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Sputum Changes In Children With Obstructive Bronchitis

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Abstract: The purpose of the study: to study the receptor activity of T-lymphocytes in the induced sputum of patients with bronchial asthma and chronic bronchitis in the dynamics of the disease and using various treatment methods. Material for study cell sediment induced sputum. Receptors were determined by immunofluorescence using monoclonal antibodies. The study showed that the content of cells having CD4, CD8, CD25 and HLA-DR receptors in induced sputum is different in patients with asthma and chronic obstructive bronchitis and in both groups depends on the period of the disease and the therapy used. This analysis of induced sputum can be used to monitor the activity of the inflammatory process in the respiratory tract in these diseases and the effectiveness of the therapy.

Key words: sputum, obstructive bronchitis, receptor, bronchitis.

To date, T-lymphocytes are considered as the main link in the progression of bronchial asthma (AD) and chronic obstructive bronchitis (COPD) in adults, both at the local and systemic levels. Moreover, cells that are in a functionally active

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state and carry detectable phenotypic molecules on their surface have pathogenetic significance. So, in patients with AD on the membranes of eosinophils and lymphocytes, the expression of SB113, SB54, SB63, SB25, SB4, and NL-BY receptors increases. With COPD, the number of SB3, SB8, SB68, SB25, and NL-BB lymphocytes increases. In this case, an increase in the number of SB8 lymphocytes is considered a biological marker of COPD. T-lymphocytes play a crucial role in the antigen-associated inflammatory process, as these are the only cells capable of recognizing antigenic material after treatment with antigen-presenting cells. Thus activated SB4 + and SB8 + T-lymphocytes produce a large number of protein mediators, including cytokines, which have the ability to control the differentiation, attraction, accumulation and activation of specific granulocytes on the surface of the mucous membrane.

One of the most valuable diagnostic methods used in the clinic of pulmonary diseases is the study of sputum. In recent years, the method of induced sputum is very common, that is, obtaining sputum after inhalation of a 3-5% hypertonic solution No. C1. The advantage of this method is the simplicity of the procedure, non-invasiveness, safety and high confidence, as well as a low number of side effects.

Objective: to study the state of SB4, SB8, SB25, and HL-BY receptors on lymphocytes of induced sputum (MI) in patients with BA and COPD during various periods of the disease and when using various methods of therapy.

Materials and methods

The study included 45 patients with AD. In the acute stage, 24 patients were examined, of which 13 were of moderate severity, 11 were severe. All patients in this period received glucocorticosteroid therapy (GCS) - budesonide at a dose of 800-1800 mcg / day, or prednisone 20 mg / day. A total of 21 BA patients in remission were examined. Of these, 12 people with moderate severity, 9 people with severe illness. All patients received basic GCS therapy - budesonide at a dose

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of 400-800 mcg / day, or combined therapy with budesonide 600-1000 mcg / day and prednisone 10 mg / day.

Also examined 40 patients with COPD. During the exacerbation of COPD, 26 people were examined, 15 of whom had a moderate course of the disease, 11 severe. All patients in the acute phase received Teopec 300 mg 1-2 times a day.

In the stage of remission of COPD, 14 people were examined, with the same number of patients with moderate and severe severity of the course of the disease. In the period of remission, 9 people received therapy with Teopecom at a dose of 300-600 mg / day.

The control group consisted of 8 healthy non-smoking volunteers without clinical, laboratory, functional and morphological signs of inflammation and obstruction of the bronchi.

The material for the study was a cell precipitate of induced sputum, which was obtained after inhalation of a 3-5% hypertonic solution using an ultrasonic nebulizer. For the dispersion and homogenization of sputum, a dithiotrietol solution was used - a substance with a low redox potential that destroys the disulfide bonds of mucous secretion glycoproteins and does not affect cellular and soluble sputum factors.

Receptors SB4, SB8, SB25 and HLB were determined by the indirect surface immunofluorescence reaction using the Clonospectrum monoclonal antibody kit. Cells were viewed under a luminescent microscope with a magnification of 90 times. Luminous, ring-shaped cells were considered positive. The content of receptors was expressed as a percentage based on 100 cells of the MI sample.

Results and discussion

In healthy individuals, the level of SB antigens is SB4 4.6 \pm 2.1%, SB8 7.2 \pm 1.5%, SB25 3.0 \pm 1.2%, and NL-BY 11.0 \pm 1.3%. With exacerbation of asthma, the activity of all the studied receptors increases. During this period of the disease,

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the number of SB4-positive cells significantly increases, the level of which is 26.4 \pm 2.1% (p <0.05 with respect to the control). The smallest changes affected SB8 cells, their activity increased to $16.5 \pm 1.3\%$ (p <0.05) (Table 1). The development of the allergic component of asthma is associated with the presence of SB4 + subpopulations of T-lymphocytes. In patients with allergies, circulating SB-4 + Tlymphocytes produce a high level of type 2 cytokines, including interleukin-5, interleukin-3 and BM-S8B, and therefore can contribute to eosinophilic inflammation. The use of GCS therapy helps to reduce the number of SB4 cells in myocardial infarction to 12.1 \pm 1.1% (p <0.05), and SB8 to 8.6 \pm 2.1. Not so long ago, O.M. Copy th al. evaluated the role of a single intravenous injection of monoclonal antibodies that bind specifically to SB-4 human antigen in the treatment of severe steroid-dependent asthma. This study showed a significant increase in morning and evening peak expiratory flow in the group with the highest dose. Additional experiments showed that the infusion of keliximab induces a quick and effective connection with all CD-4 + T cells with a transitory decrease in the number of circulating cells of this type, suggesting that therapy aimed at CD-4 T cells may be useful in treatment asthma

In the onset of asthma, we also noted an increase in the content of CD25 and HLA-DR-positive cells to $17.6 \pm 2.7\%$ and $23.5 \pm 2.5\%$, respectively (p <0.05 with respect to the control). As is known, the CD25 antigenic determinant is a receptor for interleukin-2 (IL-2), which is involved in the stimulation of proliferation of T and B lymphocytes, and an increase in its expression during exacerbation of AD is associated with the induction of an immune response. Use in the treatment of corticosteroids in the phase of exacerbation of AD leads to a decrease in the expression of CD25. However, its content remains higher than in the control group and is $13.5 \pm 1.5\%$ (p <0.05). Obviously, a decrease in the representation of CD25 in MI under the influence of GCS is associated with a decrease in the concentration of IL-2. The increase in the content of HLA-DR-positive cells is a consequence of

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the activation of effector cells and their migration into the respiratory tract. The use of GCS therapy helps to reduce HLA-DR receptors in MI, the level of which practically reaches the values of the control group and is

Table 1

The number of CD4, CD8, CD25 and HLA-DR lymphocytes in MI in AD

Patient groups Presence of receptors,%

CD4 CD25 CD8 HLA-DR

AD, remission $5.1 \pm 1.3 \ 2.5 \pm 1.1 \ 6.7 \pm 1.2 \ 4.7 \pm 1.8 *$

AD, exacerbation $26.4 \pm 2.1 * 17.6 \pm 2.7 * 16.5 \pm 1.3 * 23.5 \pm 2.5 *$

BA + GCS 12.1 \pm 1.1 * 13.5 \pm 1.5 * 8.6 \pm 2.1 11.3 \pm 3.0 *

Control $4.6 \pm 2.1 \ 3.0 \pm 1.2 \ 7.2 \pm 1.5 \ 11.0 \pm 1.3$

Note: hereinafter * - the differences are significant at p <0.05 compared with the control.

table 2

The number of CD8, CD4, CD25 and HLA-DR lymphocytes in MI in COPD

Groups Receptor availability,% of patients CD8 CD25 CD4 HLA-DR

COPD, remission $8.1 \pm 1.9 - 4.7 \pm 1.8 *$

COPD, exacerbation $37.6 \pm 2.8 * 0.9 \pm 1.5 * 4.9 \pm 2.2 6.1 \pm 1.4 *$

COPD + Theopec $13.4 \pm 2.1 * 8.5 \pm 3.3 * 5.1 \pm 2.9 * 10.4 \pm 2.3$

Control 7.2 \pm 1.5 3.0 \pm 1.2 4.6 \pm 2.1 11.0 \pm 1.3 11.3 \pm 3.0% f> 0.05).

Decreased expression of HLA-DR cells may be a result of the direct inhibitory effect of GCS on their synthesis.

During AD remission, a decrease in the expression of the presented receptors was noted. The number of CD25-positive cells decreased by almost 8 times compared with the onset of asthma and amounted to $2.5 \pm 1.1\%$, not significantly different from healthy ones. The control values and indicators of CD4 and CD8 antigen were achieved. Their level was $5.1 \pm 1.3\%$ and $6.7 \pm 1.2\%$, respectively.

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The number of cells having the HLA-DR receptor decreased by more than 5 times and became $4.7 \pm 1.8\%$, which is significantly less than in healthy ones (Table 1).

With an exacerbation of COPD, expressed expression occurs with CD8 T-lymphocytes. The number of cells with this phenotype is $37.6 \pm 2.8\%$. The representation of cells carrying CD4 receptors practically does not differ from healthy ones and amounts to $4.9 \pm 2.2\%$. Interesting data were obtained regarding CD25 and HLA-DR receptors. There is a decrease in the content of CD25 and HLA-DR-positive cells to $0.9 \pm 1.5\%$ and $6.1 \pm 1.4\%$, respectively, which significantly differs from the parameters of the control group f <0.05) (Table 2) . One of the reasons for the weakening of the expression of the considered antigenic determinants is the infection process, which often triggers an exacerbation of COB and is responsible for the violation of the immune response at the stage of antigen recognition, which is closely associated with the histocompatibility complex.

The study showed that the content of surface receptors in MI in patients with COB depends on therapy. The use in the treatment of phosphodiesterase inhibitors leads to a decrease in the expression of CD8 T-lymphocytes to $13.4 \pm 2.1\%$. The theophylline's ability to reduce inflammation is achieved by suppressing chemotaxis and proliferation of T-lymphocytes. On the contrary, the number of cells carrying CD25 increases by 8.6 times and exceeds the performance of healthy ones (Table 2). The content of HLA-DR also increases and almost reaches the control values, amounting to $10.4 \pm 2.3\%$. Using theopec in the treatment of acute exacerbation of chronic obstructive pulmonary disease changes the state of the activation signal and shifts its vector towards increased intercellular interactions aimed at resolving inflammation.

During the period of COPD remission, the content of HLA-DR and CD8 antigens sharply decreases to 4.7 ± 1.8 and $8.1 \pm 1.9\%$, respectively, and cells carrying CD4 and CD25 receptors are not detected in MI (Table 2). Obviously,

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this is due to the depression of the immune response in patients with COPD and impaired differentiation and proliferation of T cells.

Thus, both asthma and COPD are characterized by an inflammatory process, but membrane markers of cell activation are different from each other. So, in asthma, the leading cells are CD4 lymphocytes, while in COPD the increase in the number of CD8 lymphocytes is a biological marker.

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