THE ACTION OF TREATMENS WITH MICRONUTRIENS AND BIOACTIVE COMPOUNDS IN STOPPING THE EVOLUTION OF THE PERIODONTAL DISEASE

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Abstract

Periodontal disease is a significant public health concern. Inflammatory oral diseases are periodontal infections, oral mucosal lesions, pulpal and periapical lesions. The etiology is multi-factorial and is usually associated with a microbial origin, often caused by over-consumption of free sugars. However, the role of micronutrients in these processes is now becoming apparent. Trends have shifted to the role of micronutrients in oral inflammation. The progression of periodontal disease and the healing of periodontal tissues can be modulated by nutritional status. There are numerous degenerative changes in the oral mucosa that have been observed during specific micronutrient deficiencies. Attempts have been made to use dietary supplementation of certain micronutrients to treat oral inflammatory lesions in conjunction with their standard treatment procedures. Micronutrient supplementation can be administered orally or topically. The current review provides insight into the role of nutrition in oral inflammatory diseases, including periodontal disorders, oral mucosal lesions, pulpal and periapical lesions.

Keywords: micronutrients, bioactive compounds, vitamin A, vitamin B, vitamin D, vitamin E, antioxidants, copper, calcium, selenium, periodontal disease

Purpose

The aim of this study was to address the controversy surrounding the relationship between periodontal disease and changes in periodontal tissue and to assess the type of changes in periodontal disease using nutrients by measuring the clinical status of selected patients affected by periodontitis. Restoring nutrition.

Methods

The patients selected for the study were among those who requested periodontal treatment in the private practice, between August 2019 and July 2020, with symptoms specific to periodontal diseases: changes in shape, color or texture of gingival tissue, edema, bleeding caused easily or spontaneously, gingival pain and sensitivity, gingival pruritus, tooth mobility, bags of different depths, hypergrowth of different degrees, recessions, halitosis.

A total of 9 patients (4 men and 5 women) with chronic moderate / severe periodontitis aged 36-71 years.

Vitamin therapy - almost all vitamins are used: vitamin. A, complex B, vit. C, vit. D, vit. E and PP,

in the form of polyvitamins and Tarosin, which contains vitamins C + P. Vitamins activate capillary circulation, optimize vascular permeability, metabolic exchanges and oxidation-reduction reactions, cell regeneration.

Data were collected on the clinical appearance of patients prior to nutrient treatment. Periodontal control was performed with examination of the gums and periodontal pockets. The study is performed until the end of each patient's nutrient treatment.

1. Introduction

Oral diseases are common public health problems worldwide [1]. Oral diseases of inflammatory origin include periodontal diseases: gingivitis and periodontitis, inflammation of the oral mucosa, pulpal and periapical lesions [2]. Periodontal disease is one of the most common chronic inflammatory disorders [3]. Periodontitis and untreated dental caries leading to pulpal and periapical lesions are the most common cause of tooth loss in adulthood [4, 5]. Oral inflammatory pathologies negatively influence the quality of life, leading to deterioration of daily activities [6]

The etiology of oral inflammatory diseases is multifactorial [[7], [8], [9]] and usually involves a microbial component [10,11]. It is essentially a chronic inflammatory disease caused by specific oral microorganisms (e.g., *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans*), characterized by the loss of supporting periodontal ligament and alveolar bone and is the first cause of tooth loss [3]. Periodontal disease is influenced by many risk factors, such as alcohol, stress, smoking, genetics, diabetes, and hormonal alteration status (i.e., pregnancy or menopause) [4], so that the maintenance of oral and periodontal health becomes today a challenge, especially considering that periodontal disease can be itself a risk factor for a number of chronic disorders. Indeed, it is generally accepted that periodontitis is strictly associated with many chronic diseases, such as type 2 diabetes mellitus (DM2), cardiovascular diseases, inflammatory bowel disease and rheumatoid arthritis [5]. Therefore, there is a growing interest in the relationship between oral health and systemic health.

Undoubtedly, oral microorganisms are indispensable for the pathogenesis of periodontal disease, but, among risk factors, nutrition represents an influential aspect that is often neglected [1]. Indeed, nutritional factors have a vital importance for the equilibrium between oral microorganisms and the host response, from which depends the onset and progression of periodontal disease [6,7]. In the past, health-focused dietary approaches have mostly focused in reducing the consumption of “undesirable” dietary components such as sodium, refined sugars, and saturated fats. However, attention on promoting the consumption of “curative nutrients” has been expanded. This perception of “food or nutrient as a medicament” has encouraged the search of a growing number of new substances categorized from dietary supplements to “nutraceuticals”, with the aim of improving human health, making us fitter and more resilient to disease [8,9].

Numerous potential modifiable and non-modifiable risk factors associated with oral inflammatory disorders have been identified, including smoking, alcohol consumption, stress, poor oral hygiene, systemic health, genetics and epigenetic factors [7, [12], [13], [14]], [15]]. The severity and progression of oral diseases depend on these risk factors [16].

Eating habits are a pervasive modifiable determinant of oral health and play a significant role in the inflammatory processes underlying oral pathologies [17]. The role of macronutrients (carbohydrates, proteins and fats), especially free sugars, in oral health is well established and have been extensively reviewed [[18], [19], [20], [21], [22]]. More recently, the role of micronutrient imbalances found in various systemic inflammatory diseases has been of interest in oral and other inflammatory diseases [23]. Micronutrients that modulate inflammatory responses include vitamins (many of which are

antioxidants) and minerals [24,25]. Diet has also been found to influence the composition of the oral microbiome, which has been indirectly linked to the increased risk of oral diseases [[26], [27], [28]] This review will focus on the role of micronutrients and bioactive compounds in oral inflammatory conditions, including periodontal disease, inflammatory lesions of the oral mucosa and pulpal and periapical lesions.

2. The role of micronutrients in periodontal disease

Periodontal disease is a chronic inflammatory disease that involves the destruction of the supporting structures of the teeth [2,20, [29], [30], [31]]. Periodontal disease has been estimated to have an impact on up to 90% of the world's population [30]. The overall prevalence of periodontal disease increases with age [32] and in 2016 was estimated to be responsible for approximately 4.8 million years of living with disabilities affecting quality of life [33]. The incidence of the disease increases sharply in adults aged between 30 and 40 years [32,34]. Understanding the nutritional factors that contribute to this considerable burden of disease can inform new strategies for preventing and managing these diseases [20,35].

Periodontal disease consists of inflammation of the gums, called gingivitis and inflammation of the periodontal tissues, known as periodontitis [29]. Gingivitis is the mildest form of periodontal disease and is reversible [36] while periodontitis involves the destruction of connective tissue and bone, which is irreversible [37,38]. Periodontal diseases are characterized by inflammatory responses of the host to bacteria, including Porphyromonas gingi-valis and Aggregatibacter actinomycetemcomitans present in dental plaque [[39], [40], [41]] The destruction of periodontal tissues is caused by the production of inflammatory factors released by immune cells, followed by the accumulation of polymorphonuclear leukocytes [42,43]. From this results the release of reactive oxidative species with antimicrobial properties [44]. However, this inflammatory response can also affect the host tissue and, in this way, oxidative stress is associated with an increased risk of periodontal disease [[45], [46], [47]]. Therefore, inflammation is established as a key factor in the pathology of these oral diseases.

Nutrition has a significant role to play in the development of periodontal disease. Dietary deficiencies have been found to have an impact on the prognosis of malnutrition attributed to faster progression [[48], [49], [50]]. The inflammatory response, characteristic of the disorders, can be attenuated by increasing the consumption of vitamins C and D [19, 20, 51]. Research has established that dietary factors contribute to the modulation of microbes in the oral cavity that have been associated with periodontal infections [28, [52], [53], [54], [55]].

The inflammatory processes involved in periodontal disease appear to be fueled by nutritional imbalances. Research on antioxidant nutrients, vitamin D and calcium in periodontal disease has been conducted in human and animal models and highlights primarily their anti-inflammatory and anti-oxidant roles.**2.1.**

Vitamins and antioxidants

Antioxidant nutrients prevent the production of free radicals and stabilize and eliminate existing free radicals [56, 57]. Free radicals cause an oxidative chain reaction that leads to cell damage and cell death [[57], [58], [59]. Antioxidants interrupt this chain reaction by modulating oxidative stress found to be significantly associated with periodontal diseases [[60], [61], [62], [63], [64]]. A wide variety of studies, including long-term, cross-sectional, and randomized clinical trials, have explored the role of antioxidants in oral inflammatory diseases [[65], [66], [67], [68]].

2.2. Vitamin B Complex

[71]. The vitamin B complex refers to eight water-soluble vitamins, which perform many essential functions in the body. B-complex vitamins include thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), vitamin B6 (pyridoxine), folate (B9), biotin (B7) and cobalamin (B12) [69]. Research shows a role for each of the B vitamins in the progression and severity of periodontal disease. Deficiencies of B2, B3, B6 and B12 have been

correlated with hemorrhagic gingivitis and periodontitis [54,70]. Regarding periodontal disease, folic acid deficiency leads to the absence of keratinization of the gingival surface, decreased cell turnover rate, reduced resistance to infections and destruction of gingival and periodontal tissues.
[72]. In addition, folate deficiency in smokers has been reported to increase the risk of periodontitis [76,77]. In smokers, folic acid deficiency can occur even with regular dietary intake, due to the additional use of folic acid needed to convert the compounds present in cigarette smoke into biologically inactive compounds [71]. Moreover, a clinical study of 30 people showed that vitamin B supplementation after apical flap surgery, which is common in the treatment of periodontal disease, increased the level of clinical attachment compared to placebo [78]. In general, there is an increase in periodontitis in smokers, which may be due to folic acid deficiency.

2.2.1. Vitamin C

Vitamin C, also known as l-ascorbic acid, is a water-soluble vitamin that requires regular intake [79]. Vitamin C plays crucial roles in many biochemical functions, including quenching free radicals and modulating inflammatory responses [80,81]. Plasma vitamin C levels decrease during periods of infection and post-surgical stress [82], due to the additional vitamin C required for tissue repair and regeneration in these cases [81, [83], [84], [85]]. Therefore, additional intake may be required in these situations. The anti-inflammatory, anti-oxidant and regenerative actions of vitamin C are well established [62, [86], [87], [88], [89], [90], [91]], suggesting a role for this nutrient in modulating inflammatory responses in periodontal disease.

Vitamin C deficiency has been identified as a significant risk factor in periodontal disease [86,87]. It is well established that dietary deficiency of vitamin C leads to scurvy, an inflammatory disease of the gums that causes bleeding gums [89,90,92,93]. Multiple epidemiological studies have shown that vitamin C status correlates with the risk of periodontitis [[86], [87], [88], [89]]. Cross-sectional studies involving Korean and Japanese populations have found that dietary intake and serum vitamin C levels are independently linked to periodontitis [86,87]. A study using combined Finnish and Russian cohorts showed that vitamin C deficiency was associated with periodontitis, and vitamin C levels were also negatively correlated with antibody levels in Porphyromonas gingivalis [89], gram-negative bacteria effects that cause gingivitis. Moreover, the results of the Third National Health and Nutrition Survey (NHANES III, US Population Representative) showed a weak but statistically significant relationship between lower vitamin C intake and periodontal disease in the general population, and especially to tobacco consumers [88]. These studies suggest that inadequate vitamin C intake increases the risk and association with periodontal disease in different cultural demographies.

Vitamin C can also be used therapeutically to reduce the severity of periodontal disease and to improve treatment outcomes. A longitudinal study linked vitamin C-rich grapefruit consumption to improved vitamin C plasma concentrations and decreased sulcus bleeding scores [68]. The lower prevalence of deep periodontal pockets has also been linked to higher citrus intake [94]. While saturation of plasma vitamin C levels may occur at high doses [41], topical application may also influence oral health. It has also been reported that local intradermal and subepithelial vitamin C injections could be used as an adjunct in the surgical and non-surgical treatment of periodontal disease [95,96]. Moreover, local administration of vitamin C helps reduce inflammation, promotes healing, increases collagen production and improves gingival circulation [95]. Oral vitamin C supplementation as an adjunct to non-surgical treatment of chronic periodontitis has also been shown to be effective against periodontal inflammation [97]. In general, vitamin C appears to have anti-inflammatory properties and therapeutic potential in the treatment of periodontal disease.

2.3. Vitamin D

Vitamin D plays a vital role in immune responses and bone homeostasis [98,99]. Research has suggested that vitamin D deficiency may affect periodontal tissues [[100], [101], [102]]. People receiving regular periodontal treatments along with

vitamin D supplements have shown a better tendency to maintain periodontal therapy. It may be due to its indirect role in reducing gingival inflammation [103]. The third National Health and Nutrition Examination Survey (NHANES III), a representative study involving a US population, concluded that optimal vitamin D status reduced gingival inflammation [104]. The same survey found a link between the high prevalence of periodontitis and vitamin D deficiencies [105]. A case-control study found that vitamin D inadequacy is associated with periodontal disease in women during pregnancy [106]. Moreover, the results of a longitudinal study in older men suggested that vitamin D intake protects against the progression of periodontal disease [107]. A systematic analysis found that, although the literature has been mixed and limited, some data support a protective role for vitamin D in periodontal disease [108]. It is possible that this impact on periodontal health is due to its anti-inflammatory effects [[109], [110], [111]].

Epidemiological data from a Danish population have suggested that the consumption of dairy products containing vitamin D, along with calcium, casein and whey protein, have a protective effect against periodontitis. However, these studies lacked information on dietary supplements [112, 113]. Therefore, it could not be determined whether vitamin D, other dairy components or the movement of other less healthy foods by dairy products were responsible for the effect. Therefore, further studies are needed to confirm the protective effects of vitamin D in periodontitis.

2.3.1. Vitamin E

Vitamin E is a fat-soluble vitamin in various forms, mainly tocopherols and tocotrienols [114]. The main food sources include nuts and vegetable oils [115]. One of the roles of nutrients is the elimination of peroxyl free radicals, helping to stabilize membrane structures by inhibiting the reaction of free radicals [56,116]. This function of vitamin E is relevant for periodontal diseases that are characterized by oxidative stress. A cross-sectional study of an adult US population showed an inverse relationship between serum tocopherol levels and the severity of periodontitis [65]. In addition to eliminating free radicals, research has suggested that the beneficial role of vitamin E in periodontal inflammation may be due to its intrusion into prostaglandin synthesis [117]. Importantly, several studies in rats have shown that a diet rich in vitamin E reduces periodontal and gingival inflammation and improves wound healing [[118], [119], [120]]. These studies highlight the role of vitamin E in periodontal disease and indicate that dietary sources may be an area for future human research.

2.3.2. Carotenoids and flavonoids

Carotenoids and flavonoids such as β-carotene, lycopene and polyphenols are antioxidants usually obtained from vegetables and fruits [57, [121], [122], [123]]. Research has shown that these phytochemicals are effective in the prevention and treatment of periodontal disease. A recent review of antioxidant nutrients suggests that increased intake of flavonoids and carotenoids may be beneficial in preventing periodontitis [58]. Specifically, vitamin A β-carotene pre-cursor deficiency [124] has been linked to periodontitis and gingivitis [67, 125]. When vitamin A is supplemented with other therapies, studies have shown improved treatment outcomes for periodontal disease [60,126]. Research supports the importance of adequate intake of carotenoids and flavonoids for the prevention and management of periodontal disease.

Carotenoids and flavonoids are well known to have antioxidant properties and to modulate anti-inflammatory processes [116]. For example, lycopene, a bright red carotenoid, has been shown to stimulate the release of an anti-inflammatory cytokine, IL-10, and reduce TNF-α in vitro [127]. This anti-inflammatory effect was demonstrated in a randomized placebo-controlled clinical trial that tested the efficacy of orally administered lycopene (8 mg / day for two weeks) with and without dental prophylaxis [128]. Lycopene alone and in combination with dental prophylaxis significantly decreased gingival inflammation at the end of the study period [128]. Similar results were found for the efficacy of lycopene supplements in mild-to-moderate, but not more severe, gingivitis in another intervention study [129]. While these

intervention studies suggest that lycopene supplementation may be useful for reducing inflammation in periodontal disease, it is important to note that these studies were small (~ 20 participants each). Lycopene administered topically by an orally administered gel has also been shown to reduce markers of oxidative stress and gingival inflammation in periodontal disease [130].

Flavonoids found in green tea, coffee and cocoa can also be beneficial in reducing inflammation associated with periodontal disease. Green tea supplements have successfully reduced inflammation as an adjunct to mechanical periodontal therapy in a human intervention study [131]. In a large prospective cohort study in men, it was found that higher coffee consumption was associated with a small but significant reduction in the number of teeth with periodontal bone loss [132]. Mechanically, this is supported by animal models of experimentally induced periodontitis, where reduced markers of oxidative stress were observed in rats fed a diet rich in cocoa [133]. While these studies indicate a benefit for flavonoid use, further research is needed to form specific recommendations or therapeutic guidelines to be developed.

2.4. Minerals

2.4.1. Zinc

Zinc is the second most abundant trace element in the human body and is involved in the protection of cellular elements against oxidative damage [57,134]. Therefore, zinc can play a key role in maintaining periodontal health. Zinc deficiency can cause gingival inflammation by aggregating IL-1 [135]. A study of 14 Sprague-Dawley rats fed a zinc-deficient diet for four weeks after weaning found an increase in gingival index and plaque and depth of periodontal pockets.

Therefore, dietary zinc deficiency may be a potential risk factor for periodontal disease [136]. Case-control studies and cross-sections also established a link between serum zinc levels and periodontitis in healthy and diabetic patients [[137], [138], [139], [140]]. Maintaining periodontal health in diabetic patients is a crucial consideration, as periodontitis is one of the significant complications of diabetes [139,141,142]. Low plasma zinc levels have also been associated with subsequent deterioration of periodontal disease in patients with type 2 diabetes compared to healthy individuals [143]. These studies confirm that zinc deficiency is not only a risk factor for periodontal disease, but aggravates periodontitis in type 2 diabetes.

2.4.2. Selenium

The association of selenium with periodontal disease has not been as well researched to date. However, selenium is known to be essential for immune responses, and serum levels are inversely related to inflammation and tissue destruction [144]. Selenium-containing glutathione peroxidase acts as a preventive antioxidant by suppressing free radical formation [57]. It has also been reported that lower serum selenium levels may be associated with the severity of periodontal disease [141]. Although studies are limited, maintaining selenium levels in periodontal disease can help manage them.

2.5. Calcium

Calcium is known for its role in promoting bone health [145]. Periodontitis involves the destruction of the alveolar bone [145.146] and therefore calcium may be beneficial in this condition. Emerging evidence suggests that this is the case, showing that calcium status is vital in determining the risk of periodontal disease [[147], [148], [149]]. Data from NHANES III suggested that a lower dietary intake of calcium led to the progression of periodontal disease. Moreover, it has been shown that low calcium intake can indirectly affect periodontal disease; however, further studies are needed to confirm this [150].

Many intervention studies have observed the beneficial effect of combined calcium and vitamin D supplementation on the progression and maintenance of periodontal disease [109,111,151]. A randomized clinical trial involving healthy adults (> 65 years) also indicated that the use of calcium supplements improved periodontal health and tooth retention compared to placebo [110]. However, the study also involved a combination of vitamin D, which can confuse the results.

3. The role of micronutrients in lesions of the oral mucosa

Lesions of the oral mucosa are usually characterized by inflammation of the lips and mucosa and defoliation of the tongue [152]. Inflammatory lesions of the oral mucosa include foot-and-mouth ulcers, glossitis, cheilitis, and stomatitis [153]. These inflammatory lesions usually result from either local trauma or systemic pathologies [154]. Regardless of the origin, all these inflammatory pathologies are characterized by the accumulation of polymorphonuclear leukocytes, which lead to the secretion of specific chemical mediators and are responsible for the immune and inflammatory responses of the host [2,20,155,156]. The most commonly produced chemical mediators include cytokines, chemokines, and lipid mediators [2,155,157]. These inflammatory mediators play a vital role in tissue homeostasis through anabolic and catabolic processes [158]. Although nutritional deficiencies of iron are a predisposing factor in oral mucosal lesions, research focuses on the role of B vitamins, such as folic acid and vitamin B12 [[159], [160], [161]].

3.1. Vitamin B Complex

B-complex vitamins act as co-factors in energy metabolism and are essential for the production of new cells and tissue maintenance during development and healing [71,162]. Thus, the deficiency of these vitamins can lead to disruption of the oral mucosa [71]. The first signs of deficiency of some B vitamins are observed in the oral cavity and can be characterized by specific oral manifestations, as shown in Table 1 [54, [163], [164], [165], [166], [167]].

Deficiency:

Vitamin B1: Cracked lips, angular cheilitis

Vitamin B2, B3 Angular cheilitis, glossitis

Vitamin B6 Burning sensation in the oral cavity related to glossitis and stomatitis and cheilitis

Vitamin B9 Recurrent foot-and-mouth disease (RA)

Vitamin B12 Angular cheilitis, painful ulcers in the oral cavity, glossodynia (tongue pain) and RAS

Some of these oral manifestations, including angular cheilitis, glossitis, stomatitis, diffuse erythematous mucosa, and ulcerative mucosal lesion, are symptoms of early pernicious anemia (B12 deficiency) and megaloblastic anemia (B12 and folic acid deficiency) [168,169]. Vitamin B12 deficiency may be a contributing factor to recurrent aphthous stomatitis (RAS), a chronic inflammatory disease of the oral mucosa characterized by painless and recurrent ulcers, however, the exact etiology is unknown [164]. In general, B vitamins play an essential role in the health of the oral mucosa, and their deficiency contributes to the development of common oral conditions.

4.1. Vitamin E

Vitamin E supplementation may be beneficial in repairing and protecting against pulmonary inflammation. An experimental study in adult male Sprague-Dawley rats showed that vitamin E supplementation, along with dental pulp therapy, can improve pulp healing and repair [178]. It has also been shown that an isomer of vitamin E, α-tocopherol is beneficial for protecting pulp cells against toxicity leading to pulpal inflammation caused by H_2O_2 in vitro [179]. However, research is still limited.

4.2. Copper

Copper is known for its anti-inflammatory function [180,181]. Modified serum copper levels are indicative of chronic inflammatory conditions, such as increases in serum copper associated with periodontitis [138]. A case-control study involving a small Indian population showed a very significant correlation ($p < 0.001$) with copper deficiency in individuals with periapical inflammation compared to healthy individuals [182].

However, further research is needed to confirm the anti-inflammatory properties of copper in periapical inflammation.

5. Interactions between micronutrients and salivary function

Saliva is an important part of the oral environment, salivary gland dysfunction or changes in saliva composition can have implications for oral health [183]. It is well established that vitamin C deficiency influences the function of the salivary gland [184], and iron deficiency generally results in reduced salivary secretion [185]. Studies in rodents

also suggest that calcium and vitamin D deficiency may lead to salivary gland dysfunction [[186], [187], [188]]. This can further exacerbate oral inflammation or disease and further promote malnutrition through a reduced sense of taste and appetite. [189]

2. Materials and Methods

2.1 Materials

Medical consultation records highlighting the damage to the gums and the status of periodontal pockets.
Medical tests records for micronutrient deficiencies in the body (nutrigenetic test-).
Consultation kits
Treatment prescriptions

2.1. Subjects

To find out the micronutrient deficiencies in the DNA, nutrigenetics tests were performed on 9 patients (4 men and 5 women) with gingivitis and moderate periodontitis aged 36-71 years.
All subjects were systemically healthy, without symptoms of infection and did not take antibiotics for at least 3 months before the experiment.
Vitamin therapy has started - almost all vitamins are used: vitamin A, complex B, vit. C, vit. D, vit. E, PP, in the form of multivitamins and Tarosin, which contains vitamins C + P. Vitamins activate capillary circulation, optimize vascular permeability, metabolic exchanges and oxidation-reduction reactions, cell regeneration.
All subjects were informed about the purpose of the study and participated voluntarily.

2.2. Procedure

2.2.1. Performing the oral examination by the dentist with the preparation of the periodontal file in which the status of the gingival and periodontal pockets was noted before treatment (example Fig: 1,2,3)

EVALUATION OF ORAL HYGIENE INDICES

→ Oral hygiene index IHB

Plaque index: *superior arch*

$$X = \frac{\text{No. of surfaces with plaque}}{\text{Total no. of dental surfaces}} \times 100 = \frac{24}{64} \times 100 = 37{,}5\ \%$$

Bacterial plaque in the middle third of the crown. The plaque index is 2.

- inferior arch

$$X = \frac{\text{No. of surfaces with plaque}}{\text{Total no. of dental surfaces}} \times 100 = \frac{24}{64} \times 100 = 37{,}5\ \%$$

Bacterial plaque in the middle third. The plaque index is 2.

Tartar index: *superior arch*

$$X = \frac{\text{No. of surfaces with tartar}}{\text{Total no. of dental surfaces}} \times 100 = \frac{0}{64} \times 100 = 0\ \%$$

The tartar index is 0.

inferior arch

$$X = \frac{\text{No. of surfaces with tartar}}{\text{Total no. of dental surfaces}} \times 100 = \frac{12}{64} \times 100 = 18.75\ \%$$

The tartar index is 1.

IHB Oral Hygiene Index:

superior arch: oral hygiene index: 2

inferior arch: oral hygiene index: 3

Tartar and bacterial plaque in the middle third and package.

Bleeding index: *superior arch*

$$X = \frac{\text{No. of bleeding surfaces}}{\text{Total no. of dental surfaces}} \times 100 = \frac{0}{64} \times 100 = 0\ \%$$

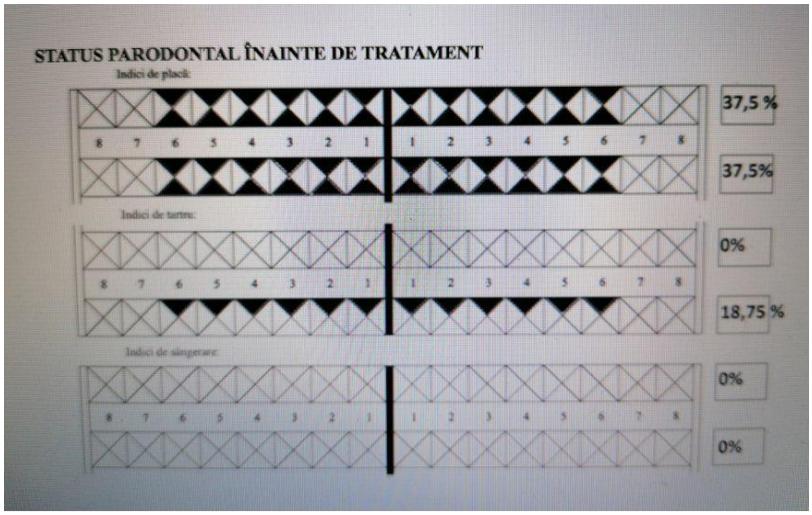
inferior arch

$$X = \frac{\text{No. of bleeding surfaces}}{\text{Total no. of dental surfaces}} \times 100 = \frac{0}{64} \times 100 = 0\ \%$$

Silness and Loe plaque index: 1 plaque is not observed with the naked eye on the dental surfaces until after scraping with the rounded tip of the periodontal probe.
Muhlmenann bleeding index: 0
Gingival retraction:

Tooth	**V (mm)**	**O (mm)**	**M (mm)**	**D (mm)**
2.1	2			
2.2	1			
3.2	1			
3.1	2			
4.1	2			

PERIODONTAL STATUS BEFORE TREATMENT



Conversion formulas (nutrient units)

Nutrient	UI → µg or mg	Equivalents → µg or mg
Vitamin A	1 Ul = 0.3 µg retinol 1 Ul = 0.6 beta-carotene	1 µg RE = 1 µg retinol 1 µg RE = 2 µg beta-carotene (supplements) 1 µg RE = 12 µg beta-carotene (foods) 1 µg RE = 24 µg alpha-carotene 1 µg RE = 24 µg beta-cryptoxanthin
Vitamin E	1 UL = 0.67 mg d-alpha-tocopherol (natural) 1 Ul = 0.9 mg dl-alpha-tocopherol (synthetic)	1 mg Vitamin E (alpha-tocopherol) = 1 mg natural alpha-tocopherol 1 mg Vitamin E (alpha-tocopherol) = 1 mg synthetic alpha-tocopherol
Vitamin D	1 Ul = 0.025 µg	1 Ul = 0.025 µg
Folate		1 µg UEF = 1µg natural folates 1 µg UEF = 0.6 µg folic acid (supplements or folic acid fortified foods)
Niacin		1 mg NE = 1 mg niacinamide 1 mg NE = 1 mg inositol hexanicotinate 1 mg NE = 1 mg niacin 1 mg NE = 60 mg tryptophan

Selenium	µg / day	400	400	400	400
Zinc	mg / day	40	40	40	40
Sodium	g / day	2,3	2,3	2,3	2,3
Chlorine	g / day	3,6	3,6	3,6	3,6

* The upper limit for magnesium is the intake of supplements and drugs and does not reflect the intake of food and water.
ND = not determined

The following tables you have highlighted the deficiencies resulting in the 9 tests of the patients before performing the drug treatments. They were tested only once in order to highlight the deficiencies and to draw up treatment plans with supplements, leaving a new set of tests to be performed after the end of each treatment.

TABLE 1: Patient no1, (66 years, gender M)
Interpretation of the nutrigenetic test.

Patient 1	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1	YES	
Vitamin B2		YES
Vitamin B3		YES
Vitamin B6		YES
Vitamin B9		YES
Vitamin B12		YES
Vitamin C	YES	

TABLE 2: Patient no2, (46 years, gender M)
Interpretation of the nutrigenetic test.

Vitamin D		YES
Vitamin E	YES	
Cooper		YES
Zinc	YES	
Selenium	YES	
Calcium	YES	

Patient 2	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1	YES	
Vitamin B2	YES	
Vitamin B3	YES	
Vitamin B6	YES	
Vitamin B9	YES	
Vitamin B12	YES	
Vitamin C		YES
Vitamin D		YES
Vitamin E	YES	
Cooper	YES	
Zinc	YES	
Selenium	YES	
Calcium		YES

TABLE 3: Patient no3 (41 years, gender M)
Interpretation of the nutrigienic test.

Patient 3	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1		YES
Vitamin B2		YES
Vitamin B3		YES
Vitamin B6		YES
Vitamin B9		YES
Vitamin B12		YES
Vitamin C	YES	
Vitamin D		YES
Vitamin E	YES	
Cooper	YES	
Zinc		YES
Selenium	YES	
Calcium	YES	

TABLE 4: Patient no4, (35 years, gender M)
Interpretation of the nutrigenetic test.

Patient 4	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1	YES	
Vitamin B2	YES	
Vitamin B3	YES	
Vitamin B6	YES	
Vitamin B9	YES	
Vitamin B12	YES	
Vitamin C		YES
Vitamin D		YES
Vitamin E	YES	
Cooper	YES	
Zinc		YES
Selenium	YES	
Calcium		YES

TABLE 5: Patient no5, (35 years,gender F)
Interpretation of the nutrigenetic test.

Patient 5	Normal Value	Deficiency
Vitamin A		YES
Vitamin B1	YES	
Vitamin B2		YES
Vitamin B3		YES
Vitamin B6		YES
Vitamin B9		YES
Vitamin B12	YES	
Vitamin C		YES
Vitamin D		YES
Vitamin E		YES
Cooper	YES	
Zinc	YES	

TABLE 6: Patient no6, (42 years, gender F)
Interpretation of the nutrigenetic test.

Selenium	YES	
Calcium		YES

Patient 6	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1	YES	
Vitamin B2	YES	
Vitamin B3	YES	
Vitamin B6	YES	
Vitamin B9	YES	
Vitamin B12	YES	
Vitamin C	YES	
Vitamin D		YES
Vitamin E	YES	
Cooper	YES	
Zinc	YES	
Selenium	YES	
Calcium		YES

TABLE 7: Patient no7, (58 years, gender F)
Interpretation of the nutrigenetic test.

Patient 7	Normal Value	Deficiency
Vitamin A		YES
Vitamin B1		YES
Vitamin B2		YES
Vitamin B3		YES
Vitamin B6		YES
Vitamin B9		YES
Vitamin B12		YES
Vitamin C	YES	
Vitamin D	YES	
Vitamin E	YES	
Cooper	YES	
Zinc	YES	
Selenium	YES	
Calcium	YES	

TABLE 8: Patient no8, (69 years, gender F)
Interpretation of the nutrigenetic test.

Patient 8	Normal Value	Deficiency
Vitamin A	YES	
Vitamin B1		YES
Vitamin B2		YES
Vitamin B3		YES
Vitamin B6		YES
Vitamin B9		YES
Vitamin B12		YES
Vitamin C	YES	
Vitamin D	YES	
Vitamin E	YES	
Cooper	YES	
Zinc	YES	
Selenium	YES	
Calcium	YES	

TABLE 9: Patient no9, (32 years, gender F)
Interpretation of the nutrigenetic test.

Patient 9	Normal Value	Deficiency
Vitamin A		YES
Vitamin B1	YES	
Vitamin B2	YES	
Vitamin B3	YES	
Vitamin B6	YES	
Vitamin B9	YES	
Vitamin B12	YES	
Vitamin C		YES
Vitamin D		YES
Vitamin E		YES
Cooper		YES
Zinc	YES	
Selenium	YES	
Calcium		YES

From performing the nutrigenetic tests we were able to create some statistics regarding: vitamin deficiency according to age, sex, and age from which it turned out that both sexes have VitD deficiencies but the higher percentage is male (100% male / 60% female). Also from here it appears that both genders have a deficiency in Vit C (50% male / 40% female). The deficiency of Vit B is higher in females than in males (60% females / 50% males). In women, compared to men, we found a deficiency of Vit A, Vit E and Cu, while in men we found a deficiency of Zn that did not occur in women.

TABLE 10 : Deficiencies according to the male gender.

Masculine	Patient 1	Patient 2	Patient 3	Patient 4
Vitamin A				
Vitamin B1	YES		YES	
Vitamin B2	YES		YES	
Vitamin B3	YES		YES	
Vitamin B6	YES		YES	
Vitamin B9	YES		YES	
Vitamin B12	YES		YES	
Vitamin C		YES		YES
Vitamin D	YES	YES	YES	YES
Vitamin E				
Cooper	YES			
Zinc			YES	YES
Selenium				
Calcium		YES		YES

TABLE 11 : Deficiencies depending on the female gender.

Feminine	Patient 5	Patient 6	Patient 7	Patient 8	Patient 9
Vitamin A	YES		YES		YES
Vitamin B1			YES	YES	
Vitamin B2	YES		YES	YES	
Vitamin B3	YES		YES	YES	
Vitamin B6	YES		YES	YES	
Vitamin B9	YES		YES	YES	
Vitamin B12			YES	YES	
Vitamin C	YES				YES
Vitamin D	YES	YES			YES
Vitamin E	YES				YES
Cooper					YES
Zinc					
Selenium					
Calcium	YES	YES			YES

In the following 4 tables, namely Table 12, Table 13, Table 14, Table 15 we highlighted the deficiency by age groups and we found that in the interval of 30-40 years and 40-50 years we have a deficiency of 100% in the case of vitD and also a deficiency of 100% but in the age ranges 50-60 years and 60-70 years we have at Vit B2, Vit B3, VitB6, VitB12

TABLE 12 : The deficiency depending on the age range 30-40 years.

AGE 30-40	Patient 4	Patient 5	Patient 9
Vitamin A		YES	YES

TABLE 13 : The deficiency depending on the range 40-50 years.

Vitamin B1			
Vitamin B2		YES	
Vitamin B3		YES	
Vitamin B6		YES	
Vitamin B9		YES	
Vitamin B12			
Vitamin C	YES	YES	YES
Vitamin D	YES	YES	YES
Vitamin E		YES	YES
Cooper			YES
Zinc	YES		
Selenium			
Calcium	YES	YES	YES

TABLE 14: The deficiency depending on the age range 50-60 years.

AGE 40-50	Patient 2	Patient 3	Patient 6
Vitamin A			
Vitamin B1		YES	
Vitamin B2		YES	
Vitamin B3		YES	
Vitamin B6		YES	
Vitamin B9		YES	
Vitamin B12		YES	
Vitamin C	YES		
Vitamin D	YES	YES	YES
Vitamin E			
Cooper			
Zinc		YES	
Selenium			
Calcium	YES		YES

TABLE 15 : The deficiency depending on the age range 60-70 years.

AGE 50-60	Pacient 7
Vitamin A	YES
Vitamin B1	YES
Vitamin B2	YES
Vitamin B3	YES
Vitamin B6	YES
Vitamin B9	YES
Vitamin B12	YES
Vitamin C	
Vitamin D	
Vitamin E	
Cooper	
Zinc	
Selenium	
Calcium	

AGE 60-70	Patient 1	Patient 8
Vitamin A		
Vitamin B1		YES
Vitamin B2	YES	YES
Vitamin B3	YES	YES
Vitamin B6	YES	YES
Vitamin B9	YES	YES
Vitamin B12	YES	YES
Vitamin C		
Vitamin D	YES	
Vitamin E		
Cooper	YES	
Zinc		
Selenium		
Calcium		

4. Discussions

The symptoms and treatment of oral inflammatory diseases are mainly influenced by the intake or supplementation of micronutrients. Micronutrients are needed to maintain the health of the mucous membranes and oral structures, as they possess anti-oxidant and anti-inflammatory properties. Deficiency of certain micronutrients can modulate the risk of oral inflammatory diseases. Also, the insufficiency of some of the B-complex vitamins helps to detect certain anemias early. Research has also confirmed that the deficiency of certain traces of minerals also aggravates certain oral diseases in the presence of chronic systemic pathologies.

Periodontitis is an inflammatory and progressive disease that affects the connective tissue around the teeth whose role, among other things, is to support the teeth.

It is accompanied by dysbiosis, ie a change in the balance of the oral microbial flora, a disorder of the immune response (chronic inflammation), oxidative

stress, destruction of the periodontium and tooth loss.

Oxidative stress is definitely a disruption of the balance between the production of reactive oxygen species (which are generated by free radicals) and antioxidant defense. Patients with periodontal disease appear to have a higher level of oxidative stress and thus there is a higher production of free radicals as well as a reduced antioxidant capacity. The role of the latter is to neutralize free radicals.

In periodontitis, recent studies have reached the following conclusions:

• oxidative stress leads to the destruction of the periodontium (destruction
bone and adjacent connective tissue);

• There is an association between periodontitis, cardiovascular disease, and diabetes
metabolic syndrome;

• there is a reduction in the body's ability to heal, caused
a significant decrease in the amount of antioxidants in the gingival tissue; therefore, antioxidant therapy should be considered;

• Diet rich in antioxidants is a key factor.

In case of periodontitis, antioxidant therapy can neutralize reactive oxygen species (toxins) and thus, can be used to treat this chronic inflammatory disease that involves the existence of increased oxidative stress.

Supplements of vitamins, antioxidants, minerals, omega 3 and phytonutrients can reduce periodontal inflammation. In addition, bioflavonoids have antimicrobial and anti-inflammatory activities.

In addition, nutritional therapy should be considered as a new adjuvant treatment for both the regeneration of damaged periodontal tissue and the reduction of gingival inflammation.

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