05). While total protein was noticed in higher mean value among dental students with moderate gingivitis, differences were statistically significant (t= -2.192, df= 65, P< 0.05).

**Table 3: The mean values of salivary vitamin C and total protein following the severity of gingival index among dental students**

| Gingival Index severity | No. | % | Vitamin C | | Total protein | |
|---|---|---|---|---|---|---|
| | | | Mean | ±SD | Mean | ±SD |
| **Mild (0.1-1)** | 50 | 74.63 | 0.062 | 0.007 | 0.258 | 0.015 |
| **Moderate (1.1-2)** | 17 | 25.37 | 0.058 | 0.003 | 0.270 | 0.024 |

Table (4) demonstrates the correlation between PI, GI with salivary vitamin C and total protein among dental students. Strong negative highly significant correlations were found between PI, GI with salivary vitamin C. While positive highly significant correlation with total protein. A strong positive highly significant correlation was recorded between PI with GI (r= 0.99, P< 0.001).

**Table 4: Correlation coefficient between plaque index, gingival index with salivary vitamin C and total protein among dental students**

| Variables | PI | | GI | |
|---|---|---|---|---|
| | r | P | r | P |
| **Vitamin C** | -0.509 | <0.001** | -0.522 | <0.001** |
| **Total protein** | 0.432 | <0.001** | 0.431 | <0.001** |

** **Highly Significant**

Table (5) represents the multiple linear regressions of GI with plaque index and salivary variables. For each one unit increased in plaque index (PI), the gingival index (GI) increase significantly by 0.991. The model was statistically significant and being able to explain 99% of variation in the gingival index (GI).

**Table 5: Multiple linear regressions of gingival index (GI) with salivary variables and plaque index**

| Variables | Partial Regression Coefficient | Standardized Coefficient | P-Value |
|---|---|---|---|
| **Vitamin C** | -1.731 | -0.025 | 0.051 |
| **Total protein** | -0.290 | -0.011 | 0.364 |
| **PI** | 0.991 | 0.989 | 0.000** |

**P (model) < 0.001 $R^2$= 0.99 **Highly significant**

# DISCUSSION

In the present study mild and moderate type of gingivitis were affected dental students while severe type was not noticed this finding due to the careness of dental students for their oral health as well as the oral cleanliness by the use of oral hygiene measures. The study reported a positive strong highly significant correlation between gingival inflammation and plaque index. Dental plaque is the main local factor related to the variation in the prevalence of periodontal disease [13]. This finding was also reported by Iraqi studies and other studies in the world [14-18].

The study revealed a higher mean value of vitamin C among dental students with mild type of gingivitis compared to those with moderate type, as a negative highly significant correlation was recorded with plaque and gingival index. This finding was also reported by other studies [19-21]. As the most effective physiological antioxidant, vitamin C may also generate a disadvantageous environment for the optimal growth and survival of P. gingivalis. Accordingly, it is possible that an extremely low vitamin C concentration may increase colonization of P. gingivalis, but it is also conceivable that it disturbs the healing of the periodontal tissues. Since the major function of ascorbic acid is its involvement in the synthesis of collagen fibers, a very low vitamin C status may prevent the regeneration of periodontal tissues. However, the attachment ligaments or alveolar bone lost due to the inflammation response will not be revived [22-25].

Salivary total protein is a vital component of saliva, with salivary proteins, predominantly comprising proline rich proteins, mucin, amylase, immunoglobulins, statherin and antibacterial factors, and these are responsible for most of the functions of saliva [26]. The present study recorded a positive highly significant correlation between total protein with plaque and gingival index as a higher mean value was noticed among dental students with moderate type of gingivitis. The same findings were recorded by other studies [2,21,27,28]. In general, the major factors affecting the protein concentration and composition of whole saliva are the salivary flow rate, protein contributions of the glandular saliva and crevicular fluid proteins. Thus, the elevated protein levels are most likely due to enhanced synthesis and secretion by the individual glandular saliva. Also, glandular-derived proteins, Cystatin C and amylase showed significant rise in periodontitis subjects, proving the glandular origin of these proteins [29]. Reactive Oxygen Species (ROS) are implicated in the destruction of the periodontium during periodontitis. The imbalance in oxidant activity may be a key factor and the elevated level of total proteins may provide a protective rule against Reactive Oxygen Species (ROS) [1,30,31]. The present study concluded that the protective rule of salivary vitamin C and total protein may offer a route to improve oral healthcare.

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