Immunological aspects of inflammatory periodontal disease (analytical review)

Vladimir Evstropov[1], *Galina* Zelenkova[1,*], *Sergei* Tresnitskii[1], *Anna* Spirina[2], *Pavel* Bykadorov[3], *Mikhael* Yenin[3], *Ekaterina* Danileyko[3], and *Alexey* Ermakov[1]

[1]Don State Technical University, sq. Gagarina, 1, Rostov-on-Don, 344003, Russia
[2]Veterinary clinic "Blizneci-Dentalvet", Aviakonstruktora Milya ul., d. 2 korp. 1, Moscow, 109431, Russia
[3]State Educational Institution of the Lugansk People's Republic Lugansk National Agrarian University, ,the town LNAU, 1, Lugansk, , 91008, Lugansk People's Republic

Abstract. Clinical and immunological parallels in inflammatory periodontal diseases are considered taking into account some features of the functioning of general and local structures of the immune system in periodontitis of varying severity, chronic generalized periodontitis, aggressive periodontitis. In the analysis of immunopathogenesis of inflammatory periodontal diseases, an essential role is given to an imbalance in the immune and cytokine system.

1 Introduction

The term "periodontal disease" unites a wide range of chronic inflammatory conditions of the gums, alveolar bone and periodontal ligaments holding the tooth (in foreign dentistry, there is no concept of "periodontium" – a complex that includes the gum, alveolar bone, periodontal ligaments and tooth root cement. this is denoted by the term "periodontium" and accordingly, the term "periodontal disease" is most accurately translated as "periodontitis").

Periodontitis (from the ancient Greek παρα – "about", ὀδούς – "tooth", Latin *itis* – a suffix indicating the inflammatory nature of the disease), otherwise an inflammatory disease of the periodontal tissues, characterized by progressive destruction of the normal structure of the alveolar process of the upper jaws or (and) the alveolar parts of the lower jaw. Periodontitis is widespread enough, like other periodontal diseases.

Table 1 - Classification of periodontitis according to ICD-10 (1997)

Acute periodontitis (K05.2):	K05.20 - periodontal (periodontal) abscess of gingival origin without fistula;
	K05.21 - periodontal (periodontal) abscess of gingival origin with a fistula.
Chronic periodontitis (KO5.3):	K05.30 - localized;
	K05.31 - generalized;
	K05.32 - chronic pericoronitis;
	K05.33 - thickened follicle (papilla hypertrophy).

* Corresponding author: zelenkovalex@rambler.ru

Severity classification:
light - periodontal pockets no more than 4 mm, destruction of bone tissue of the inter-root septum up to 1/3 of the length of the roots, no pathological mobility;
medium - pockets from 4 to 6 mm, destruction of the bone tissue of the septa by 1/3-1/2 of the root length, pathological mobility of the I-II degree.
severe - the depth of the pockets is more than 6 mm, the destruction of the bone tissue of the septa is more than ½ of the length of the roots, pathological mobility of the II-III degree.

By prevalence:
localized (focal), generalized.

2 Discussion

A number of researchers, who assign a leading role in the etiology of periodontitis to local factors, since the 60s of the XX century, have actively studied the microflora of dental plaque. Substantiation of the hypothesis of an infectious etiology of any diseases is compliance with the fundamental postulates of Koch-Pasteur (1884). For infections associated with the microflora of the oral cavity, the postulates of Koch-Pasteur were modified by Socransky S., and currently contain the following criteria: 1 - the pathogen is associated with the localization of the disease and is absent in healthy areas or in other forms of the disease; 2 - exclusion of the pathogen leads to recovery; 3 - there is an inadequate cellular or humoral immune response to a possible pathogen with a simultaneous normal response to other microorganisms; 4 - experiments on animals can serve as a relative basis for the development of practical recommendations; 5 - confirmation of virulence factors is required.

There are three types of periodontal infection: 1 - endogenous / exogenous infection with bacteria that belong to the normal flora of the skin, nose, oral cavity, intestinal and urogenital tracts (Porphyromonas gingivalis, Bacteroides forsythus, Actinobacillus actinomycetemcomitans); 2 - endogenous / opportunistic infection that occurs in a systemically or locally weakened organism (Prevotella intermedia, Fusobacterium spp., Peptostreptococcus micros, Eicenella corrodens, Treponema denticola, Candida spp.) And 3 - exogenous / superinfection in the cavity, bacteria that are not normally found mouth (Enterobacter, Esherichia coli, Klebsiella, Pseudomonas, Staphilococcus, Candida spp.).

The development of periodontitis is closely related to the microbiota of dental plaque. The microbiota penetrates under the epithelial tissue and further into the stroma of the connective tissue, forming an inflammatory process. The inflammatory process increases the amount of cerebrospinal fluid, which is a good breeding ground for the microbiota of the gingival pockets. The inflammatory process causes a defect in the epithelial cover of the bottom of the SG, contributes to the cutting off of the periodontal ligaments. The growth of the epithelium in depth is limited by the limit of the compact plate of the periodontal fissure. Periodontium replaces granulation tissue, significantly increasing the surface area of tissues infiltrated by plaque microbiota.The main pathogenetic link that transforms the microbiota of dental plaque is overcoming the epithelial cover, the formation of an inflammatory infiltrate into the connective tissue of the periodontium behind the SG junction and penetration into the systemic circulation.

The area of the surfaces of the periodontal teeth is 75 cm^2. Thus, if a patient has atrophy of half (1/2) of the bone of the alveolar process of the jaw, then the wound surface infected with plaque microbiota will be from 30 to 40 cm^2. Such an infected wound area significantly increases the risk of plaque microbiota entering the systemic circulation. Moreover, if with extensive periodontitis pathological gingival pockets reach a depth of 4–5

mm, then in aggregate they represent a chronically infected wound with an area of 10 to 20 cm^2.

The prevalence of periodontal disease in clinical observations shows that the inflammatory process in the oral cavity can lead to prolonged severe bacteremia, which reduces immunity and depletes the immune system.

Microorganisms of the oral microbiota against the background of periodontitis affect health in four ways: bacteremia, systemic dissimation, locally formed inflammatory mediators that provoke an autoimmune response, aspiration of bacterial contents and their ingestion into the respiratory and digestive system.

Modern models of the functioning of the microbiota of the biotope of the oral cavity, digestive, respiratory systems and the skin indicate that local antigens of the microbiota, produced by bacterial plaque on the outer membranes, provide local and systemic immune responses of the body. Local antigens of the oral microbiota are actively involved in the "systemic transmission of information" through a series of nuclear factors - kappa and beta - synthesis and secretion of cytokines and chemokines that regulate the inflammatory process locally and systemically. Most of the periodontal cells, gum tissue fibroblasts secrete prostaglandins, interleukin-1-beta, interleukin-6, interleukin-8, tumor necrosis factor (TNF) and interferon gamma. Mediators affect the inflammatory process both locally and systemically.

The relationship between inflammation of periodontal tissues and the pathophysiology of chronic inflammatory diseases of the body is explained by two mechanisms: the first is associated with the influence of the microbiota of the oral cavity biotope and its metabolic products on the pathogenesis of atherosclerotic plaques in myocardial infarction; the second - with the established influence of inflammatory mediators caused by periodontal pathogens on the pathogenesis of atherosclerosis, diabetes mellitus, which have a multifactorial etiology. These diseases usually arise as a consequence of chronic periodontal disease [1].

Chronic periodontitis can make a substantial role in the violation of human health. Cardoso E.M. and colleagues [2] reports on the relationship of chronic periodontitis with cardiovascular diseases, diabetes, cancer and chronic respiratory diseases, believing that this relationship may be largely due to an increase in inflammatory cytokines in these patients (TNF-alpha, IL -1 and IL-6). The relationship between periodontitis and type 2 diabetes, rheumatoid arthritis, osteoporosis, Parkinson's disease, Alzheimer's disease, psoriasis, is also associated with the presence of common inflammatory pathways for these diseases. It has been shown to be associated with late Alzheimer's disease with clinical signs of periodontitis and the level of circulation of antibodies against bacteria associated with periodontitis [3], which is proved in our opinion, the essential role of the immune system in the occurrence and development of both periodontitis and comorbid diseases.

S. Amar and M. Engelke [4] showed that a significant part of the population (up to 46%) detected periodontal pathogens, but many of them limit the progression of the disease or purify the body in case of infection, which is apparently due with the protective potential of the immune system. M.P. Cullinan et al. offer a multifactorial etiology of periodontal disease, in which the host's immune system and environmental factors play a significant role. Moreover, the characteristics of the individual's immune response to infectious agents largely determine the severity of periodontal disease, and the dysregulation of the mechanisms of innate immunity plays a major role in the development of periodontal disease.

According to modern concepts, periodontitis can be attributed to non-specific infectious diseases initiated by various associations of microbes [5]. Gram-negative anaerobes (Prevotella, Leptotrichia, Veillonella, Porfiromonas, Treponema) are more frequently infected with parodontal [6]. P.gingivalis, T.forsythia, T.denticola, P.intermtdia are usually considered as the most likely causative agents of initiation of chronic periodontitis, while

aggressive periodontitis is more often associated with colonization of the oral cavity A. Actinomycetemcomitans, P. Gingivalis.

When evaluating the contents of periodontal pockets in patients with inflammatory periodontal diseases in cytological preparations, some clinical and immunological parallels were shown [7]. In particular, when studying cytograms of epithelial cells and leukocytes in patients with inflammatory periodontal diseases of various degrees of severity, various changes were recorded correlated with the level of desquamation of the squamous epithelium, its proliferative and functional activity, the formation of periodontal pockets, the state of factors of natural immunity.

There are some clinical and immunological parallels in the study of indicators of innate immunity in patients with periodontitis of varying severity of the disease. Mild periodontitis is characterized by increased expression of the HBD-3 genes in epithelial cells of the periodontal mucosa and the production of cytokine IL-6, which is interpreted as a reaction of innate immunity to the aggression of microbial paradontopathogens. With moderate and severe periodontitis, there is a sharp increase in the expression of TLR4 genes in epithelial cells of the periodontal mucosa and an increase in the cytokine TGFβ-1 content in the gingival fluid, causing periodontal tissue destruction [8].

In patients with chronic generalized periodontitis, both at the local and systemic levels, an imbalance of immune system cells, a decrease in the functional activity of neutrophils, an increase in the content of proinflammatory cytokines and C3, C4 components of complement was noted. The immune mechanisms of this disease reflect the presence of autosensitization and a decrease in non-specific resistance in this disease. In the early stages of the disease, activation of immune mechanisms is noted, in the late stages, their suppression (secondary immunological failure) with marked suppression of regenerative processes. An immunological study of patients with chronic generalized periodontitis of a severe degree reveals a decrease in the level of T3D + T cells, T4D + lymphocytes, an increase in the content of SD8 + lymphocytes in the blood, the TDS4 + / SD8 + index, and a decrease in the level of SD16 + lymphocytes (natural killers) in the blood. The Ig A level is regularly exceeded, circulating immune complexes are detected [9].

Chronic generalized periodontitis is often accompanied by significant contamination of the contents of periodontal pockets by yeast-like fungi of the genus Candida, the intensity of which correlates with the severity of the inflammatory process. Major changes in the functioning of the immune system are manifested by an imbalance of pro (INF-γ TNF-α, IL-8) and anti-inflammatory (IL-4) cytokines in the contents of periodontal pockets and peripheral gum blood, which clearly correlate with the severity of periodontitis [10]. Chronic inflammation is characterized by periodontal infiltration by macrophages producing cytokines TNF and IL-1.6, which increase the activity of osteoclasts, which leads to the destruction of alveolar bone tissue [11].

Immune disorders in aggressive periodontitis are characterized by hyperimmunoglobulinemia M, against the background of hypoimmunoglobulinemia A and G, lymphocytosis, a violation of the CD4 / CD8 index and the functioning of phagocytes [12]. According to data obtained [13], localized cultures of blood cells with subgingival plaque material from patients with aggressive periodontitis demonstrate higher levels of cytokines (G-CSF, INFγ, IL10, IL12p40, IL1β, IL-6, IL-8, MCP-1, MIP-1α and TNFα) than control cultures. In the gingival fluid of patients with aggressive periodontitis, only the level of γ-interferon is reduced [14], and in the blood plasma, the levels of interleukin (IL-10, IL-13, IL-1Ra) decrease, monocytic chemoattractant-protein-1 (MCP-1) regulated on an activated T-cell expressed and secreted (RANTES), granulocyte colony-stimulating factor (G-CSF), granulocyte-macrophage factor-CSF (GM-CSF). The ratio of Th1: Th2 in the fluid of the gingival sulcus and Treg: Th17 in plasma is significantly lower in these patients. The authors of the study concluded that a decrease in the level of cytokines found

in the plasma of patients may affect the immune response, and a decrease in the ratio of Th1:Th2 and cytokines in the gingival fluid suggests an increase in the role of Th2 in the local area of the lesion.

The immune system plays such an essential role in the pathogenesis of periodontitis that one can speak of the immunopathogenesis of periodontitis. Modern ideas about the functioning of the immune system include the characteristics of its broad extra-immune activity, based on the close interaction of the immune system with the nervous and endocrine systems. During inflammation in the periodontal tissues, the function of the immune system extends to the stimulation and regulation of mucosal homeostasis. However, in this situation, the mechanisms of local immunity determine the violation of the integrity of cell membranes, dysfunction of enzyme complexes, as well as damage to the tissues of the oral cavity [15]. As a result of phagocytosis of microorganisms, the complement systems and immune cells are activated, the synthesis of mediators is intensified, and subsequent antigen-specific immune responses are formed [16].

Immunopathogenesis of inflammatory periodontal disease consists of several stages. With the initial response of resident leukocytes and endothelial cells to a bacterial biofilm (stage 1), only histological changes are noted. The metabolic products of bacteria then activate cell-mediated cytokine production and stimulate neurons to produce neuropeptides. Neutrophils from the vessel migrate to the site of inflammation in response to chemokines. The increase in the number of neutrophils in the connective tissue and the appearance of macrophages, lymphocytes, plasma cells and mast cells in it leads to the development of early lesions. Proteins of the complement system are activated. The epithelium is growing, there are clinical signs of inflammation of the gums (stage 2). The next stage is interpreted as an established lesion, which from the standpoint of immunology is considered as the period of transition from the innate immune response to the adaptive one. Immunocytologically, it is characterized by the dominance of macrophages, plasma cells and T lymphocytes, the presence of subclasses (subpopulations) of BG lymphocytes with expressed IgG1 and IgG3. Blood flow is impaired, collagenolytic activity is activated and collagen production by fibroblasts increases. Clinically, this stage is manifested by gingivitis. At the final stage (periodontitis) bone loss occurs, the spread of inflammatory lesions affects the alveolar bone.

The resolution of inflammation in the immunological aspect is characterized by the production of lipid mediators (immunoresources), resolvins, lipoxins and marezins [17]. Resolvins are supposed to prevent neutrophil penetration, limiting inflammation at the local level and promoting tissue regeneration [18], and cytokine imbalance serves as the basis for the development of chronic inflammatory reactions [19].

It is of interest to study the participation of various types of immunity in the development of periodontitis. Grover, H. S. et al. [20] reports that neutrophils, macrophages, dendritic cells, mast cells, neutrophils, natural killer cells, and eosinophils are the cellular basis of innate immunity, including periodontitis. The cellular mechanisms of adaptive immunity are represented by macrophages, T-and B-cells of various subclasses. Clinically, the expression of periodontal disease can occur in the form of a relatively stable or progressive lesion. A stable response is mediated by T-helper cells of the Th1 subclass and predominates in the presence of T-cells, whereas a progressive lesion is formed against the background of the prevalence of B-cells and through the T-helper response of the Th2 subclass.

According to the literature, the structural and functional imbalance between key subpopulations of T-cells can be crucial for the pathogenesis of periodontal disease [21, 22]. In particular, the level of Th17 cells is associated with the activity of periodontal inflammation and tissue destruction [23], whereas Treg cells, on the contrary, play a protective role, participating in protection against the development of periodontitis [24]. As

for B-cells, despite the increase in their number in case of progressive periodontal lesions, the presence of a dual function of B-cells in periodontitis is suggested: protective - by improving bacterial clearance and destructive - by increasing inflammation, bone resorption and matrix dissolution [25]. This is possible due to the fact that B cells produce not only various anti-inflammatory cytokines (IL-10 and TGF-β tumor growth factor), but also pro-inflammatory factors (tumor necrosis factor TNF-α, IL-6, matrix metalloproteinases that contribute to the degradation of the connective fabrics) [26].

It is known that the kappa-B receptor activator (RANKL) and osteoprotegerin are key regulators of bone remodeling and are directly involved in the differentiation, activation and survival of osteoclasts and precursors of osteoclasts [27]. RANKL can express not only osteoblasts and stromal cells, but also cells of the immune system: activated T-cells and B-cells, monocytes / macrophages and dendritic cells. This fact indicates possible functional connections between the functioning of the cells of the bone and immune systems, which can be disrupted during the development of periodontitis.

Authors acknowledge the support of the Government of the Russian Federation (contract No. 075-15-2019-1880).

References

1. O. Shushlyapin, Immunology and Allergology, **3-4**, 15-27 (2010)
2. E. Cardoso, C. Reis, M. Manzanares-Céspedes, J. Postgraduate Medicine, **130(1)**, 98-104 (2018)
3. B. J. Balin, A. P. Hudson, Curr. Allergy Asthma Rep, **14(3)**, 417 (2014)
4. S. Amar, M. Engelke, Mol. Oral Microbiol, **30(3)**, 171-85 (2015)
5. X. Li, X. Wang, M. Zheng, Q.X. Luan, Exp. Cell. Res, **347(1)**, 212-221 (2016)
6. J. Wang, J. Qi, H. Zhao et al., Scientific Reports, **31843**, 168-174 (2013)
7. A. Bulgakova, N. Vasil'eva, Yu. Andreeva, Parodontologiya, **3**, 22-26 (2012)
8. O. Zorina, L. Gankovskaya, R. Balykin, O. Svitich, T. Ivanyushko, Stomatologiya dlya vsekh, **3**, 40-44 (2017)
9. S. Blashkova, N. Makarova, Institut stomatologii, **2(47)**, 54-55 (2010)
10. S. Melekhov, N. Kolesnikova, E. Ovcharenko, Parodontologiya, **1**, 3-9 (2013)
11. T. Murakami, Clin. Calcium, **26(5)**, 766-772 (2016)
12. T. Tarasevich, T. Zakirov, E. Brusnicina, Rossijskij immunologicheskij zhurnal, **3**, 607-609 (2014)
13. L. Shaddox, W. Spencer, I. Velsko et al., J. Clin. Periodontol, **43(9)**, 746-753 (2016)
14. H. Zein Elabdeen et al., Acta Odontologica Scandinavica, **75(8)**, 616-622 (2017)
15. N. Gogebashvili, L. Dzhashi, T. Sanikidze, Allergologiya i immunologiya, **1**, 34-37 (2014)
16. L. Alieva, A. Borisenko, A. Kurchenko, Immunologiya ta allergologiya, nauka i praktika, **3-4**, 76-81 (2010)
17. G. Bannenberg, C. Serhan, Biochim. Biophys. Acta, **1801(12)**, 1260-1273 (2010)
18. H. Hasturk, A. Kantarci, E. Goguet-Surmenian et al., J. Immunol, **179(10)**, 7021-7030 (2007)
19. S. Blashkova, E. Krikun, Parodontologiya, **4**, 25-28 (2017)

20. H. Grover, R. Saini, P. Bhardwaj, A. Bhardwaj, Indian J. Multidiscip. Dent, **5(2)**, 62-67 (2015)
21. N. Huang, F.C. Gibson, Current Oral Health Reports, **1(2)**, 124-132 (2014)
22. M. Akdis, O. Palomares, W. van de Veen, M. van Splunter, C.A. Akdis, J. Allergy, Clin. Immunol, **129(6)**, 1438-49 (2012)
23. M. Adibrad, P. Deyhimi, M. Ganjalikhani Hakemi, P. Behfarnia, M. Shahabuei, L. Rafiee, J. Periodontal Res, **47(4)**, 525-531 (2012)
24. G. P. Garlet, C. R. Cardoso, F. S. Mariano et al., J. Clin. Periodontol, **37(7)**, 591-600 (2010)
25. M. Zouali, Autoimmunity, **50(1)**, 61–70 (2017)
26. Z. Vadasz, T. Haj, A. Kessel, E. Toubi, FEBS Letters, **587(13)**, 2074-2078 (2013)
27. A. Cekici, A. Kantarci, H. Hasturk, T.E. Van Dyke, Periodontol 2000, **64(1)**, 57-80 (2014)
28. A. Zelenkov, A. Ermakov, G. Zelenkova et al., *E3S Web of Conferences*, **135**, 01088 (2019)
29. A. Zelenkov, A. Ermakov, G. Zelenkova et al., *IOP Conf. Series: Earth and Environmental Science*, **403**, 012020 (2019)
30. A. Zelenkov, G. Zelenkova, A. Ermakov et al., *E3S Web of Conferences*, **164**, 06032 (2020)