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Immunological Aspects of the Development of Oral Lesions in Inflammatory Bowel Disease

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Abstract: According to modern concepts, immune disorders play one of central roles in the pathogenesis of IBD. The main immunopathological mechanism is an imbalance between subpopulations of CD4 cells: Th1- and Th2- lymphocytes. According to most authors, CD and UC differ in types cellular immune response. CD is characterized by a cell-mediated reaction of immune damage to the intestinal wall of the Th1 type with a predominance of eat CD4+ - and CD8+ -cells. UC is thought to be a Th2 disease. Type with an increase in the population of B-lymphocytes (CD19 and CD20) and plasma cells resulting from the development of humoral immunity. According to other scientists, in UC there are violations of both humoral, and cell-mediated immunity. Considering different types development of the immune response, immunohistochemical studies of cell composition of lymphocytes are of practical value in the differential diagnosis of CD and UC.

Keywords: CD5 (T-lymphocytes), CD16 (T-lymphocytes, NK-cells), CD57 (NK cells), CD20 (B-lymphocytes), CD31 (vascular endothelium).

Relevance. Immunocompetent cells respond to the processes occurring in the body by changing the degree of expression, the appearance or disappearance of surface or intracellular functional molecules. In the process of immunohistochemical study of COR and intestinal biopsy specimens, the expression of CD5 (T-lymphocytes), CD16 (T-lymphocytes, NK-cells), CD57 (NK cells), CD20 (B-lymphocytes), CD31 (vascular endothelium) was determined.) and tryptase (TK).

Thus, we studied the cells involved in the immune response and representing the system of humoral and cellular immunity: T-lymphocytes, a subpopulation of lymphocytes responsible mainly for the cellular immune response, and B-lymphocytes, a subpopulation of lymphocytes that synthesize antibodies and responsible for the humoral (B-cell) immune response.

It is known that the activation of subpopulations of T-lymphocytes, leading to the formation of inflammatory mediators - eicosanoids, platelet activating factor, histamine, kinins, CK, active oxygen radicals. They play an important role in the progression of inflammation and involvement in pathological the process of new tissue sections.

Immunohistochemical study of markers of immunocompetent cells of the buccal mucosa in patients of the control group (dental patients without IBD) showed a negative expression of CD5, CD16, CD20, weak expression of CD31 and single CD57+ - cells in the stroma and mucus epithelium - stop shell. At the same time, all studied cells were found in both groups of IBD patients.

In terms of expression of CD16, CD31 and CD57, significant differences in CD and UC. The degree of expression of these markers in the oral cavity at CD is much more pronounced than in UC, which indicates a general activation of the T-cell link of the immune system in CD.

Predominance of CD16+ subpopulation found -cells over subpopulation CD5+ - lymphocytes. This may be explained by the fact that CD5+ - and CD16+ -cells in the oral cavity are involved in the immune response process to varying degrees.

Expression of CD31 in the capillary endothelium is more pronounced in CD than in UC, and indicates chronic inflammation of the COR. Moderate expression CD16+ - and CD57+ -cells in CD indicates the activity of inflammation in the cavity mouth. In our opinion, these cells form the composition of the infiltrate determined by morphological studies. In UC, in 100% of cases, a weak degree of expression of CD16 and CD57 was noted.

Analysis of immunohistochemical parameters of the intestinal mucosa in CD and UC showed differences between groups for all markers, especially for area CD16+ - and CD20+ -cells. Area CD16+ - Cells are much more CD (10.15±0.60% versus 3.99±0.10% in UC). Investigation of circulating CD16+ -monocytes in CD, conducted by S. Koch et al., also showed an increased number of CD16+ -cells in the inflammatory infiltrate of the mucosa intestinal lining, especially with high disease activity and involvement in colon process. According to the authors, it is local rather than peripheral CD16+ -cells make up the inflammatory infiltrate in CD and are target for anti-inflammatory therapy.

Area CD20+ -cells are significantly more in UC (11.84±0.36%) than in BC (1.26±0.14%), which is explained by violations of B-cell immunity in UC.

Comparison of the degree of expression of the studied markers in the mucosa cheeks and intestines revealed no statistically significant difference in the expression compared markers in different parts of the digestive tract in CD.

Moderate expression of CD31 in the capillary endothelium indicates an increase in the number of vessels and chronic inflammation in the oral cavity and gut. It is known that CD31 (PECAM-1), being a marker of inflammation, ensures the exit of phagocytes from the vascular bed through the endothelial wall for further migration to the focus of inflammation. Our data are consistent with the results of other researchers who found an increase in CD31 expression in intestinal mucosa and determined that an increase in the area of expression CD31 is accompanied by an increase in the frequency of occurrence of lympho-macrophage infiltrates and does not depend on the number of degranulating MCs in tissues. It is known that participation in inflammation of the vascular endothelium is manifested changes in permeability and diffusion of proteins into the lesion, secretion of endothelial CK, and complement deposition in vascular endothelium.

The T cell population is heterogeneous and consists of various functionally competing (cytotoxic and immunosuppressive) subpopulations.

Increasing the number of CD8+ - and CD57+ -T cells indicate high cytotoxic activity, play a role in the autoimmune response, and are associated with more severe disease. It is known that CD57 is expressed both on NK cells and on T-lymphocytes, activating cytotoxicity and cytokine activity products. Moderate expression of CD16 and CD57 in CD both in the cheek and in the gut confirms the activity of inflammation in the oral cavity and indicates a cell-mediated type of immune response.

According to J. G. De Tena et al., CD57 expression on T cells correlates with clinical activity and laboratory markers of inflammation. at BC. In our observations, a large number of CD16+ - and CD57+ -cells are located in the stroma and intraepithelially. Studies carried out by K. Geboes and et al. showed that the accumulation of cells of the inflammatory infiltrate interepithelially in the walls of the crypts with their subsequent release into the lumen of the crypts,

as a result of leukopedesis or partial destruction of the epithelium, leads to the formation so-called crypt abscesses, which are a characteristic sign of IBD activity.

We did not reveal any relationship between immunohistochemical parameters and localization of lesions in CD (terminal ileitis, colitis, ileocolitis).

In UC, the expression of all immunohistochemical markers in the COR is weakly expressed. Single CD20+ -lymphocytes in the stroma and epithelium of the cheek, as well as a pronounced expression of CD20 in the intestine, indicate a local B-cell immune response in UC. Moderate expression of CD16 and CD57 in the gut stroma confirms the activity of inflammation in the gut.

According to the results of immunohistochemical studies, we have identified the most important indicators that were used as differential diagnostic criteria for CD and UC according to the state of the oral cavity. Created mathematical model including 4 most significant features (area staining and the degree of expression of CD16, the intensity of staining of CD31 and the degree expression of CD57).

Simultaneous immunohistochemical study of markers of lymphocyte subpopulations (CD5, CD16, CD57, CD20, CD31) performed in our work in the SOR and biopsy specimens of the intestinal mucosa obtained in the zone of the greatest bowel lesions in CD and UC have not been previously performed.

In recent studies conducted by B. M. Sinčić et al., established the clinical significance of CD68+ aggregations -macrophages in the SOR in patients with IBD, significantly more often present in the normal mucosa cheeks in CD than in UC, independent of the characteristics of the microbiota of the cavity mouth and not associated with aphthous lesions. There were no statistical differences in the frequency of occurrence of macrophage microaggregates in patients with active CD and in remission of the disease. Similar microaggregates CD68+ -macrophages found in biopsies of normal gastric and duodenal mucosa in CD and not detected in UC. To assess the relationship between the nature of the course of aphthous stomatitis and immunohistochemical parameters, the dependence of the frequency of exacerbations was analyzed stomatitis from the local immunological status of patients.

A direct relationship was found between the expression area of CD16 in the COP and the nature of the course of aphthous stomatitis in CD ($p<0.05$), which allows us to consider CD16 as a predictor of the frequently relapsing course of ASD. The CD16 protein is known to be a marker of natural killer cells (NK cells).

Pronounced expression of CD16 confirms the role of high activity of inflammation in the progression of the disease. Area CD16+ -cells are much more with frequent exacerbations of ASD (3-4 times a year) against the background of CD progression than with long-term remission of ASD ($6.7\pm0.1\%$ versus $1.7\pm1.2\%$). Relationships between immunohistochemical parameters and course aphthous stomatitis was not detected in UC ($p>0.05$).

Thus, the relationship between the frequency of exacerbations of aphthous stomatitis from CD activity and the absence of such in UC, confirming the hypothesis that the pathological conditions of ORS in CD and UC, despite similar clinical manifestations, have different mechanisms that reflect the pathogenesis of the underlying disease.

Previous studies have shown changes in functional activity of TC and indicators of cellular renewal of epithelial cells COR against the background of lesions of the gastrointestinal tract, indicating common mechanisms of development of inflammatory-dystrophic changes in the oral cavity and underlying parts.

The role of TC has been studied mainly in cases of immediate type hypersensitivity; their functional significance in the mechanism of development of chronic inflammation is still not clear. MCs synthesize and secrete a large number of active proteinases, including tryptase, chymase, carboxypeptidase, and cathepsin G, the rapid release of which is induced by numerous factors. To search for additional differential diagnostic criteria for CD and UC, we conducted an immunohistochemical study of TC of the buccal mucosa using anti-tryptase antibodies.

Significant differences in the area and brightness of tryptase expression were revealed. TC of the buccal mucosa in CD and UC. It has been proven that the area of expression tryptase is an important indicator that can be used as a differential diagnostic criterion between CD and UC. Defined threshold value of tryptase expression area, at which the maximum accuracy of the differential diagnosis of CD and UC according to condition of the oral cavity. It was found that the operational characteristics of the immunohistochemical method exceed the indicators of diagnostic efficiency.

We have found a relationship between the area of MC expression and the nature of the COR infiltrate. The change in the functional activity of MC largely determines the degree of inflammatory responses of the SOR against the background of IBD. It is believed that TCs are involved in the regulation of the permeability of blood vessel walls. According to recent studies, MCs may be involved in tissue remodeling during BK. R. Middel et al. proved that chemokines released by MC can selectively attract macrophages and CD4+ -cells and exacerbate the inflammatory response at the site of injury.

Despite the high expression area of TK tryptase in CD, confirming the participation of these cells in inflammatory reactions, were not found associations between tryptase expression and the incidence of ASD exacerbations.

The effect of TA on collagen metabolism in fibrosis is known. We found a correlation between IFD indicating the degree of fibrosis in SF OCT, and the expression area of tryptase TK.

The results of immunohistochemical studies show that CD is characterized by a general activation of the T-cell link of the immune system, a generalized inflammatory process in the gastrointestinal tract with signs of immune inflammation in oral cavity, while in UC the pathological process is limited to the intestine and does not affects the upper part of the digestive system.

The study of CK is considered a promising direction in the study of the pathogenesis of IBD. It is known that CK imbalance plays a significant role in chronicity and progression of diseases of the digestive system. For BC and Yak an increase in the content of key pro- and anti-inflammatory CKs is characteristic.

An increase in the concentration of IL-1β, -2, -6, -8, -12, IF-γ, TNF-α in the early stages and in the height of the disease reflects an increase in adhesive, chemotactic, cytotoxic activity, synthesis of biologically active substances, acute phase proteins, and free radicals. These processes cause disturbance of microcirculation, development of hyperemia, edema, necrobiosis. In later periods, under the influence of the Central Committee, damaged cells are phagocytized, the destructive material is utilized, the processes of regeneration, angiogenesis, restoration of the epithelial layer increase, growth of fibrous tissue.

M. Yu. Ignatov et al. indicate the dominant role of CK in the interaction of immunocompetent cells in the regulation of local cavity immunity mouth. Studies conducted by O. A. Tutina revealed a direct correlation cytokine profiles of the oral fluid and blood serum, which allows the use of saliva as an adequate biological material for assessing the activity of IBD.

Our work established significant differences in the content of IL-6, IL-8, IL-10 in patients with IBD and dental patients without somatic diseases. There is an obvious imbalance in the secretion of pro- and anti-inflammatory CKs (IL-8 and IL-10), as well as in the levels of individual CKs with a similar direction of biological action (IL-6 and IL-8) in patients with CD and UC. According to the content of IL-6 and IL-10 did not reveal any significant differences between CD and UC. The content of IL-8 is significantly higher in the group of patients with UC. A direct correlation has been established between the activity of IL-6 and IL-8 and between IL-6 and IL-10. A direct correlation of CD31 expression with the activity of IL 6 and IL-8 was revealed. It is known that IL-6 and IL-8 are pro-inflammatory CKs, i.e. stimulating and supporting the development of an inflammatory response. IL-6 promotes the synthesis of acute inflammatory proteins, and IL-8 causes chemotaxis neutrophils to the area of inflammation. It is possible that increased synthesis of anti-inflammatory IL-10 is associated with the body's reaction aimed at limiting the excessive production of pro-inflammatory CK.

CK are produced mainly by lymphocytes, macrophages, neutrophils, vascular endothelial cells and, to a lesser extent, epithelial and TK. Since all of these cells make up the inflammatory infiltrate and participate in inflammatory reactions, changes in the content of CK in tissues in IBD is quite natural.

The study of the cytokine profile in ASD also shows an increase in the concentration of proinflammatory CKs. Our data are consistent with the latest studies by E. M. Vakhrushina, demonstrating a high concentration CD8+ - and CD16+ -T-lymphocytes in the zone of inflammatory reaction in the stage of alteration in ASD and confirming the hypothesis of the participation of these cells in the cytotoxic mechanisms of destruction of keratinocytes. At present, there is no doubt about the connection many pathological processes in the oral cavity with diseases of internal organs, metabolic disorders, changes in the immune status.

According to the existing ideas, the change in the functional activity of cells of the immune system COR can be considered as a nonspecific response of the upper digestive system involved in the pathological process. Our data support the hypothesis that

The structural and functional organization of the immune system in various chronic diseases of the digestive system is characterized by a different degree of activation of its components.

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