

Evaluating Cytokines and Antimicrobial Peptides as Diagnostic Biomarkers in Colorectal Cancer and Adenomatous Polyps

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Abstract: *This study investigates the role of interleukin 6 IL - 6, tumor necrosis factor TNF - , and fatty acid binding protein FABP as diagnostic biomarkers in patients with colorectal cancer and adenomatous polyps. Analyzing blood serum samples from 80 colorectal cancer patients, 17 adenomatous polyp patients, and 25 healthy individuals, the study explores the diagnostic potential of these biomarkers. The findings indicate significant differences in the levels of these biomarkers among the groups, underscoring their potential in differential diagnosis. Considerable reduction of content of FABP in patients with malignant tumors of bowels on the relative control is also revealed. In patients with adenomatous polyposis, a significant increase in FABP was observed than with bowel tumors. Thus, all these markers can be used for early diagnosis and screening for bowel cancer.*

Keywords: IL - 6 interleukin 6, TNF - tumor necrosis factor, FABP fatty acid binding protein

1. Introduction

The article aims to investigate the diagnostic value of interleukin 6 IL - 6, tumor necrosis factor TNF - , and fatty acid binding protein FABP in differentiating between colorectal cancer and adenomatous polyps.

The article's significance lies in its contribution to understanding the diagnostic potential of specific biomarkers in colorectal cancer and adenomatous polyps, aiding in early detection and effective treatment strategies.

Colorectal cancer is characterized by the difficulties of clinical, radiological and morphological diagnosis. The non-specificity of the clinical symptoms observed during colorectal cancer and the similarity to non-oncological diseases should lead to some diagnostic errors in the early stages of tumor development. Most researchers point out that new endogenous factors, cytokines, biologically active substances such as activators and inhibitors of apoptosis play an important role in the investigation of tumor tissue development mechanisms and metastasizing ability in the pathogenesis of colon cancer. It is considered that the study of interleukins and antimicrobial peptides is important for early diagnosis of the disease and selection of adequate treatment tactics [1].

The rapid and aggressive clinical course of these pathologies, with unsatisfactory results of treatment, requires special attention. The detection of specific lesions mainly in adults reflects the importance of this problem from the social and economic point of view [2].

An important step in the diagnosis of malignant tumors, requires the application of special examination methods that allow the removal of material. However, some of these methods are traumatic, tumor dissemination can be dangerous, and can be accompanied by vascular damage and bleeding [3].

It is known that the tests used in clinical practice are not ideal. It is important to learn the capabilities of each method for the optimal selection of available inspection methods and interpretation of the obtained results. The reliability of the method is characterized by its specificity, sensitivity, as well as accuracy of the results and usability of the research results [4].

Recently, ROC (Receiver Operating Characteristic) research is used to check the effectiveness of laboratory tests. The ROC method is characterized by a trade-off between sensitivity and specificity. ROC allows to objectively assess the diagnostic value of laboratory tests. Through this method, the tests used in the diagnosis of various diseases can also be evaluated comparatively. This method allows determining the individual criteria of the interpreters used in the evaluation of the researched tests [5].

The purpose of the research work the role of cytokines and antimicrobial peptides, which play an important role in the molecular mechanisms of the oncological process, in the diagnosis of colorectal cancer and adenomatous polyp was compared.

2. Research materials and methods

The methods used in the research, including patient selection, clinical - instrumental examinations, and statistical analysis, appear sound and appropriate for the studies objectives. The research includes the materials of 80 patients who were examined and treated with the diagnosis of colorectal cancer at the Oncology Clinic of the Azerbaijan Medical University, 17 patients who were examined and treated with the diagnosis of adenomatosis at the Teaching - Surgical Clinic of AMU, and 25 practically healthy individuals. Patients included in the study were divided into 4 groups. The first group included 26 patients diagnosed with benign colon cancer, the second group included 19

patients with malignant colorectal cancer, the third group included 35 patients diagnosed with metastatic (secondary) colorectal cancer, and the fourth group included 17 patients with adenomatous polyps.

All patients underwent clinical and instrumental examinations, including radiography and ultrasound of the abdominal cavity, small pelvic organs in women, computer and magnetic resonance imaging as prescribed.

The concentration of IL - 6 (interleukin 6) and TNF - α (tumor necrosis factor) in blood serum was carried out by the IFA method using the reagent kit belonging to the "RD" company of the United States. The concentration of FABP (fatty acid binding protein) in blood serum was determined using a reagent kit from Cloud - Clone Corp.

The variations indicated in the groups were sorted and the average indicator (M), standard error (m), minimum (min), maximum (max) values of this indicator were calculated for each variation order.

In this way, the specificity and sensitivity of the biological markers included in the study were studied using the ROC analysis method. Reference indicators are reflected in the ROC curve based on the formula $L(0, 1) = \sqrt{(1 - \text{specificity}, SP)^2 + (1 - \text{sensitivity}, SE)^2}$.

3. Research results and their discussion

The level of IL - 6, TNF - α and FABP in blood serum was determined and the results were compared (table 1).

Table 1: Concentration of IL - 6, TNF - α and FABP in the blood serum of the control group and patients. (M \pm m)

Groups \ Indicators	Year - 6 (pg/ml)	TNF - α	FABP (pg/ml)
Control	1.3 \pm 0.3	1.7 \pm 0.7	13.6 \pm 0.6
Benign bowel cancer	3.1 \pm 0.2**	2.0 \pm 0.5	8.4 \pm 0.8**
Malignant colorectal cancer	13.9 \pm 5.8**	6.1 \pm 1.3*	6.4 \pm 1.2*
Metastatic colorectal cancer	6.5 \pm 0.5 **	12.0 \pm 2.3**	3.3 \pm 0.6 **
Adenomatous polyp	13.6 \pm 2.9**	2.4 \pm 0.8	19.4 \pm 1.8*

Note: statistical integrity of the difference with the indicator of the control group - *p<0.05; **p<0.001

From the obtained results, it is known that IL - 6 in the blood of patients included in the research oliectin is increased most in patients with malignant colorectal cancer and adenomatous polypola. In our entire study, this indicator was 4.5 times higher in the blood of patients with malignant colorectal cancer than in benign tumors, and 2.1 times higher than in patients with metastatic colorectal cancer. In patients with adenomatous polyps, IL - 6 is 4.4 times higher than in benign colorectal cancer. Therefore, if the level of IL - 6 in the blood serum is significantly higher than in the control group, benign colorectal cancer should be ruled out in such patients. Il - 6 activates tumor tissue and accelerates resorption processes [6]. It is known that colorectal cancer,

like adenomatous polyp, develops mainly in children and young people. The initial symptoms of these diseases are almost similar to each other. In this case, the symptoms of malignant colorectal cancer and adenomatous polyp should be examined in detail in specialized institutions and the appropriate diagnosis should be confirmed.

In addition to the accepted standard examination methods, the analysis of IL - 6 in the blood serum informs the doctor that the process has a malignant or benign nature. This helps patients receive timely and adequate treatment.

The statistical indicators of the ROC curve of IL - 6 and TNF - α are shown in Figure 1.

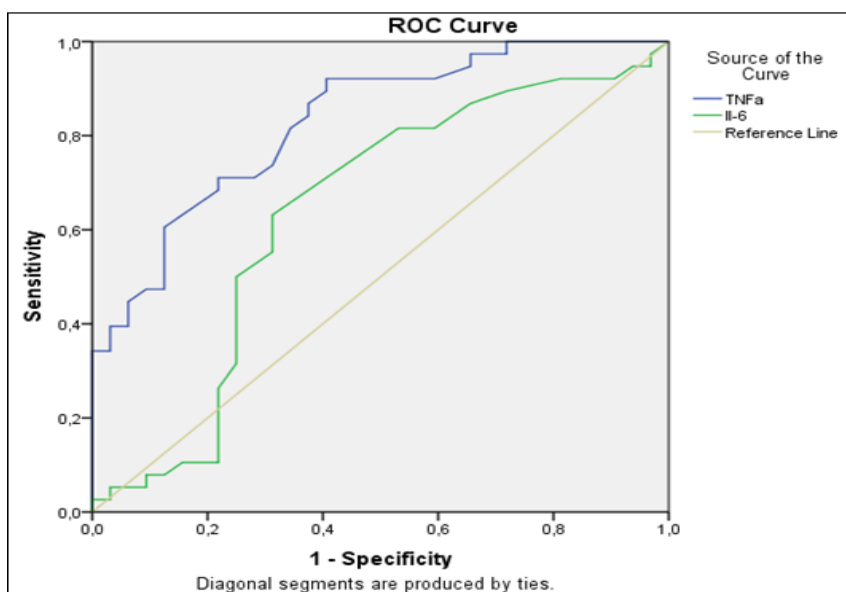


Figure 1: Statistical results of the ROC curve of IL - 6 and TNF - α (ROC curve, specificity, sensitivity, reference line) According to the 95% confidence interval of the ROC curve of IL - 6, the lower limit is 0.737; the upper limit is 0.924 and the area under the curve is 0.831.

Based on the 95% confidence interval of the ROC curve of TNF - α , the lower limit is 0.498; the upper limit is 0.774 and the area under the curve is 0.636.

It is known that in an ideal test, the ROC curve is located in the upper left corner of the graph, when the true positive results are 100%. Accordingly, the lower the curve of the test, the higher the quality of the test. The graph is completed by the straight line $x=y$. So, if the ROC line is below the $y=x$ equation, that test is considered to be of poor quality. As can be seen from the ROC curve of IL - 6, this test is an important screening method with high sensitivity and specificity.

TNF - α is a multifunctional cytokine and is mainly produced by macrophages, but also by neuroglial cells, mast cells, endothelial cells, LAK (lymphokine - activated killer cells) and natural killer cells. TNF - α is almost absent in the blood of a healthy person [7, 8].

The level of TNF - α is higher in the blood of patients with malignant, especially metastatic colorectal cancer. In this regard, the analysis of TNF - α can play an important role in differential diagnosis. In patients with malignant colorectal cancer, the level of this indicator is 3 times higher than in benign colorectal cancer, and 2.5 times higher than in adenomatous polyps. In the case of metastatic colorectal cancer, this difference is characterized by larger numbers, in patients with metastatic colorectal cancer, it is 6 times higher than in patients with benign colorectal cancer, 5 times higher than in patients with adenomatous polyp, and 2 times higher than in patients with malignant colorectal cancer.

Of the patients included in the research contingent, 43 patients had higher than normal levels of TNF - α , and 32 patients did not change. This indicator was not checked in 22 patients. According to the ROC curve obtained on the basis of these figures, it can be noted that considering TNF - α as a test with a high diagnostic value, it can be widely used in the diagnosis and differential diagnosis of colorectal cancer.

We have also obtained results that FABP from antimicrobial peptides plays an important role in the differential diagnosis of colorectal cancer and adenomatous polyp. From our results, it is known that the level of FABP increases compared to the control group only in the case of adenomatous polyp, the level of this indicator was 1.4 times ($p<0.01$) higher in the case of adenomatous polyp compared to the control group. In those with colorectal cancer, the level of FABP decreases compared to the control group. Thus, in patients with benign colorectal cancer, the FABP level decreases by 1.6 times compared to the control group, and by 2.3 times compared to the group of patients with adenomatous polyps. FABP levels in patients with malignant colorectal cancer are 3 times lower than those with polyps, 2.1 times lower than those in the control group, and 1.3 times lower than those in patients with benign colorectal cancer.

Therefore, when it is difficult to distinguish a malignant intestinal tumor from an adenomatous polyp in a patient, performing an examination of FABP in the blood by the

immunoenzyme method can be of important diagnostic importance for differential diagnosis. The fact that the level of this indicator is much lower than the norm can be considered typical for colorectal cancer. FABP is abundant in the liver. However, it is synthesized in the intestinal wall, pancreas, kidney, lung and stomach mucosa. FABP constitutes 5% of the proteins in hepatocytes. In addition to binding fatty acids, this protein also binds coenzyme A, alcohols, and carcinogens [9]. According to the results of the study, the level of FABP in metastatic colorectal cancer is 5.8 times lower than in patients with polyps, 4.1 times lower than in controls, 2.5 times lower than in patients with benign colorectal cancer, and 1.9 times lower than in patients with malignant colorectal cancer. . Thus, we believe that using FABP for colorectal cancer screening is appropriate.

FABP 's ROC curve are shown in Figure 2.

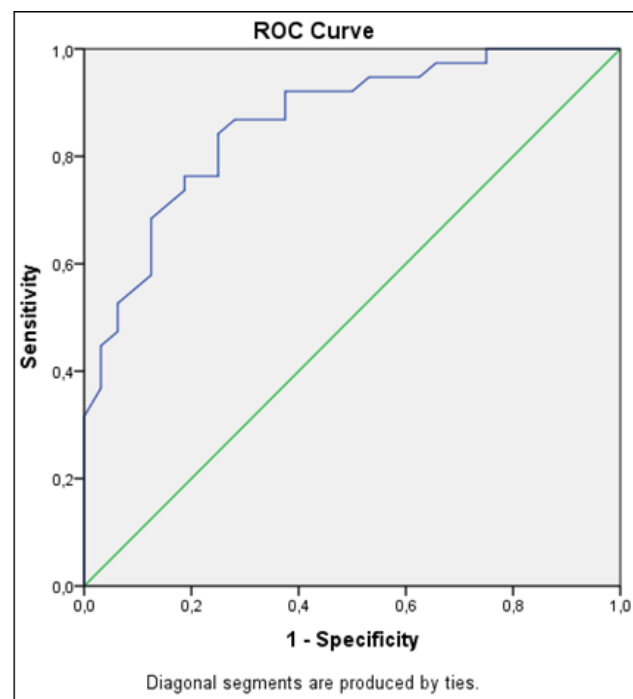


Figure 2: Statistical results of FABP (fatty acid binding protein) ROC curve (ROC curve, specificity, sensitivity, reference line) d di 0.781 based on the 95% confidence interval of the ROC curve of FABP; the upper limit is 0.948 and the area under the curve is 0.864.

The results of FABP were positive in 38 of the patients we studied, and negative in 32. This examination was not performed in 27 patients. The ROC curve of FABP was located below the straight line $y=x$, the AUC area was small. Therefore, based on our results, FABP is statistically evaluated as a test with low diagnostic value.

From the parameters obtained from the research work, it is known that IL - 6, TNF - α and FABP analysis in blood serum are among the generally accepted examination tools for the differential diagnosis of colorectal cancer and adenomatous polyp. The results of these examinations allow the attending physician to make a timely and accurate diagnosis by providing information about the nature of the disease.

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