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Original Article

Significance of Antioxidants in Diabetic Individuals with Periodontitis

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Abstract

Introduction: The purpose of this research was to evaluate the serum level of vitamin C in patients with type 2 diabetes mellitus with and without periodontitis, and also to interpret whether raised or lessened serum vitamin C levels might be associated to a possibility for building oxidative stress in type 2 diabetes mellitus with periodontitis. **Material and Methods:** Investigation was conducted on diabetic subjects with and without periodontitis. The serum vitamin C level was determined using the 2, 4- dinitrophenylhydrazine method, and the spectrometer was used to read the concentration of serum vitamin C. The data attained were statistically calculated using the ANOVA test and post-hoc test. **Results:** The findings revealed that average serum vitamin C levels were reduced considerably among test groups (groups B, C, D) ($p< 0.0001$) in contrary to the healthy control group. Conclusion: The existing results are proof that reduced concentration of antioxidants such as vitamin C is related to a greater threat to the increase of oxidative stress in patients with type 2 diabetes mellitus and periodontitis.

Keywords: Antioxidants, Periodontitis, Oxidative stress, Type 2 diabetes mellitus, Vitamin C.

Introduction

Periodontal infection is one of the most common diseases that destroy the soft tissue, as well as the hard tissues surrounding the teeth. If such a condition is not managed, the bone supporting the teeth resorbs, leading to tooth loss [1, 2]. Hypothetically, individuals with periodontitis are at greater possibility of acquiring diabetes in addition to what is universally expected [3]. Diabetes is associated with compromised wound healing, exaggerated monocyte reaction to dental plaque microorganisms [4] in addition to decreased neutrophil chemotactic reactions, all of which can lead to increased local tissue damage. It has been assessed that diabetes mellitus affects over 552 million individuals globally by the year 2030 [5].

Vitamin C is an essential water-soluble micronutrient that has antioxidant, anti-carcinogenic, as well as anti-inflammatory properties [6-8]. Epidemiological reports state an adverse association among plasma vitamin C concentration as well as the severity of periodontitis [9, 10]. Vitamin C insufficiency leads to scurvy with decreased growth and collagen production, intensified periodontal destruction, bleeding, as well as tooth loss. On the other hand, Staudte et al. stated that the consumption of grapes leads to an upsurge in serum vitamin C concentration, and it lessens gingival bleeding [11]. It is very probable that these biochemical cycles of vitamin C [12, 13] result mainly from its chemical features of a reducing and chelating agent.

All these interpretations suggest that vitamin C is more closely associated with human pathophysiology than formerly believed. This study was done to assess the serum vitamin C concentration in patients with type 2 diabetes mellitus with and without periodontitis.

Material and Methods

Study Subjects and Ethical Approval

The study was conducted at the A. B. Shetty Institute of Dental Sciences in the outpatient department

of periodontics located in Mangalore City, India. After obtaining ethical approval from the institutional ethics committee, a single-blinded study was conducted. The research comprised of a sum of 40 subjects in the age group 25 to 60 years. The individuals were separated into four categories, comprising of 10 members in every group as follows:

- Group A – Control healthy subjects
- Group B – Healthy subjects with periodontitis
- Group C- Subjects with type 2 diabetes mellitus and periodontitis
- Group D - Subjects with type 2 diabetes mellitus without periodontitis.

Selection criteria were established by inclusion and exclusion measures. The inclusion criteria were as follows: individuals diagnosed with diabetes taking oral anti-hyperglycemic agents according to the classification of the American Diabetes Association [14], systemically healthy individuals for group A, individuals diagnosed with chronic periodontitis, according to the 1999 classification of the American Academy of Periodontology for groups B and C [15]. The exclusion criteria involved pregnant or lactating women, smokers, individuals taking dietary supplements that underwent periodontal therapy and diabetic individuals with uncontrolled blood sugar levels

Clinical Assessment of Study Subjects

A regular proforma comprising the following information: name, age, gender, previous treatment record (if any), gingival score (Loe and Silness Gingival Index) [16], plaque score [16], clinical attachment and periodontal pockets according to the 1999 classification of chronic periodontitis [15] was recorded. Every individual was inspected by means of a mouth mirror, as well as Williams' graduated periodontal probe under artificial light. Written informed consent was obtained from each patient before the examination and drawing blood.

After the initial screening of all patients, the subjects were divided into groups, and 5 ml of venous blood was drawn to assess the Vitamin C level using the dinitrophenylhydrazine method, and the results were read using a visible spectrophotometer.

Analysis

The obtained results were tabulated and subjected to statistical analysis; the mean values and standard deviation were calculated. Tukey's post-hoc comparison test was carried out for comparison between the groups.

Results

The mean and standard deviation for serum Vitamin C is significantly higher in the control group in contrast to the test groups.

The ANOVA test showed significance in all parameters between groups and within groups, $p < 0.001$.

Discussion

Several human ailments are linked with a build-up of oxidative stress causing moreover from the distorted formation of free radicals or commencing the modified antioxidant elements or actions. Metal ions are recognized to perform an important part in living organisms, both in the development and physiological functions. Reduced absorption of trace components is witnessed in diabetic patients with periodontitis. Amongst the antioxidants usually eaten by humans, the antioxidant Vitamin C is of precise significance. Our study revealed a statistically significant negative correlation of Vitamin C among groups (B, C, D), as represented in Tables 1-3.

One chief association among periodontitis, as well as the chronic systemic inflammatory condition, is that they are all intensely related to the occurrence of oxidative stress. Oxidative stress reinforces the pathogenesis of periodontitis [17] and type 2 diabetes mellitus [18]. Oxidative stress may upshoot mitochondrial proton as well as electron seepage, which causes the main influence on mitochondrial linking inefficiency, leading to the formation of reactive oxygen species. Anoth-

Table 1: Mean and Standard Deviation for Serum Vitamin C.

Group	Vitamin C in mg/dl: -mean -std dev
Healthy	1.40 0.565
Healthy with Periodontitis	1.01 0.287
Type 2 Diabetes with Periodontitis	0.920 0.23
Type 2 Diabetes without Periodontitis	0.93 0.29

Table 2: Quantitative analysis of Vitamin C was done using ANOVA.

Parameters	Groups	Sum of Square	Mean squares	F	P
Vitamin C in mg/dl	• Between groups • Within groups • Total	3.850 13.12 16.97	1.28 0.13	9.38	<0.001

Table 3: Post-Hoc test for Vitamin C.

Groups	Mean Difference	Std Error	P value
Healthy vs. Healthy with Periodontitis	0.389	0.104	0.002 <0.05
Healthy vs. Type 2 diabetes with Periodontitis	0.482	0.104	<0.001 significant
Healthy vs. Type 2 diabetes without periodontitis	0.465	0.104	<0.001 Significant
Healthy with periodontitis vs. Type 2 diabetes with periodontitis	0.093	0.104	0.806 i.e.>0.05
Healthy with periodontitis vs. type 2 diabetes without periodontitis	0.077	0.104	0.881 i.e.>0.05
Type 2 diabetes periodontitis vs. type 2 diabetes healthy	0.166	0.104	0.999 i.e.>0.05

er method by which dietary processed carbohydrates, as well as saturated fat consumption, produce free radicals is through to the role of advanced glycation end products (AGEs), and their receptor (RAGE), which plays a key role in the inflammatory actions leading to the development of diabetes in addition to its complications, or when minimally modified LDL activates Toll-like receptors, even reported of bounding to the C-reactive protein, which initiates metabolic disorders. Another reason of free radical production in humans is because of continuous production by antimicrobial peptides in the course of a degenerative action after a stimulus, for example, pathogens coated with opsonin, bacterial DNA or peptides, and stimulation of the hexose monophosphate shunt which makes use of molecular oxygen and NADPH as an electron donor [19].

Our results, as shown in Tables 1-3, are similar to previous studies where it was noticed that adults, as well as old aged individuals with type 2 diabetes mellitus, develop a drop in vitamin C levels, in addition to the upsurge of oxidative stress biomarkers [20] when matched to healthy subjects. Since Vitamin C derivatives aggregate in water solution, it has been suggested that it is guided and bounds at the outer layer of lipid bilayers tissues, thus leading to active retardation of free radicals occurring in the aqueous solution, and in effect, the restoration of fat-soluble antioxidants [21]. High blood sugar-induced oxidative stress plays an important part in the progress of diabetic complications, which is thought to be caused by increased reactive oxygen types production, in addition, due to reduced antioxidant protective action. An upsurge in blood glucose values in diabetic rats was detected after the initiation of diabetes by streptozotocin [22].

The association concerning periodontitis with vitamin C was also established by a British report [23], which revealed a converse association among plasma vitamin C concentration and serum antibody concentration of Porphyromonas gingivalis in an unmethodical subsample of the European population. Furthermore, in a non-scorbutic trial periodontitis rat model, it was revealed that vitamin C additions decreased the degenerative activity in the gingiva [24].

In a study conducted by Amaliya et al. that involved Indonesian inhabitants underprivileged to receive dental care, a noteworthy converse association among plasma vitamin C values and tissue degeneration was noted [25]. Similarly, a converse association between

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Vitamin C and periodontitis was found in the present study.

Vitamin C is a dominant antioxidant radical scrounger in the aqueous stage. Antioxidants help to soothe these extremely oversensitive free radicals, thus preserve the organizational as well as working integrity of cells. The relationship with periodontal-pathogenic microorganisms interrupts this delicate equilibrium, causing collagen breakdown and subsequent periodontal pocket development. An upsurge in the serum vitamin C concentration will have a beneficial result on periodontitis. As this vitamin is water-soluble, its oxidative counteraction activity depends mainly on the hydration status, identical to how leucocytes gather vitamin C to defend themselves against oxidative stress during phagocytosis.

Definite methods of conveyance, as well as digestion, assemble vitamin C in the tissues to improve its purpose as an enzyme cofactor and a foraging antioxidant [26]. Conversely, it is uncertain how vitamin C consumption disturbs gingival oxidative stress, gene expression encoding inflammation, as well as cell behavior in periodontal lesions, even though certain reports propose that vitamin C upsurges the number of collagen fibers in the renewing periodontal tissue in addition to clearing histamine in gingivitis [27]. Tests showed that the antioxidant action of Vitamin C includes the hydrogen atom transfer more than the transfer of electrons.

It was earlier projected that a continuous photochemical, as well as ambient production of active oxygen types, may compose a noteworthy threat in the etiology of periodontitis. On the other hand, under circumstances of raised radical formation as well as/or reduced antioxidant scavenging action, oxidative tissue injury may happen. In case of type 2 diabetes mellitus with periodontitis, reactive oxygen species formed by vitamin C might not only perform a part in the host protection contrary to the bacteria but also may affect oxidative injury to host tissues.

Conclusion

We assume that a drop in Vitamin C levels is linked to a more significant threat for type 2 diabetes mellitus with periodontitis when related to healthy individuals. Vitamin C acts as an effective scrounger of water-soluble free radicals as well as oxidants, thus guarding new biomolecules against oxidative destruction. More well-regulated clinical trials are essential to assess the result of reduced values of vitamin C on the threat of acquiring disease pathogenesis and the development of type 2 diabetes mellitus in patients with periodontitis, as well as before making general public health recommendations.

Conflict of Interest

The authors declare no conflict of interest.

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