PHYSIOLOGICAL CHANGES IN THE BLOOD COMPOSITION OF RATS EXPERIMENTALLY INFECTED WITH CROHN'S DISEASE

ФИЗИОЛОГИЧЕСКИЕ ИЗМЕНЕНИЯ СОСТАВА КРОВИ КРЫС С ЭКСПЕРИМЕНТАЛЬНОЙ МОДЕЛЬЮ БОЛЕЗНИ КРОНА

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Crohn's disease is an inflammatory bowel disease, that is characterized by chronic inflammation of any part of the gastrointestinal tract, has a progressive and destructive course and is increasing in incidence worldwide. Several factors have been implicated in the cause of Crohn's disease, including a dysregulated immune system, an altered microbiota, genetic susceptibility and environmental factors, but the cause of the disease remains unknown. In this paper, we analyzed the protein, carbohydrate, and cholesterol content of the blood of rats with chronic experimental Crohn's disease.

Болезнь Крона – воспалительное заболевание кишечника, характеризующееся хроническим воспалением любого отдела желудочно-кишечного тракта, имеющее прогрессирующее и деструктивное течение и увеличивающее заболеваемость во всем мире. В причину болезни Крона вовлечено несколько факторов, в том числе нарушение регуляции иммунной системы, измененная микробиота, генетическая предрасположенность и факторы окружающей среды, но причина заболевания остается неизвестной. В данной работе мы проанализировали содержание белков, углеводов и холестерина в крови крыс с хронической экспериментальной болезнью Крона.

Keywords: CD - Crohn's disease, UC - Ulcerative colitis, IBD - Inflammatory bowel diseases.

Ключевые слова: БК – болезнь Крона, ЯК – язвенный колит, ВЗК – воспалительные заболевания кишечника.

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Crohn's disease is an inflammatory bowel disease characterized by chronic inflammation of any part of the gastrointestinal tract, with a progressive and fatal course, and the incidence rate is increasing worldwide. Multiple factors have been implicated in the causation of Crohn's disease, including a dysregulated immune system, altered microbiota, genetic predisposition, and environmental factors, but the cause of the disease remains unknown [1]. The global prevalence of inflammatory bowel disease (IBD) has been increasing since 2000, and IBD now affects 1 in 200 people in Western countries. IBD includes two distinct diseases that differ in pathophysiology, gastrointestinal (GI) tract involvement, symptoms, complications, disease course, and treatment: Crohn's disease (CD) and ulcerative colitis (UC). The cause of CD is still unclear, but genetic, immunological, and environmental factors contribute to the risk of onset and progression of the disease [2]. CD is characterized by intestinal lesions (i.e., areas of inflammation interspersed with normal-appearing mucosa) anywhere in the gastrointestinal tract and involves chronic, relapsing transmural inflammation, which is characterized by chronic abdominal pain. causes diarrhea, obstruction, and/or perianal lesions. CD is progressive and destructive - 21-47% of patients also present with systemic, extraintestinal manifestations, strongly affecting patients' quality of life and long-term outcomes, including hospitalizations, complications, and surgery in a variety of ways. makes a secret. In addition, half of all patients with CD develop bowel complications such as strictures or fistulas within 10 years of diagnosis. Up to 30% of patients with CD have been found to have bowel involvement at diagnosis, and half of these patients require surgery within 20 years of diagnosis [3]. Although most cases of CD occur in patients under 30 years of age, the incidence is increasing in the elderly. Most studies have found no gender difference in incidence in Western countries, whereas the incidence of CD is higher in men than in women in Asian populations .

Causes of Crohn's disease. Crohn's disease is caused by the interaction between genetic susceptibility, environmental factors and intestinal microflora, which disrupts the mucosal immune response and disrupts the epithelial barrier function.

Environmental factors. In Western countries, smoking has been identified as the only modifiable risk factor for CD. Smoking doubles the risk of developing CD among more women (albeit age-dependent). Smoking is also associated with earlier disease onset, need for immunosuppression, increased need for surgery, and higher rate of postoperative disease recurrence. Intestinal dysbiosis is a characteristic feature of CD, and diet is an environmental factor (which has changed over the past decade) that may affect the gut microbiota. In particular, the host organism–gut microbiota relationship has changed with changes in dietary composition and the shift from high-fiber, low-fat foods to processed foods containing food additives . Reduced dietary fiber intake and frequent fluctuations between high-fiber and low-fiber foods lead to decreased gut microbiota diversity and predisposes to the development of CD. Two prospective studies in Sweden found

that greater adherence to a Mediterranean diet could significantly reduce the risk of subsequent CD development. As low-risk countries such as Japan, China, and India adopted Western lifestyles, the incidence of Crohn's disease increased dramatically. Despite high rates of smoking in Asia and Africa, Crohn's disease is extremely rare. Conversely, Crohn's disease is very high in northern European countries despite low smoking rates [4].

Several studies have shown that the composition of the gut microbiota can change in response to diet. In addition, dietary components can affect epigenetic modification, thereby causing long-lasting phenotypic changes . Elucidating the complex interplay between diet and gut dysbiosis in CC may improve our understanding of the role of diet in the pathogenesis of CC. Frequent use of antibiotics in childhood increases the risk of developing CK. In addition, oral contraceptives, aspirin, and NSAIDs have been reported to increase the risk of CC [5]. Among the environmental factors associated with reduced risk of CC, breastfeeding reduces the risk of CC and statins help reduce the risk [6]. Because potentially relevant environmental factors differ in different populations, interventions selected to prevent disease may be targeted to specific populations. Smoking modification, judicious use of antibiotics, promotion of breastfeeding, and advice on proper nutrition may serve as reliable approaches to reduce the development of CC and improve long-term outcomes . Although better clinical trials are awaited, dietary interventions and new elimination diets have the potential to better control the disease or prevent complications .

Genetic factors. Compared to environmental factors, more progress has been made in identifying genomic variation that determines disease risk. It was found that CD is inherited from generation to generation. In twins developed from the same egg, the development of CD is 50% higher than that of UC. About 12% of patients have a family history of Crohn's disease [16]. Genomic association studies have identified more than 200 alleles associated with inflammatory bowel disease, of which 37 are unique to Crohn's disease [7]. Only 13·1% of disease heritability is explained by genetic variation, emphasizing the importance of epigenetic and other non-genetic environmental factors. Despite all the advances, genetics alone has not been able to explain disease variation and phenotypes, and therefore genetic evaluation is not used in clinical practice. Following the seminal discovery in 2001 of a coding mutation in the NOD2 (also known as CARD15) intracellular pattern recognition receptor gene selectively associated with CD risk, more than 200 loci associated with CD in more than 70,000 individuals identified. Most of the small risk foci for CD are shared by a wide range of immune-mediated diseases . Importantly, most of the overall genetic risk is explained by variation in several loci, including the autophagy genes NOD2 and ATG16L1 (both specific for CD) and the IL-23 receptor gene IL23R (which increases the risk of CK and UC).

Microbiota. Metagenomic studies show that four major bacterial phyla (Bacteroidetes, Firmicutes, Actinobacteria, and Proteobacteria), composed of thousands of predominantly anaerobic species, colonize the human gut with a steep, gastric acid-driven, proximal-to-distal gradient. Species diversity in the gut also typically varies with temporal, individual, dietary, and drug-related factors. However, the variability of the healthy gut microbiota is generally stratified and not continuous. Comparative studies have shown clustering and reduced diversity in patients with Crohn's disease, especially Firmicutes and Bacteroides phyla. It was associated with an increased risk of postoperative recurrence of Crohn's disease in the ileum, and its experimental recovery had an anti-inflammatory effect. Crohn's disease is not only due to reduced commensal diversity, but also requires a susceptible genotype - relevant susceptibility mutations have been confirmed by studies in mice with humans.

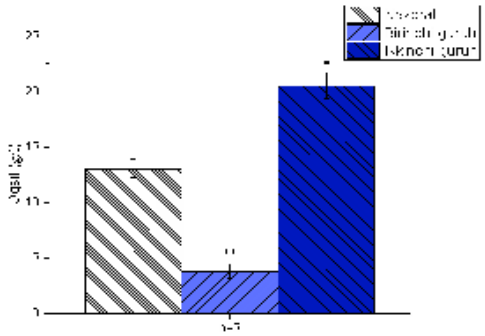
The location of the disease. About one-third of patients with CC have colonic disease, one-third have ileocolonic disease, and one-third have small bowel disease. The prevalence of upper gastrointestinal tract involvement in CC varies considerably between studies. Upper gastrointestinal tract involvement was initially thought to have a low prevalence (0.3–5 %), but a higher prevalence (30–75 %) has been reported over the past two decades [2]. "Involvement of the upper gastrointestinal tract" refers to involvement of the esophagus, stomach, duodenum, and jejunum individually or in combination with other sites. Up to one-third of patients have bowel strictures or penetrating complications at diagnosis, and half of all patients experience bowel complications within 20 years of diagnosis [3]. A large proportion of patients (40 %) have bowel damage within 1 year of diagnosis. Treatment with immunomodulators or TNF antagonists in the first 2 years after the diagnosis of CK reduces the risk of developing intestinal stricture, compared with starting treatment with these drugs more than 2 years after diagnosis. In addition, early treatment with immunomodulators reduces the risk of intestinal surgery, perianal surgery and any complications [1].

Symptoms. Symptoms can be subtle, non-specific and depend on the location and severity of the disease. Some patients may have symptoms for years before the diagnosis of CK [24]. Diarrhea and abdominal pain are the main symptoms reported by patients with CK [5]. Other symptoms include fatigue, weight loss, fever, anemia, and recurrent fistulas or other perianal findings (sores or fissures). Constipation in patients with constipation causes a lack of bowel movement, which can lead to hyperactive bowel sounds, nausea, and vomiting. Fistulas or abscesses may be a manifestation of penetrating disease. When an abscess is present, patients may have systemic symptoms such as fever and chills. Symptoms resulting from fistulas depend on the location of the fistula: diarrhea in the case of enteroenteric fistulas, and urinary tract infections in the case of enterovesicular (between the intestine and the bladder) or enteroureteral fistulas. Symptoms are similar in patients with early-onset CK and patients with late-onset CK, but there are some differences. [6]

Material and methods. A 4 % solution of acetic acid was used to create a chronic experimental Crohn's disease model in animals. For this purpose, the rats were taken care of for 1 week and adaptation was made. Then, 2 ml of 4% acetic acid was rectally injected into the hind intestine of the isolated animals using a polyurethane tube. After 24-48 hours from the time of administration, the following changes are observed in the colon wall: neutrophilic infiltration of the

intestinal wall, necrosis of the mucous and submucosal layers, dilatation of vessels, submucosal edema layer and gastric ulcer. Also, the disadvantages of using acetic acid are the significant frequency of death in experimental animals due to the perforation of the intestinal wall, significant bleeding and the development of peritonitis.

After creating an experimental Crohn's model, the protein in the blood was determined: rats infected with Crohn's were the first group, and the protein in the blood of rats infected twice with Crohn's was taken as the second group.

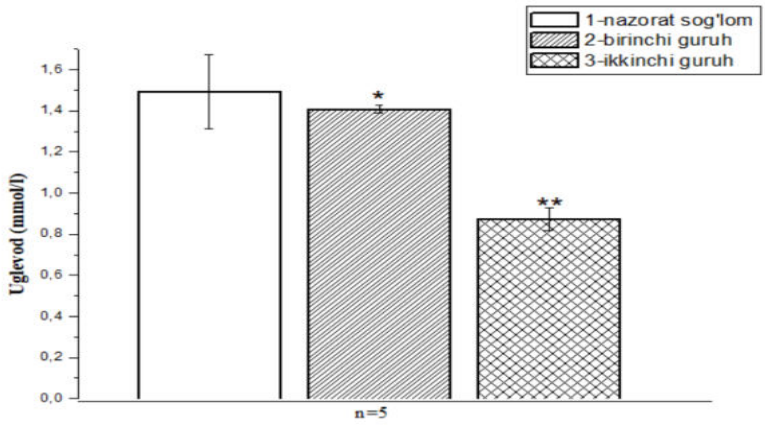


The amount of protein detected in the rat blood in the experimental model

*Note: *- P<0.05; **-P<0.01; ***-P<0.001*

When the results were analyzed, it was found that the average value in the control group was 13,044 g/l, in the first group it was 3,798 g/l, and in the second group it was 20,524 g/l. It was found that the amount of protein decreased by 70 % in a group with Crohn's disease. It was assumed that this was the result of complete absorption in the intestine. In the group with two cases of Crohn's disease, an increase in the amount of total protein in the blood was observed and increased by 36.44 % compared to the control. It was concluded that an increase in the amount of protein in the blood was observed due to the increase of immunological reactions and the occurrence of apoptosis in the cells when the disease was intensified 2 times.

After creating the experimental Crohn's model, the carbohydrate content of the blood was determined: the rats infected with Crohn's were the first group, and the carbohydrate content of the blood of the rats infected twice with Crohn's was taken as the second group.



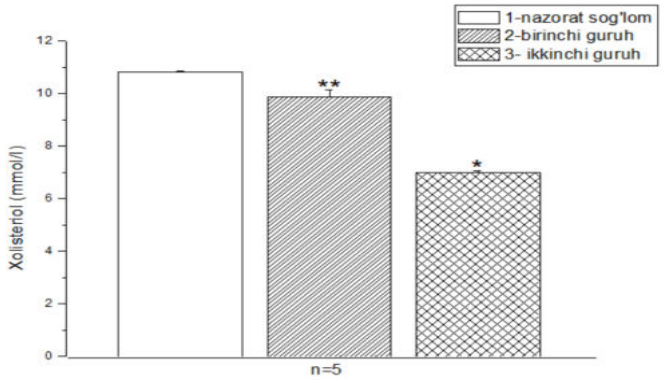
Carbohydrate content determined in rat blood in an experimental model.

*Note: *- P<0.05; **-P<0.01; ***-P<0.001*

When analyzing the results, it was found that the average value in the control group was 1.492 mmol/l, in the first group it was 1.408 mmol/l, and in the second group it was 0.874 mmol/l. A 5.63 % decrease in carbohydrates was found in the group with Crohn's disease. In the group with two cases of Crohn's disease, carbohydrate content decreased by 41.4 %.

Cholesterol in the blood was determined when the experimental Crohn's model was created: rats infected with Crohn's disease were the first group, cholesterol in the blood of rats infected twice with Crohn's disease was taken as the second group.

When the results were analyzed, it was found that the average value in the control group was 10.83 mmol/l, in the first group it was 9.872 mmol/l, and in the second group it was 7.002 mmol/l. Cholesterol was found to decrease by 8.84 % in the group with Crohn's disease once. Cholesterol content was reduced by 35.3 % in the group with two cases of Crohn's disease.



Cholesterol content determined in rat blood in an experimental model

*Note: *- P<0.05; **-P<0.01; ***-P<0.001*

Summary. Crohn's disease is an inflammatory bowel disease characterized by chronic inflammation of any part of the gastrointestinal tract, has a progressive and fatal course, and the incidence rate is increasing worldwide. Multiple factors have been implicated in the causation of Crohn's disease, including a dysregulated immune system, altered microbiota, genetic predisposition, and environmental factors, but the cause of the disease remains unknown. The appearance of the disease at a young age in most cases requires prompt but long-term treatment to prevent the development of the disease with exacerbation of the disease and intestinal complications. Clinical symptoms usually manifest as abdominal pain, chronic diarrhea, weight loss, and fatigue.

In the experimental Crohn's model, due to changes in digestion and absorption, body weight was found to decrease by an average of 2.3 times.

The amount of protein in the blood of the first group of rats with Crohn's disease decreased by 3.4 times compared to the value of healthy rats, glucose decreased by 44 %, cholesterol by 13.5 %, carbohydrates by 26 % due to the corrective effect of dihydroquercetin and tissue peptides. It was found that the protein increased by 7 % and by 42.8 %.

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ВЛИЯНИЕ МЕЗЕНХИМАЛЬНЫХ СТРОМАЛЬНЫХ КЛЕТОК И ПЛАЗМЫ, ОБОГАЩЕННОЙ РАСТВОРИМЫМИ ФАКТОРАМИ ТРОМБОЦИТОВ, НА ВЫЖИВАЕМОСТЬ ЛИМФОЦИТОВ СЕЛЕЗЕНКИ КРЫС IN VITRO

THE EFFECT OF MESENCHYMAL STROMAL CELLS AND PLASMA ENRICHED WITH SOLUBLE PLATELET FACTORS ON THE SURVIVAL OF RAT SPLEEN LYMPHOCYTES IN VITRO

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