Chapter

Use of Biomarkers for the Diagnosis of Periodontitis

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Abstract

Periodontal disease is the most common oral condition of human population; if periodontitis is not treated in its initial stages, it can cause the loss of teeth. The diagnosis of periodontitis is based on clinical measurements. However, currently with the advancement of technology, other diagnostic and monitoring options are being search. In fact, different types of biomarkers have been evaluated where different biological fluids have been used as a source of the sample. We will try to summarize existing biomarkers of different periodontitis stages and make a comparison of the periodontal biomarkers evaluated so far and their usefulness in diagnosis and monitoring of periodontitis.

Keywords: biomarkers, periodontitis, dentobacterial plaque, gingivitis

1. Introduction

Health in general is fundamental in humans, oral health plays an important role, and any alteration in it can influence the general welfare of individuals. Diseases of the oral cavity are very important due to its high incidence and prevalence according to the World Health Organization [1].

Regarding the epidemiology of the disease, we can say that from 5 to 15% of the population of the United States suffers from severe periodontitis [2]. Data from the Department of Health of Mexico mention that approximately 8.8% of the Mexican population has chronic periodontitis. This is more common among subjects 35 years of age and older, where it is estimated that the frequency is 22% [3].

Periodontitis is a chronic inflammatory disease that compromises the integrity of the tissues that support the teeth, which include the gingiva, periodontal ligament, dental cement, and alveolar bone, and are collectively known as the periodontium [4, 5].

This disease is caused by specific microorganisms or groups of specific microorganisms, which in the end produce a greater formation of probing depth, recession, or both. When these conditions remain, they cause the tissue to be destroyed and the tooth to be lost. This disturbs the mastication, phonation, and esthetics of the patient, which affect the quality of life [4].

The traditional treatment for periodontitis decreases the microbial presence by means of the mechanical interruption and the elimination of the bacterial layers that form in the surfaces of the teeth and adjacent soft tissues [2].

In the pathology of periodontitis, the clinical, radiographic, and histological characteristics of the gingival groove and pouch epithelium, the underlying connective tissue and the types of resident and infiltrating blood cells in the initial, early, established periodontal injury are known [6].

Currently, the diagnosis requires rapidity, sensitivity, and specificity since determining the stage in which the patient is located is fundamental for a good treatment; for this reason molecules are currently being sought that vary when the person is healthy and when the person has the disease. However, despite all the researches that exist regarding chronic inflammation, the diagnosis of periodontitis is based on clinical measurements; these not only show low sensitivity and specificity as diagnostic tests but are also subjective and laborious. The objective of this chapter is to try to describe what a biomarker is, the types of biomarkers evaluated in periodontitis, the sources to obtain these biomarkers, and their usefulness.

2. Biomarkers

The definition of biomarkers as established by the National Institute of Health (NIH) is as follows: biomarkers are the biological, biochemical, anthropometric, physiological, etc. characteristics, which are objectively measurable, capable of identifying physiological or pathological processes, or a pharmacological response or a therapeutic intervention [7].

There are different types of biomarkers; the ideal biomarker must be specific, sensitive, predictive, rapid, economical, noninvasive, and stable in vivo and in vitro. Additionally, it must have enough preclinical and clinical relevance to modify decisions regarding the pathological process in which applies [7].

Before a biological marker is used in human health studies, its validation is fundamental; therefore, the selection and approval process requires careful consideration of specificity and sensitivity, establishing accuracy, precision, quality assurance, analytical procedure, and interpretation of measurement data, which must be compared with other variables [8].

3. Biomarkers for the diagnosis of periodontitis

3.1 Importance of the different biomarkers used for the diagnosis of periodontitis

Because there are certain molecules, like trace elements, proteins (cytokines), and proteolytic enzymes, these have been considered as possible biomarkers of periodontal disease, we will try to discuss the relevance of these groups below.

3.1.1 Proteins (cytokines) involved in the inflammatory process of periodontitis

Inflammation has evolved as a protective response to an injury, is a primordial response that eliminates or neutralizes foreign organisms or materials, in general; the innate inflammatory response starts in minutes and, if all is well, resolves in a matter of hours. In contrast, chronic inflammation persists for weeks, months, or even years [9]. The inflammatory response that occurs in periodontal disease is mediated mainly by B and T lymphocytes, neutrophils, and monocytes/macrophages. These are activated to produce inflammatory mediators, including cytokines and chemokines [10]. Several pro-inflammatory cytokines including interleukins like IL-1, IL-6, IL-12, IL-17, IL-18, and IL-21; tumor necrosis factor alpha (TNFα); and interferon (IFN-γ) have been demonstrated to be involved in the pathogenesis of periodontitis [2].

3.1.2 Metalloproteinases

There is significant evidence showing that collagenases, along with other matrix metalloproteinases, play an important role in periodontal tissue destruction. The main group of enzymes responsible for the collagen and other protein degradation in extracellular matrix (ECM) is matrix metalloproteinases (MMPs) [11]. Several works have shown that matrix metalloproteinases are upregulated in periodontal inflammation; transcription of matrix metalloproteinase genes is very low in healthy periodontal tissue. In periodontal disease, secretion of specific matrix metalloproteinases is stimulated or downregulated by various cytokines [12].

3.1.3 Calcium

The importance of calcium in the development of periodontal disease has been recognized since the 1980s [13]. In addition to this, a relationship has been found between people who suffer from periodontitis and who also have osteoporosis [14].

3.1.4 Alkaline phosphate

Alkaline phosphatase (ALP) is an intracellular enzyme. It is considered that when this enzyme increases in saliva, it can be determined that there is inflammation and destruction of healthy tissues [15]. It is worth mentioning that other enzymes representative of tissue degradation are aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transferase (GGT), alkaline phosphatase (ALP), and acid phosphatase (ACP) [16].

3.1.5 Phosphate

Phosphorus is an essential element and plays an important role in multiple biological processes, due to the fact that maintaining physiological phosphate balance is of crucial biological importance for bone health [17]. Approximately 85% of phosphorus is in the bone, primarily compounded with calcium (Ca^{2+}), the most abundant mineral in hydroxyapatite (HAP) crystals deposited on the collagen matrix [18].

The importance of phosphate in periodontal disease has been observed in X-linked hypophosphatemia (XLH); this disease is a rare skeletal genetic illness in which increased phosphate in the kidney produces hypophosphatemia and prevents normal mineralization of the bone and bone dentine. In a study of 2017, it was observed that the frequency and severity of periodontitis increased in adults with XLH and that the severity varied according to the treatment of hypophosphatemia. Patients who benefited from early and continuous phosphate supplementation during childhood had less loss of periodontal attachment than patients with late or incomplete supplementation [19].

3.1.6 pH

Although the pH of the oral cavity is between 5 and 9, it is also known to vary widely depending on several factors. There are studies that report that there is a statistically significant correlation between pH and periodontal pocket formation [20].

3.1.7 Oxidative stress

Periodontitis is an inflammatory disease of the supporting tissues of the teeth, it is defined as a complex infectious disease that results from the interaction of the

bacterial infection and the response of the host to the bacterial challenge, and the disease is modified by environmental factors, acquired risk factors, and genetic susceptibility [21]. In recent years, the inflammatory response has been associated with oxidative stress, specifically with reactive oxygen species since it is considered to play a central role in the progression of many inflammatory diseases [22].

Oxidative stress create multiple products in affected tissues, such as reactive oxygen species which are free radicals and other non-radical derivatives which are involved in normal cell metabolism [23], other metabolites can damage DNA such as 8-hydroxy2′-deoxyguanosine (8-OHdG) or 8-oxo-7,8-dihydro-2′-deoxyguanosine (8-oxodG) which are two of the predominant forms of free radicals induced by oxidative lesions. In fact, 8-oxodG has been widely used as a biomarker for oxidative stress [24].

3.1.8 Telopeptide

Bone resorption is a basic physiological process that is central to the understanding of many key pathologies, with its most common oral manifestation seen as the alveolar bone destruction in periodontitis [25]. The osteoid matrix consists principally of collagen (90%), other smaller proteins, and proteoglycans. The main structural protein of the bone is type I collagen. Consequently, most available bone resorption markers are based on degradation products of type I collagen. According to Koizumi et al. [26], ICTP (telopeptide) is one of the best markers for clinical use.

3.1.9 miRNAs (microRNAs)

Nowadays, RNAs that do not code for protein have taken on great importance because, in addition to maintaining their importance in the determination of cellular phenotypes [27], now they are recognized as dynamic participants in the performance of cellular activities [28].

It has been mentioned and demonstrated that miRNAs are involved in bone metabolism, in fact, some studies have shown that they are associated with the activator of the nuclear factor receptor kappa-B ligand (RANKL) induced osteoclastogenesis. Within these miRNAs, miR-223 [29] was the first to associate with periodontal tissue, although other miRNAs such as miR-15a, miR-29b, miR-125a, miR-146a, miR148 / 148a, miR-223 and miR-92 have been identified more recently as important in periodontal health and have even been considered potential biomarkers [30].

3.1.10 Other markers of periodontal disease

Other biomarkers have been analyzed to determine periodontitis. One of these is chondroitin sulfate, which is a natural glycosaminoglycan (GAG) present in the extracellular matrix [31]; chondroitin sulfate is recognized for its immunomodulatory effects, such as the reduction of nuclear translocation NF-κB, the decrease in the production of pro-inflammatory cytokines interleukin-1β (IL-1β) and tumor necrosis factor alpha (TNF-α), and the decrease in expression and activity of nitric oxide synthase-2 (NOS-2) and cyclooxygenase-2 (COX-2) [32]. Another molecule that has been proposed as a possible biomarker is MUC-4. Mucins are high-molecular-weight glycoproteins, are involved in diverse biological functions, are members of transmembrane mucin family, and are expressed in airway epithelial cells and body fluids like saliva, tear film, ear fluid, and breast milk [33]. It has been reported that the production of MUC-4 could be regulated by inflammatory cytokines [34].

3.2 Biomarkers determined in saliva

Saliva is a seromucous secretion, consisting of 99% water; however, saliva is also composed of glycoproteins, phosphate ions, bicarbonate, sodium, chlorine, fluorine, calcium, and potassium and has a neutral pH [35], which forms a film of liquid consistency that covers the surface of the oral mucosa, with the purpose of lubricating it and keeping it moist among many other characteristics for the maintenance of oral health [36]. The composition of saliva varies from one place to another in the oral cavity of each individual [35]. If there are changes in its composition, there may be significant alterations in deterioration of the health of the host [37].

Due to the described characteristics of saliva, several authors have claimed that these salivary constituents may actually be useful indicators of both local and systemic disorders. These revelations have formed the basis of the field of saliva diagnosis and, therefore, have triggered research that culminated in the identification of saliva-based biomarkers for disorders ranging from cancer to infectious diseases [38]. In addition to the above, saliva has several advantages when compared to other sources for diagnosing diseases since saliva is easily collected and stored and ideal for early detection of disease as it contains specific soluble biological markers [39]. Saliva has been used to diagnose diseases as diverse as autoimmune disorders, cardiovascular diseases, diabetes, HIV, oral cancer, and oral diseases [40].

3.2.1 Proteins (cytokines) determined in saliva that could be used as biomarkers

In our review we observed that there are about 15 works that were dedicated to investigate the possible use of these proteins determined in saliva as biomarkers to determine periodontitis. We can say that of all the works, the majority focuses on comparing healthy groups with periodontitis; only three researches include the group of gingivitis, which indicates that this group should be used more for this type of studies. We need to remember that gingivitis is considered an intermediate stage that may or may not lead to periodontitis [41], and if the patient performs good dental hygiene in combination with the treatment, in general, progression to periodontitis can also be stopped [42].

On the other hand, the cytokine that has been most explored and that better results have given as biomarkers to detect in saliva is the cytokine IL-1β [43–48]; this must be due to the recognized importance of interleukin-1β, as an important mediator in the pathophysiology of periodontitis [49].

However, other cytokines such as IL-6 and IL-2 have also been explored [43, 44, 48, 50]; IL-6 is recognized for playing a role as a pro-inflammatory cytokine acting on bone resorption in the presence of infections [51]. Regarding IL-2, a study that investigated cytokine profiles at different stages of the development of periodontitis found that levels of mRNA for IL-2 were significantly associated with the phase of resolution of the disease [52]. This agrees with reports that IL-2 has been implicated in the stimulation of osteoclasts [53].

Regarding MCP-1, Gupta et al. [54] conducted a study in 45 patients with an average age of 43 years for healthy patients and 41 for patients with periodontitis, with results similar to Nisha et al. [55]. In both studies it was found that the levels of MCP-1 in saliva can be a good biomarker for the development of periodontal disease. One difference between the studies is that Nisha's work included a group of patients with gingivitis, while Gupta's study does not include it.

Regarding the possibility of using prostaglandin E2 (PGE2) as biomarkers in saliva to diagnose gingivitis, Syndergaard et al. [56] conducted a study with 80 participants, 40 without gingivitis and 40 with gingivitis, and found that the levels of PGE2 in the group with gingivitis were significantly higher compared with the

control group. This study reported that PGE2 remained high after prophylaxis. As for other studies conducted with the purpose of comparing the concentrations of PGE2, Sanchez et al. [82] conducted a study in which the population was 74 adult subjects who were grouped according to the progress of the periodontal disease in mild, moderate, and severe; the conclusion was that the levels of PGE2 increase as the severity of the periodontal disease progressed. In addition a high sensitivity and specificity were reported.

When TNF-α is evaluated as a possible biomarker for the diagnosis of periodontitis, we found that there are discrepancies since in some studies, such as Eivazi et al. [57] who conducted a study with one healthy group and another with chronic periodontitis which reported that before and after starting treatment, the concentrations of TNF-α in saliva were higher in the healthy group than in the periodontitis group. In contrary to the results reported by Yue et al. [48], they found that TNF-α concentrations were higher in the saliva of patients with advanced periodontitis than in the saliva of healthy subjects. Yue's study is supported by studies that investigate the loss of the alveolar bone since the concentration of TNF-α in subjects with alveolar resorption is low [58].

Yue et al. [48] found that in the saliva of patients with aggressive periodontitis (AP) have higher levels of IFN-γ in the saliva compared to subjects without AP; this decrease was statistically significant throughout the course of treatment ($p < 0.05$).

3.2.2 Metalloproteinases

Matrix metalloproteinases (MMPs) are key proteases involved in destructive periodontal diseases. A total of 23 MMPs have been described. These MMPs can be found in periodontal tissues as pro-forms, active forms, complex species, fragmented, and cell-bound species [59]. MMPs are the most important group of proteinases responsible for the degradation of extracellular matrix proteins during periodontitis, and any imbalance between MMPs and their inhibitors can trigger the degradation of the ECM, the basement membrane, and the alveolar bone [60].

In this way and due to the importance of MMPs, several researchers have been dedicated to try to determine if MMPs are opportune as biomarkers. Gursoy et al. [61] did a study with the objective of detecting possible markers of periodontitis in saliva, with high sensitivity and specificity; to determine this, the salivary concentrations of MMP-8, MMP-9, and MMP-13 among others were measured in 230 subjects. The concentrations of MMP-8, MMP-9, and MMP-13 in saliva were higher in subjects with generalized periodontitis than in controls; however, according to the authors, MMP-8 was the only marker capable of differentiating subjects with severe bone loss of those who presented mild bone loss, so they consider that MMP-8 is a strong candidate to detect alveolar bone destruction [61]. These results were corroborated by Rathnayake et al. [46] who found that MMP-8 could be used as a marker of periodontal disease in large patient populations. An interesting fact that they reported is that smokers compared to non-smokers showed slightly lower concentrations of MMP-8.

Another interesting fact regarding MMP-8 by Ebersole et al. [25] in a study that included 30 healthy volunteers and 50 patients diagnosed with chronic periodontitis is that MMP-8 (among others) was investigated as a biomarker associated with inflammatory and destructive processes of periodontal disease and reported that the levels of MMP-8 of patients who have periodontitis are very different from the normal levels found in healthy subjects and showed a particular diagnostic potential.

Morelli et al. [50] examined 168 participants and found higher salivary levels of matrix metalloproteinases, MMP-3, MMP-8, and MMP-9, in diseased groups compared to healthy. In the same year, Miricescu et al. [62] carried out a study where 20

patients were also included with chronic periodontitis and 20 controls and different biomarkers were evaluated including matrix MMP-8, and as a result it was found that the levels of MMP-8 were significantly increased in patients with chronic periodontitis compared to controls.

Ebersole et al. [43] conducted a study that included 65 healthy subjects, 43 subjects with gingivitis, and 101 subjects with periodontitis. In this study, the levels of MMP-8 very similar to the previous studies stood out in a significant way in the group of periodontitis compared to those of gingivitis and healthy subjects. In a more categorical way, Borujeni et al. [63] reported that MMP-8 provides a substantial sensitivity with which physicians can use the test for MMP-8 and thus detect periodontitis in their patients.

Similarly, Gupta et al. [64] made an investigation with the objective of establishing MMP-8 as a noninvasive marker for the early diagnosis of chronic periodontitis. The study included 40 subjects who were divided into two groups: 20 healthy subjects and 20 patients with chronic periodontitis. The results of this study demonstrate high concentrations of MMP-8 in individuals with chronic periodontitis.

Already Lira et al. [65] continued to explore the importance of MMP-8 and conducted a study that aimed to evaluate the levels of markers related to innate immunity, the MMP-8 in the saliva from patients with aggressive generalized periodontitis, and patients with gingivitis and healthy. In the saliva, MMP-8 levels were higher in aggressive periodontitis than in healthy patients; in this way it is reaffirmed that MMP-8 can be an important biomarker of periodontitis.

Other researchers such as Virtanen et al. [66] continued to look for other metalloproteinases as potential biomarkers, and some reaffirm that the salivary concentrations of matrix metalloproteinases such as MMP-8 and MMP-9 are slightly higher in patients with periodontitis, although they report that the differences between the groups were not significant. Interestingly, this group reports that MMP-13 values were significantly higher in the group without periodontitis compared to patients with periodontitis and also report that the concentration of MMP-13 may have some gender implications in periodontitis.

Following with the MMP-8, Mauramo et al. [67] studied whether the levels of MMP-8 in the saliva are associated with periodontitis in 258 subjects. Periodontitis was more frequent among subjects with high levels of MMP-8 in the saliva. The highest levels of salivary MMP-8 were associated with any periodontal diagnosis (mild, moderate, or severe). They concluded that elevated levels of MMP-8 in the saliva are associated with periodontitis in a normal adult population.

3.2.3 Calcium

When we search for studies that have explored the detection of calcium present in saliva as a biomarker, we find that while some studies report the usefulness of calcium because the subjects in the periodontitis group had significantly higher levels of salivary calcium than gingivitis and healthy group [68, 69], another work find that high salivary calcium content can be correlated with good dental health but not with periodontal bone destruction [70].

3.2.4 Phosphorus

According to Patel et al. [69] study, phosphorus can be considered a biomarker for the diagnosis of sick and healthy periodontal tissues. The study concludes that as the severity of periodontal disease increases, it also increases total phosphorus levels.

3.2.5 Alkaline phosphatase

Alkaline phosphatase has been evaluated in saliva as a possible biomarker for the detection of periodontitis. Dabra and Singh [16] first study 20 healthy subjects with gingivitis, and 20 with chronic periodontitis were included. This investigation showed a statistically significant increase in alkaline phosphatase activities in the saliva of patients with periodontal disease compared to the control group. A recent study of Patel et al. [69] included 150 healthy subjects, 50 patients with chronic generalized gingivitis, and 50 with periodontitis. In this study it was shown that alkaline phosphatase can be considered for the diagnosis of diseased and healthy periodontal tissues; since as the severity of periodontal disease increases, it also increases alkaline phosphatase levels.

3.2.6 pH

The pH of the saliva has been evaluated, and it has been found that there is a significant change in the pH depending on the severity of the periodontal condition, so the pH can be useful as a rapid diagnostic biomarker in the consultation. The study suggests that the pH becomes alkaline when patients have chronic gingivitis, but it becomes acidic when there is periodontitis [71].

3.2.7 Telopeptide (ICTP)

A 2015 study found that the concentrations of ICTP were higher in the group with periodontitis and lower in the group with healthy patients; this study suggests that the level of ICTP in saliva increases as the patient presents with gingivitis and periodontitis, since the periodontitis samples had the maximum concentration of salivary ICTP. The authors suggest that more studies with a larger sample size be conducted to establish a correlation between the concentrations of ICTP and the individual clinical parameters [72].

3.3 Biomarkers determined in crevicular fluid

In the oral cavity, we find three fluids: the gingival crevicular fluid, the serum, and the total saliva. The gingival crevicular fluid is an exudate, and at present the quantification of its constituents is a current method to identify specific biomarkers with a reasonable sensitivity [73].

The gingival crevicular fluid is an exudate secreted by the gums that can be found in the crevices located at the point where the gumline meets the teeth. The concentrations of this fluid are usually low but may increase when an inflammatory process occurs in the oral cavity [74].

It is considered that due to the noninvasive and simple nature of its collection, the analysis crevicular fluid can be beneficial in determining the periodontal status [75].

3.3.1 Proteins (cytokines) involved in the inflammatory process of periodontitis

Regarding the cytokines that have been evaluated, we can say that they are very similar to those that were evaluated in the saliva; besides that the results are also similar since it has been found that IL-β is the most important cytokine since diverse studies confirm high levels in periodontal disease compared to healthy [48, 76, 77].

In the same way it happens with IL-2 and IL-6, where several studies conclude that both interleukins are important as biomarkers to identify patients with periodontal disease [48, 76, 78].

Regarding MCP-1, the levels of this biomarker were significantly higher in crevicular fluid than healthy subjects ($p < 0.001$) [54].

Regarding IFN-γ and TNF-α, both biomarkers were significantly higher in patients with aggressive periodontitis; this correlates with the findings in saliva where both biomarkers were elevated in sick patients [48].

3.3.2 Metalloproteinases

With respect to metalloproteinases, studies of biomarkers in crevicular fluid have shown that MMP-7 could be useful as a potential new biomarker for periodontitis [79]. Similar to that determined in saliva, MMP-8 is increased in patients with periodontitis and provides a good sensitive measure to establish differences between patients and healthy individuals [77]. It is also useful as a complementary tool in the periodontal diagnosis [67]. Another metalloproteinase that has proved useful is MMP-9. This metalloproteinase correlates with clinical measures and results in good sensitivity to predict the progression of periodontal disease [77].

3.3.3 Alkaline phosphatase

A study was conducted to determine the usefulness of alkaline phosphatase as a biomarker, and this study showed a correlation with the clinical characteristics when mediated in crevicular fluid [80].

3.3.4 miRNAs

According to Mico et al. [81], epigenetic regulation by miRNAs has not yet been studied in periodontal disease using crevicular fluid. They analyzed the possible use as biomarkers of six miRNAs: miR-671, miR-122, miR-1306, miR-27a, miR-223, and miR-1226. Of the six miRNAs analyzed, only miR-1226 can be used as a promising biomarker for periodontal disease since it had statistically significant differences between the healthy group and patients with periodontitis.

3.3.5 Oxidative stress

As previously mentioned, 8-OhdG is a marker of DNA damage and is considered a biomarker to detect oxidative stress [24], that is why it is not surprising that its usefulness as a biomarker of periodontitis was explored. This study was conducted in crevicular fluid, and it could be determined that evaluating this biomarker in crevicular fluid is more effective than in saliva and that it can be useful as a biomarker for determining periodontitis since according to the authors, the severity of the periodontal disease can be revealed [82].

3.3.6 Telopeptide

Telopeptide has also been evaluated as a biomarker in periodontitis in the study by Aruna [83], which suggests that this biomarker could be useful as a specific marker of bone turnover in patients with periodontitis.

3.3.7 Other markers of periodontal disease

Chondroitin sulfate is a biomarker that, due to its results in patients with chronic periodontitis, suggests that it is important in the diagnosis to evaluate the severity

of alveolar destruction [80]. MUC4 protein was measured in crevicular fluid and according to this study can be considered as a new biomarker to rule out patients with periodontitis from healthy ones ($p < 0.01$) [79].

3.3.8 Osteoprotegerin (OPG)

A final marker that we have considered for crevicular fluid is osteoprotegerin; this biomarker was studied by Kinney et al. [77]. They studied healthy patients, with gingivitis and periodontitis, finding that the biomarker OPG was elevated in patients with periodontal disease with a significant difference when compared with healthy patients, so they conclude that this biomarker has a good sensitivity to rule out periodontal disease of gingival health.

3.4 Biomarkers determined in serum

Serum in humans is a matrix commonly used in clinical and biological studies. Many authors recommend using the correct matrix. Both plasma and serum are derived from whole blood that has undergone different biochemical processes after blood extraction. The serum is obtained from the blood that has been clotted [84].

3.4.1 Proteins (cytokines) involved in the inflammatory process of periodontitis

In a study conducted by Nile et al. [85], interleukin IL-17 was identified as a reliable biomarker in 40 patients with chronic periodontitis, later these subjects underwent periodontal therapy, and the values of this interleukin decreased; thus, they consider that IL-17 can be a valuable protein to monitor healing processes after a periodontal intervention.

Another protein related to the immune response MCP-1 was studied by Boström et al. [86]. They studied healthy patients with periodontitis and detected that the MCP-1 protein was increased in the serum and inflamed connective tissue comparing it with healthy patients. In this way, they considered that MCP-1 can help identify patients with periodontitis.

3.4.2 Metalloproteinases

Within the metalloproteinases studied in serum, Lira et al. [65] reported that MMP-8 is elevated in patients with aggressive generalized periodontitis compared with the rest of the patients.

3.4.3 Oxidative stress

Sreeram et al. [87] studied the transpeptidase biomarker (GGT) in healthy subjects and with periodontitis. This biomarker showed elevated levels in patients with periodontitis with respect to healthy. Among the conclusions found in this study was the GGT is useful, economic, and easy to use.

Onder et al [88] studied 4-hydroxynonenal (4-HNE) as a biomarker in serum, concluding that biomarker 4-HNE was at high levels in patients with periodontitis.

In another serum study, other biomarkers of oxidative stress as total antioxidant capacity (TOS) and oxidative stress index (OSI) used in patients with periodontitis and healthy found high levels of TOS and OSI in patients with chronic periodontitis, this suggests that these biomarkers play important roles in periodontitis [89].

3.4.4 Others

Calprotectin was studied by Lira et al. [65]; this biomarker was analyzed in patients with aggressive generalized periodontitis, found elevated levels in these patients compared with healthy and with gingivitis. In addition to the previous study, Nizam et al. [90] studied myeloperoxidase (MOP) and found that it increases in patients with generalized periodontitis compared to healthy ones; nevertheless Meschiari et al. [91] reported similar levels in patients with periodontitis and healthy. This discrepancy between both authors is possible due to the demographic variation and probably the anatomical site where the sample was extracted for analysis.

3.5 Biomarkers determined in plasma

Blood can be a universal reflection of the state or phenotype. Its cellular components are erythrocytes, thrombocytes, and lymphocytes. The liquid portion is called plasma, when all the components are retained. The concentrations of various plasma components are routinely determined in clinical practice [92]. In this way, it is not surprising that biomarkers are sought in plasma.

3.5.1 Proteins (cytokines) involved in the inflammatory process of periodontitis

Among the cytokines evaluated in plasma, we can say that IL-8 and IL-10 were useful to discriminate patients with aggressive periodontitis from healthy ones, and we can add that they were interleukins different from those expressed in the crevicular fluid where IL-2 and IL-6 were relevant [78].

On the other hand, IFN-γ is a biomarker that has been determined in plasma and was significantly high in patients with aggressive periodontitis, this correlates with the findings in crevicular fluid where this biomarker was elevated in sick patients [78].

Regarding MIP-1α, when it was determined in plasma, it was determined that it could be useful to discriminate patients with aggressive periodontitis from healthy ones, since it was found to be elevated [78].

4. Discussion

Periodontitis is one of the most prevalent illnesses in humans [93]. One of the main challenges faced by the periodontics field is to improve the methods for diagnostic and prognostic of periodontitis [94]. Biomarkers, previously described, can be useful in monitoring the current state of the disease, the effectiveness of the treatment, and possibly predict the progression. However, currently the single ideal biomarker displaying high specificity and sensitivity for discriminating and monitoring this disease has not been determined. Thus, the combination of different biomarkers could be more advantageous than single biomarkers [93]. This would provide a more accurate panorama of the state of periodontal disease.

Classic methods for periodontitis diagnosis as the inspection and the palpation by the specialist can be relatively inaccurate. Additionally, the use of periodontal probes, and radiography could only provide information about previous periodontal damage rather than the current state [93]. Thus, biomarkers have been proposed as complementary methods to defeat the mentioned limitations monitoring the clinical response to an intervention and future risk [95].

Desirable's characteristics in test using biomarkers in periodontitis are easy to perform, rapid, and low cost, which could allow clinicians to perform early diagnosis and more effective personalized treatment.

Different factors are involved in the development of periodontitis, and a complex interaction between bacteria and immune system is observed. Additionally, periodontitis has been linked to at least 43 systemic diseases [96]. Thus, it is important to be careful when interpreting the results of biomarker tests because different factors could have a confounding impact on potential biomarkers [97]. Additionally, further large-scale studies are needed to prove specificity and sensitivity of the biomarkers analyzed in periodontitis and for utilization in routine clinical practice in the future.

5. Conclusions

Currently, a number of biomarkers have been sought for the detection of periodontal disease, but so far an ideal biomarker has not been found that helps early detection of the disease; perhaps the combination of several is the most appropriate.

The search for biomarkers continues, we suggest for further studies in search of new biomarkers, it should be consider having a larger sample size, a random source and keep a follow-up.

6. Future recommendations

Periodontal disease is already a very common problem in many countries; due to the above, the monitoring and reduction of the progress of periodontitis through surveillance and health promotion are part of the national health goal of countries like the United States [98].

Due to this, there has been an exhaustive search in recent years of biomarkers obtained from various sources, with saliva being the most used; however, we believe that new studies should include groups of patients with gingivitis on a daily basis, since this is considered an intermediate phase in which the patient can (if he/she carries out good dental hygiene and continues the treatment) stop the development of periodontitis [42], so that limit levels of these biomarkers may be detected.

In addition to this, numerous studies have shown so far that among the best options for biomarkers are proteins such as IL-1β, MMP-8, and ICTP.

From our point of view, we should also include and explore molecules such as miRNAs and other noncoding RNAs such as lncRNA and circRNA, in addition to the classical molecules that are already known to directly participate in the development of inflammatory pathology, since the study of these molecules could yield new perspectives on the development and progression of periodontal disease, which at some point may have important applications as biomarkers with leading activity in the development and manifestation of periodontitis.

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