The Role of Proinflamatory Cytokines in the Development of Clinical Picture of Myoma and Adenomyosis

G.S. Babadjanova[1], S.I. Abdurakhmanova[2], Muratova N.D.[3], Suleymanova N.Zh[4]

[1]Doctor of Medical sciences, MD, Professor, Department of Obstetrics and Gynecology No.1, Tashkent Medical Academy, 2 Farobiy Street, Almazar District Tashkent City, Uzbekistan. 100109, [2]PhD, MD, Department of Obstetrics and Gynecology No.1, Tashkent Medical Academy, 2 Farobiy Street, Almazar District Tashkent City, Uzbekistan. 100109, [3]Doctor of Medical Sciences, MD, Docent, Department of Ophtalmology with Gynecology Course, Tashkent State Dental Institute, Address: 103 Traqqiyot Str., Tashkent City, Uzbekistan. 100047, [4]PhD, MD, Docent, Department of Ophtalmology with Gynecology course, Tashkent State Dental Institute, Address: 103 Traqqiyot Str., Tashkent City, Uzbekistan. 100047

Abstract

The goal of research was to study the cytokine status in women with a combination of fibroids and adenomyosis to determine their role in the course of the disease. Method 165 women with uterine myoma and/or adenomyosis were examined. Patients were divided into 3 groups: 1-group - 75 women with a combined form of myoma and adenomyosis, 2-group - 47 women with diffuse adenomyosis, 3-group - 43 women with uterine myoma. The study was conducted in 2017-2018, on the basis of the Tashkent Medical Academy in the department of gynecology of the obstetric-gynecological complex No. 9 of Tashkent city.

The research method were a dynamic study of node blood flow by ultrasound with dopplerometry and an immunological blood test. For myoma 5 mg ulipristal acetate was prescribed for treatment for 3 months for at least 2 courses by 3 months with a 2 month break, and for adenomyosis - 2 mg dienogest for 6 months continuously. For the combined form of myoma and adenomyosis, treatment was started with dienogest for 6 months continuously, then UPA was prescribed for 3 months.

Results: The determination of the level of pro-inflammatory cytokines in patients before treatment showed the following results: the level of IL-1, IL-6 and TNF-α was higher in all three groups than in the control group, and in the group with combined pathology IL-6 was 1,5 times higher than in groups with isolated pathologies. After therapy, the decrease in the levels of IL-1, IL-6, and TNF-α was observed in all three groups. Also the level of anti-inflammatory cytokines IL-10 and VEGF exceeded significantly the control group, especially in the group with combined pathology, after the treatment, their level decreased significantly.

Discussion: An increase in the level of cytokines in the blood is most pronounced in combined forms of myoma and adenomyosis compared with isolated forms of myoma and adenomyosis.

Keywords: *Uterine myoma, adenomyosis, pro-inflammatory cytokines IL-1, IL-6, TNF, anti-inflammatory cytokines IL-10, VEGF.*

Introduction

This indicates the participation of the immune system in the development and progression of such hyperplastic processes of the uterus as myoma and adenomyosis[1].

Uterine fibroids and adenomyosis are ranked among the hyperplastic processes and diseases of the uterus[2]. However, each of these diseases has its own pathogenetic features, clinical implications and is characterized by various laboratory parameters. Uterine myoma have clinical symptoms more often with a proliferating node type. The most characteristic signs of it are menstrual irregularities, infertility and, in some cases, functional abnormality of adjacent organs[3]. Uterine adenomyosis is also manifested by disorder of menstrual and

reproductive function, pain syndrome[5]. At the same time, each of these pathologies has its own therapeutic approaches, including conservative prevention, which allows achieving positive results [6].

Modern ultrasonographs can not only distinguish myoma and adenomyosis, but also determine the degree of activity of the process[4]. However, over the past decades there has been an increase in the frequency of combined forms of myoma and adenomyosis among patients of reproductive and climacteric age. Often the presence of adenomyosis in the setting of myoma is detected only after a histopathological examination of the extirpated uterus. The relevance of the question lies in the fact that when identifying the combined form, the specialist raises the question: what diagnosis is a priority, what preparations the patient should be treated with, because not all preparations are uniquely effective for myoma and adenomyosis.

Recent studies have shown that the condition and function of various body systems, including the immune system is important in the development of myoma and adenomyosis,. The participation of the immune system has been studied well in endometriosis, but few studies have been conducted with its combination with uterine myoma, and their results are contradictory.

All this indicates the necessity for further research to improve the diagnosis of combined forms of diseases, to study the role of the immune system in the pathogenesis of these diseases.

The goal of this research was to study the cytokine status in women with a combination of myoma and adenomyosis to determine their role in the course of the disease.

Material and Research Method

The study was conducted in 2017-2018, on the basis of the Tashkent Medical Academy in the department of gynecology of the obstetric-gynecological complex No. 9 of Tashkent city. We examined 165 women with uterine myoma and/or adenomyosis. All the patients were divided into 3 groups: 1-group - 75 women with a combined form of myoma and adenomyosis, 2-group - 47 women with diffuse adenomyosis, 3-group - 43 women with uterine myoma. The study groups were formed according to the results of ultrasound, while the number of myomatous nodes and the degree of adenomyosis were payed attention to. The control group consisted of 29 healthy women. The age of the examined ranged from 20 to 45 years, the average age was 36.1±0.9 years.

The research method were a dynamic study of node blood flow by ultrasound with dopplerometry and an immunological blood test. An ultrasound study with dopplerometry made it possible to determine the presence of a simple or proliferating type of myoma node in patients and to establish the degree of adenomyosis, as well as to evaluate the effectiveness of the treatment by comparing the results of ultrasound before and after treatment. The effectiveness of treatment was also evaluated when questioning patients after treatment for the disappearance or decrease in the intensity of the pain syndrome.

An immunological study allowed us to determine the characteristics of the parameters of the immune system, including determining the level of pro-inflammatory cytokines IL-1, IL-6 and TNF and anti-inflammatory cytokines IL-10 and VEGF, to determine their role in the development of combined forms of uterine myoma, and in some cases infertility in women of reproductive age, compare indicators before and after treatment.

Interleukin-1 (IL-1) - a cytokine, a mediator of inflammation and immunity, is synthesized by many cells of the body, primarily activated macrophages, keratinocytes stimulated by B-cells and fibroblasts.

Interleukin-6 (IL-6) is a cytokine, an indicator of the immune response in diseases accompanied by inflammation, i.e. inflammatory mediator. IL-6 is a cytokine that coordinates immune and acute phase inflammatory responses, as well as oncogenesis and hemogenesis. Cytokines are specific proteins by which cells of the immune system can exchange information with each other and coordinate activities. Cytokines include some protein molecules - interferons, tumor necrosis factor (TNF), a number of interleukins, colonie-stimulating factor (CSF) and others.

Interleukin-10 anti-inflammatory cytokine is a human gene product. IL-10 has multiple pleiotropic effects on immunoregulation and inflammation. It reduces the expression of Th1 cytokines, increases the survival of B cells, their proliferation and antibody production.

Vascular endothelial growth factor (VEGF) is a signaling protein produced by cells to stimulate vasculogenesis and angiogenesis (the growth of new

vessels in an existing vascular system). The main functions of VEGF are the creation of new blood vessels, the enhancement of muscle growth after exercise, the maintenance of collateral circulation (the creation of new vessels by blocking existing ones). But, increased VEGF activity can lead to various diseases. Tumors (as well as uterine myoma and adenomyosis) cannot grow larger than a certain limited size without receiving adequate blood supply; and the tumors themselves are capable of expressing VEGF in order to grow.

Tumor necrosis factor (TNF - alpha, cachexin, cachectin) is an extracellular protein, a multifunctional pro-inflammatory cytokine, synthesized mainly by monocytes and macrophages. It affects lipid metabolism, coagulation, insulin resistance, endothelial function, stimulates the production of IL-1, IL-6, IL-8, interferon-gamma, activates leucocytes, one of the important factors of protection against viruses and intracellular parasites.

Excessive production of TNF causes hemodynamic disorders, diffusely increases the capillary permeability, the cytotoxic effect on body cells.

In our study, we performed a quantitative analysis of pro-inflammatory and anti-inflammatory cytokines in blood serum in 60 patients. 15 patients of each studied group, as well as in the control group donated blood for immunology research before treatment and after the course of therapy. The treatment was carried out depending on the revealed pathology: for myoma, ulipristal acetate 5 mg (UPA) was prescribed for 3 months at least 2 courses of 3 months with a two-month break, for adenomyosis - 2 mg dienogest for 6 months continuously. For the combined form of myoma and adenomyosis, treatment was started with dienogest for 6 months continuously, then UPA was prescribed for 3 months.

The Results of the Research

Ultrasound studies showed that the 1st group patients with the combined form of adenomyosis and myoma with clinical symptomatology (algodismenorrhea, infertility) revealed single (30 cases) or multiple (from 2x to 5 nodes - 45 cases) myomata.

Dopplerometry of the blood flow in nodes made it possible to determine a simple or proliferating myomatous node by the degree of quality of its blood supply. So in the 1st group with a combined form, about half of the patients (46.7%) had several myomatous nodes in combination with diffuse adenomyosis. A single myomatous node was combined often (25.3%) with a diffuse form of adenomyosis, and in some cases (14.7%)-combined with a diffuse focal form of adenomyosis. Several nodes of proliferating fibroids were also combined with a diffuse focal form of adenomyosis (13.3%).

Determination of pro-inflammatory cytokines level in patients before treatment showed the following results: the level of IL-1 in all three groups was almost equal (Table 1). In the 1st and 3rd groups, the level of IL-1 was 3.4 times, and in the 2nd group, IL-1 was 3.0 times higher than in the control group (P <0.001). A high level of IL-6 was observed more often in patients of the 1st group and was 3.3 times higher than in the control group (P <0.001).

Table 1: The level of pro-inflammatory cytokines in the blood serum of the examined patients before and after treatment (M±m)

№	Group	IL-1		IL-6		TNF-α	
		Before	After	Before	After	Before	After
1	The combination of myoma and adenomyosis n = 15	7,5±0,39* P<0,001	3,5±0,12[a] P<0,001	8,2±0,45* P<0,001	4,6±0,24[a] P>0,05	8,7±0,93* P<0,001	10,3±1,02 P<0,001
2	Adenomyosis n=15	6,7±0,38* P<0,001	3,2±0,22[a] P<0,01	6,0±0,79* P<0,01	4,3±0,26[a] P<0,01	12,7±1,12* P<0,001	14,6±0,95 P<0,01
3	Uterine myoma n=15	7,4±0,68* P<0,001	3,4±0,25* P<0,05	5,1±0,24 P<0,01	3,6±0,19 P<0,01	11,3±1,91* P<0,001	12,6±1,21 P<0,001
4	Control group n=15	2,2±0,15		2,5±0,21		1,5±0,07	

Note: *-acurate compared with the control group, [a] - acurate compared with the data before treatment

Also, compared with 2- and 3-groups, IL-6 exceeded 1.5 times. In the 2 group, this indicator exceeded the control group by 2.4 times (P <0.01), and in the 3 group 2 times (P <0.01).

In the 1-group with combined pathology TNF-α was 5.8 times, and in the second group for women with adenomyosis it was 8.5 times, in the 3-group it was 7.5 times higher than the control group, the accuracy was P <0.001.

After the therapy, decrease in the levels of IL-1 and IL-6 was observed in all three groups. The decrease in IL-1 was observed in all groups by an average of 2 times and the average values closed in on the control values. Also, a decrease in the cytokine IL-6 in the 1 group was observed by 1.8 times, and in the 2 and 3 groups by 1.4 times.

After treatment a decrease in the average of TNF-α, which is a pro-inflammatory cytokine was revealed in all groups. In the 1st group, the cytokine level was increased now only 2 times. The increase in its level was noted only 1.1 times in the 2nd and 3rd groups. accuracy P <0.001.

Determination of anti-inflammatory cytokines level showed that the level of IL-10 in patients with combined pathology was 3.2 times higher than in the control group (Table 2). In patients with isolated pathologies, the indicator was 1.9 times higher than in the control group. After treatment, the level of IL-10 decreased by 2 times in the 1-group. In the second group, a decrease was observed by 1.6 times and in the third group by 1.4 times.

A study of vascular endothelial growth factor (VEGF) indices showed that the highest indices were in patients of groups 1 and 2 and almost 4 times exceeded the control group (P <0.001). In the 3-group there was also a high indicator, but lower than in the 1- and 2-group, and 2.2 times higher than in the control group (P <0.001).

After the treatment, an accurate decrease in VEGF level was noted, compared with the indicators before treatment. In the 1st group the VEGF level decreased by 2 times, in the 2nd group by 1.9 times and in the 3rd group by 1.8 times.

Table 2: The level of anti-inflammatory cytokines in the blood serum of patients before and after treatment (M±m)

№	Group	IL-10		VEGF	
		Before	After	Before	After
1	The combination of myoma and adenomyosis n = 15	10,6±0,67*	5,0±0,3[a]	1282,6±119,8* P<0,001	628,7±27,4[a] P<0,05
2	Adenomyosis n=15	6,2±0,24*	3,8±0,21[a]	1243,5±127,4* P<0,001	653,2±30,36[a] P<0,01
3	Uterine myoma n=15	6,3±0,74* P<0,001	4,4±0,37[a] P<0,05	725,4±35,2* P<0,001	410,2±20,2[a] P<0,05
4	Control group n=15	3,3±0,14		325,0±22,0	

Note: * - the differences are accurate compared with the control group, [a] - the differences are accurate compared with the data before treatment

Discussion

Thus, the presence of diseases such as myoma, adenomyosis, or their combined forms in the examined patients contributes to the development of an imbalance of pro- and anti-inflammatory cytokines. The increase in their level in the blood is the most significant with combined forms of myoma and adenomyosis compared with isolated forms of myoma and adenomyosis. This indicates the participation of the immune system in the development and progression of such hyperplastic processes of the uterus as myoma and adenomyosis.

The choice of priority for prescribing preparations depended on the activity of myomatous nodules. With proliferating myomata, the preparation UPA was

prescribed first, then dienogest, and for simple myomata – vice versa. The treatment of these diseases contributed to the incremental recovery of pro- and anti-inflammatory cytokines level, which was close to the indicators of the control group.

Simultaneous ultrasound with dopplerometry after the treatment showed that stabilization of the growth of nodes in diameter of 5 and> cm and decrease in the size of small nodes were noted. The grade of blood flow in myoma nodes and in focal adenomyosis worsened, which was characterized by a decrease in the level of VEGF in the blood serum of patients, i.e. indicated the positive changes.

According to the patients survey, the clinical picture of the disease after treatment has also changed. Algomenorrhea disappeared in 24.2% of cases and its intensity decreased significantly in 61.8%, a slight improvement was noted by 14.0%. Reproductive function recovered in 67.9% within a year after the treatment.

Conclusions

1. The combined forms of myoma and adenomyosis in women of reproductive age are characterized by more significant imbalance of pro- and anti-inflammatory cytokines.
2. After the treatment, an accurate decrease in the level of pro-inflammatory cytokines is noted, which confirms the role of immunity in the development of myoma and adenomyosis.
3. The effectiveness of the treatment of combined forms of myoma and adenomyosis is characterized by the improvement in the clinical progression of the disease and restoration of fertility in 67.9% of cases.

Ethical Clearance: No ethical approval is needed.

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Conflict of Interest: Nil

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