REVIEW
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Examination of Research on Vitamin C in Periodontology

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Abstract

Periodontitis is a chronic, multifactorial disease characterized by the destruction of periodontal tissues, in which tooth loss can occur if left untreated. Periodontitis occurs as a result of the interaction between periopathogenic bacteria in the biofilm layer and the host response. More than 10% of the world's population is affected by severe periodontitis. The effect of vitamin C on gingival diseases has been a subject that has been emphasized a lot in previous studies and has been re-focused on recently. Considering the effects of vitamin C on collagen synthesis, immune cells, and antioxidant capacity, it is thought that it may be beneficial for periodontal tissues. In this review, we aimed to analyze recently published studies in which the areas of vitamin C use in periodontology practice and how they can be used in the future were examined.

Keywords: Periodontology, periodontitis, depigmentation, review, vitamin C, ascorbic acid

Periodontitis is a chronic, multifactorial inflammatory disease associated with dysbiotic plaque biofilms and characterized by progressive destruction of the tooth-supporting apparatus. Its primary features include the loss of periodontal tissue support, manifested through clinical attachment loss (CAL) and radiographically assessed alveolar bone loss, presence of periodontal pocketing, and gingival bleeding.[1] Individuals affected by gingivitis and periodontitis constitute 47% of the US population and 43% of the Australian population. More than 10% of the world's population is thought to have advanced periodontitis.[2,3]

Destruction of periodontal tissues, clinical attachment loss, and bone loss are observed in periodontitis.[4,5] With the help of histopathological examinations, pathological pocket formation, displacement of the junctional epithelium toward the apical region of the enamel–cementum border, increased accumulation of polymorphonuclear leukocytes (PMNL) in the junction and pocket epithelium, and infiltration of macrophages, lymphocytes, and plasma cells are observed. Although the tooth-associated biofilm plays a crucial role in the initiation and progression of periodontitis, it is primarily the host inflammatory response that inflicts irreversible damage to the periodontal tissues leading, in some cases, to tooth loss.[5,6] Early bacteriological studies revealed dramatic differences in the composition of the periodontal microbiota in health and disease. This shift in bacterial community composition could be interpreted in 2 ways. First, it could be taken as a sign that specific bacteria are involved in the etiology of periodontitis, in that the disease-associated microbiota contained novel species or periodontal pathogens, which were either not present or barely detectable in the healthy state. Second, it could imply that the disease is caused by dysbiosis of the periodontal microbiota. In that case, a change in the relative abundance of individual components of the microbiota compared with their abundance in health leads to alterations in the host–microbial crosstalk sufficient to initiate inflammatory disease. The quest to identify specific periodontal pathogens has led to significant progress, including the identification of some candidates, mostly gram-negative anaerobic bacteria that colonize subgingival tooth sites. Foremost among this group are 3 species that comprise the so-called "red complex," are frequently isolated together, and are strongly associated with diseased sites in the mouth: *Porphyromonas gingivalis*, *Treponema denticola*, and *Tannerella forsythia*. Research work has been directed toward understanding the pathogenic mechanisms

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and virulence determinants of these 3 bacterial species.[5-7] However, periodontitis is not observed in every individual with these pathogenic microorganisms; this situation reveals that most individuals are in balance with their biofilm, but when the current balance is disturbed, the disease develops. Dysbiosis occurs as a result of environmental factors, genetic factors, systemic diseases, oral hygiene habits, stress, smoking, etc. that cause the increase of pathogenic microorganisms or insufficient host defense mechanisms.[1,2,8]

The new classification system published by the American Academy of Periodontology in 2017 evaluated the course of the disease and the presence of systemic diseases with clinical attachment loss around the teeth, radiographic bone loss, tooth loss, and bleeding on probing. In the initial evaluation, the patient is given a stage and grade based on the below-mentioned criteria. The classification of periodontitis is based on stages defined by severity (according to the level of interdental clinical attachment loss, radiographic bone loss, and tooth loss), complexity, and extent and distribution. Stage 1 periodontitis is based on 1-2 mm interdental CAL and <15% radiographic bone loss. Stage 2 periodontitis is based on 3-4 mm interdental CAL and 15%-33% radiographic bone loss. Stage 3 is the condition in which there is severe damage to the periodontal tissues and tooth loss if not treated. Stage 4 is usually expressed as damage to periodontal tissues causing tooth loss and difficulty chewing food because of periodontal tissue destruction. Classification of periodontitis is based on grades that reflect biological features of the disease including evidence of, or risk for, rapid progression, anticipated treatment response, and effects on systemic health. The grade is graded as A, B, or C, with C being the most severe.[4,9]

Main Points

- The only proven indication of vitamin C deficiency in clinical practice is scurvy, and current studies are refocusing on the clinical benefits of vitamin C.
- It is a misconception to think of vitamin C as a medicine and a therapeutic agent, but it has been observed to be beneficial when used as an adjunct to treatment.
- The effect of vitamin C is significant in the destruction caused by oxidative stress. For this reason, it is more rationale to consider vitamin C supplementation in the case of diabetes, rheumatoid arthritis, and heavy smoking accompanying periodontitis.
- Vitamin C is also an auxiliary agent in treating depigmentation, apart from periodontitis, and promising results have been obtained.
- Considering that it is an easily accessible, easily tolerated, and economically viable agent, it is considered a promising agent that can help the physician's treatment in clinical practice. Still, in particular, clinical studies on the subject should be increased. The dose, frequency of use, and what patient groups would benefit from Vitamin C should be investigated in more depth.

An initial periodontal treatment includes debridement, root planning, and providing oral hygiene education to the patient, sometimes with systemic or local antibiotics. Although this treatment protocol is quite effective, it can be limited as it depends on patients' oral hygiene habits and continuing their dental examinations.[5] Despite microorganisms being the primary factor in periodontitis, the host response determines the prognosis of the disease.[10,11] The host's response to periopathogenic bacteria is influenced by the host's systemic factors such as diabetes and cardiovascular disease and environmental factors such as smoking.[11] Reactive oxygen species (ROS) and inflammatory cytokines such as IL-1beta and IL-6 are effective in the modulation of the host response.[12,13] Reactive oxygen species affect host modulation by causing DNA damage. Antioxidants, on the other hand, are thought to reduce the attacks on DNA by free radicals, thus protecting mutations that cause disease.[13,14] Antioxidants are divided into 2 groups as natural and synthetic antioxidants. Enzymes (superoxide dismutase [SOD], CAT, expression of glutathione peroxidase [GSH-Px], cytochrome C oxidase, hydroxyperoxidase), macromolecules (transferrin, ferritin, myoglobin, haptoglobin), and micromolecules (β-carotene, vitamin A, vitamin C, ubiquinone) are examples of natural antioxidants. Unnatural antioxidants, on the other hand, are synthetic substances used to extend the shelf life of foods in industrial production. Vitamin C, an important antioxidant, is present in body fluid as ascorbate and protects biological membranes from oxidative damage by inhibiting peroxide radicals in the water phase.[15] Vitamin C has an important role in processes such as oxidative stress reduction, iron absorption, carnitine synthesis, and various hormone functions.[15,16]

Previous studies have been conducted on the use of vitamin C in patients with gingivitis and periodontitis, and this issue has been reviewed in current studies.[16-18] The only documented indication of vitamin C in clinical practice is scurvy.[19,20] When the first studies in the literature were examined, the studies on ascorbic acid started in 1907. In 1912, considering that scurvy disease was caused by deficient intake of a factor in food, researchers named this substance they found as "antiscorbutic vitamin." In the study published by Drummond et al in 1975, "vitamin C" was started to be used instead of antiscorbutic vitamin.[19-21] The serum/plasma concentration of vitamin C is lower in individuals with gingivitis and it has been reported that bleeding and gingivitis are reduced when vitamin C is supplemented in addition to periodontal therapy in these patients.[21,22] It has been reported that vitamin C supplementation reduces oxidative stress in gingival tissues in rats with experimental periodontitis.[23] An inverse relationship was also established between the severity of necrotizing ulcerative gingivitis and vitamin C plasma levels. It is thought that this situation is due to collagen synthesis; therefore, vitamin C deficiency causes a decrease in healing capacity.[21,22,24-26] Woelber et al[27] published a study with 30 participants to evaluate the effect of an anti-inflammatory

diet on gingivitis. Significantly reduced gingivitis in a clinically relevant range was shown in the experimental group, while serological inflammatory parameters and the subgingival microbiome seem to be unaffected by the diet.[27] Martinon et al[28] evaluated the relationship between periodontal and chronic diseases and diet. As a result of this study, it has been concluded that a diet with low sugar, high fiber, and high omega-6, omega-3 fatty acid ratio reduces the risk of cardiovascular disease, diabetes, cancer, and periodontal disease. In a randomized, double-blind, placebo-controlled multicenter study published in 2019, combinations of vitamin C, vitamin E, lysozyme, and carbazochrome (CELC) were used in addition to periodontal treatment in chronic periodontitis patients; the effects on gingivitis were investigated by evaluating the gingival index, plaque index, and periodontal pocket depth; when the results at both the fourth and eighth weeks were compared in terms of the gingival index parameter, a statistically significant result was obtained in favor of the test group. However, other parameters were similar between the groups. Consequently, researchers suggested that a larger sample is required to clarify the clinical efficacy and benefits of CELC.[29] In another study, researchers evaluated the effects of an optimized diet on the health of oral tissues on periodontal clinical parameters over a 4-week period. They evaluated periodontal status through plaque index, gingival bleeding index, periodontal pocket depth, and bleeding on probing. During these 4-week periods, the participants in the experimental group were fed a diet low in carbohydrates and rich in omega-3 fatty medical s, vitamins C and D, antioxidants, and fiber. At the end of the study, despite the similar plaque values in both groups, it was observed that all inflammatory parameters evaluated in the experimental group were significantly reduced than in the control group.[30]

In periodontitis, it has been observed that ascorbic acid supplementation slows down the progression of the disease and induces regeneration by stimulating the progenitor cells in the periodontal ligament. It is thought that ascorbic acid, an effective antioxidant that reduces oxidative stress and participates in various biochemical reactions, may reduce proinflammatory cytokine responses.[31] Studies state that the plasma concentration of vitamin C is lower in periodontitis patients compared to healthy individuals and they show that there is an inverse correlation between serum/plasma vitamin C levels and the severity of periodontitis.[32,33] How vitamin C supplementation affects gingival oxidative stress, gene expression in inflammation, and cell behavior in periodontal tissues has not been certainly defined yet, but studies show that the number of collagen bundles in tissues given vitamin C increases and histamine is detoxified.[23,24] The study by Kuzmanova et al[32] showed vitamin C helps in the bactericidal activity of PMNLs and monocytes by increasing nitric oxide synthesis and reducing DNA damage due to oxidative stress.

In vivo studies have shown that smokers have lower serum levels of vitamin C compared to non-smokers and the decrease in serum vitamin C concentration is thought to be related to the number of cigarettes smoked. It is thought that the low vitamin C level detected in smokers is due to the activation of leukocytes and the formation of ROS as a result of this situation.[33-35] It was determined that the concentration of vitamin C in serum samples obtained from smokers was lower than in the samples from non-smokers. The possible reason is that it may occur in the form of decreased absorption, increased metabolic destruction, increased excretion, and combinations of these conditions. There are many studies showing that daily vitamin C intake is lower in the smoking population than in the non-smoker population, but even if the necessary dietary adjustment is made and the vitamin C intake of both groups is equalized, the serum vitamin C level is lower in the smoking group.[36-39] Smoking and periodontitis increase oxidative stress.[40-43] Studies that evaluate the effect of vitamin C on periodontitis's progress in periodontitis patients during smoking are increasing. In a study about periodontally healthy smokers and non-smokers, the levels of vitamin C and E in the gingival crevicular fluid were compared and it was reported that the concentration of vitamin C decreased in the smoking population.[44] A study published by Nishida et al[16] was conducted with 12 419 participants and the effect of dietary vitamin C on periodontal diseases was evaluated. As a result, a dose-dependent relationship was found between decreased dietary vitamin C and the increased risk of periodontal disease. When the clinical attachment level was evaluated, a weak but statistically significant association between vitamin C intake and periodontal disease was observed in smoking participants. The group that benefited most from vitamin C supplementation was determined to be smokers.

The importance of using vitamin C against periodontal diseases as a powerful antioxidant has been increasing in recent years. Studies defined the relationship between vitamin C and periodontal health and they stated that there is an inverse relationship between plasma/serum vitamin C concentration and periodontitis. Recent studies have focused on the importance of the antioxidant properties of vitamin C against ROS and leukocyte function for patients with periodontitis. The role of both ROS and immune cells is very important in terms of the pathophysiology of periodontitis. In addition to promising studies stating that vitamin C storage in monocytes increases glutathione, reduces oxidative DNA damage, and may be protective for cells, there are also long-term follow-up studies showing that vitamin C supplementation has no effect on oxidative damage biomarkers.[32,45-47] Bogdan et al's[48] study showed a significant link between vitamin C intake and periodontitis progress in periodontitis patients in the course of type 2 diabetes and it was observed that diabetic subjects were more susceptible to oxidative stress and periodontitis development when they received insufficient vitamin C.

A systemic review published in Nutrients in 2020 observed that the vitamin C level in the groups with diabetes and

periodontitis was lower than those in healthy groups. Although the positive effects of vitamin C supplementation were observed in the review, it was stated that the available data are insufficient to reach a definite conclusion and more research is needed to determine the appropriate dose and regimen.[48] When the studies conducted to evaluate the interaction between vitamin C and periodontitis in the literature are examined, contradictory results can be encountered. To obtain correct result, the selected parameters should be determined carefully and accurately. However, when the studies are examined, the general view is that vitamin C intake alone is not sufficient for the treatment of periodontal diseases, but it can be used in addition to the treatment. In addition, there are studies expressing that vitamin C combined with chlorhexidine can prevent and slow the progression of periodontitis.[48,49]

In the medical field, vitamin C is frequently used in dermatology for spot treatment. Similarly, it is suggested that vitamin C could be used as a depigmentation agent in dentistry, but studies on the subject in dentistry are limited. Ascorbic acid can be used as a depigmentation agent because it acts as a reservoir for ROS, copper, and calcium in cells. After ascorbic acid enters the target tissue, it binds to melanin and causes ROS, copper, and calcium deficiencies, resulting in a decrease in melanin production. Ascorbic acid relies on the destruction and affects the function of melanocytes. Other approaches affect the number of melanocyte.[50] In the literature, it is stated that positive results have been obtained in case reports on the use of vitamin C in the treatment of hyperpigmented spots, but researchers state that more studies on the subject are needed.[50]

Relapse of pigmented areas affects the treatment method, number of recall periods, genetic and ethnic factors, tobacco consumption, and hormonal factors. Repigmentation was observed after 6 months in the patient who applied vitamin C with a derma pen. In the case where depigmentation was performed with a scalpel and local ascorbic acid was applied, positive results were obtained but recurrence was reported 9 months later. Most of the available literature shows a lower recurrence rate for cryosurgery and lasers.[50-53] According to the systemic review, vitamin C is widely used in dermatology in the treatment of hyperpigmented spots, and its use for the treatment of melanin hyperpigmentation in dentistry shows promising results, but there is limited documentation on the use of vitamin C as a depigmentation agent in dentistry and the relevant data should be increased by determining more different doses and periods.[44,50,51]

It would be wrong to think of vitamin C as a therapeutic agent alone in periodontology practice, but its use in particular groups and in addition to treatment contributes to recovery. Existing studies have not reached a definite conclusion yet. Clinical studies on the subject, in particular, should be increased; the dosage of the ascorbic acid, the frequency of use, and the subjects in which patient groups benefit more should be determined. However, considering that it is easily accessible, easily tolerated, and economically viable, it is considered to be a promising agent that can help the physician's treatment in clinical practice.[8,9,47,54]

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