The Significance of Cytokine Content and Substance P In the Diagnostics And Treatment Of Chronic Salpingoophoritis With Chronic Pelvic Pain Syndrome

Ziyaeva E.R Andijan State Medical Institute

ABSTRACT

Chronic salpingoophoritis (CSO) is rightfully considered the most common pathology in practical gynecology, its frequency reaches 60-65%. CSR in 24-30% of cases causes the development of chronic pelvic pain syndrome. The presence of CPPS often causes pathology in both the physical and mental spheres, which leads to maladaptation in marriage, and, accordingly, an increase in divorces, frequent temporary or even permanent loss of ability to work, which makes it a medical and social problem.

CSO complicated with CPPS is an urgent problem in gynecology. We decided to study the pathogenetic factors of CSO and CPPS based on changes in cytokines and substance P(SP).

Keywords: Chronic salpingoophoritis, chronic pelvic pain syndrome, cytokines, substance P.

Chronic salpingoophoritis (CSO) (adnexitis) is rightfully considered the most common pathology in practical gynecology, its frequency reaches 60-65%. The duration of treatment for this pathology in many cases is comparable to the duration of the patient's fertility. According to WHO, about 70% of patients with CSO receive therapy at least twice annually, but 40% of them are dissatisfied with its effectiveness.

CSO has an extremely negative effect on the reproductive function of patients, especially when the effectiveness of therapy is low. CSO in 24-30% of cases causes the development of chronic pelvic pain syndrome (CPPS), in 1-5% of cases it leads to ectopic pregnancy, while about 40% of patients with CSO suffer from infertility [29] .

The presence of CPPS often causes pathology in both the physical and mental spheres, which leads to maladaptation in marriage, and, accordingly, an increase in divorces, frequent temporary or even permanent loss of ability to work, which makes it a medical and social problem. CPPS is characterized by a protracted and often recurrent course, often accompanied by CPPS, adhesions, infertility, the development of purulent processes in the pelvis - pyosalpinx, pyovar, tubo-ovarian abscesses[33] .

CSU complicated with CPPS is an urgent problem in gynecology. We decided to study the pathogenetic factors of chronic CSO and CPPS based on changes in cytokines and substance P(SP)[32, 36].

Cytokines are mainly simple proteins or glycoproteins with a molecular weight of up to 30 kDa; only some cytokines are oligomers with high molecular weight. Cytokine production is regulated by various inducers at the level of transcription or translation. Cytokine production is transient and their range of action is usually short (typical action is autocrine or paracrine, not endocrine). Cytokines realize their effects through specific high-affinity receptors with Kd = 109–1012 M. Phenotypically, the action of cytokines leads to an increase (or decrease) in the rate of cell

proliferation, changes in cell differentiation and the manifestation of various functions of somatic cells. Although the range of biological effects of different cytokines may vary, the targets of most cytokines are hematopoietic cells[13, 20].

The role of cytokines in the regulation of body functions can be divided into 4 main components:

1. Regulation of embryogenesis, formation and development of organs, including organs of the immune system;
2. Regulation of certain normal physiological functions;
3. Regulation of the body's defense reactions at the local and systemic levels;
4. Regulation of tissue regeneration processes.

Expression of genes for individual cytokines occurs stage-specifically, at certain stages of embryonic development. Stem cell factors, transforming growth factors, TNF family cytokines and chemokines regulate the differentiation, immigration of various cells and the formation of immune system organs. After this, the synthesis of some cytokines may not resume, while others continue to be regulated.

Cytokines have a number of common biochemical and functional characteristics that distinguish them from other classes of regulatory molecules, among which the following are considered the most important: characteristic biochemical properties (cytokines are proteins or polypeptides with a molecular weight of 5 to 50 kDa), manifestation of biological activity in picogram concentrations, absence of tissue, lack of tissue and antigen specificity, pleiotropy and interchangeability of biological action, signal transmission through interaction with specific cell receptors, formation of a cytokine network. In this regard, cytokines can be isolated into a new independent regulatory system that exists along with the nervous and endocrine maintenance systems[20].

Methods for determining the concentration of cytokines are carried out using immunochemical methods. Immunochemical methods are extremely diverse and are represented by the following types of methods for quantitative determination of the concentration of soluble cytokines in various biological fluids. Enzyme-linked immunosorbent assay in various modifications of ELISA (Enzyme linked Immuno sorbent Assay ELISA) [13, 20]

Enzyme-linked immunosorbent assay in various modifications of ELISA (Enzyme linked Immuno sorbent Assay ELISA).

Radioimmunoassay (RIA). The sensitivity of the method is quite high, but in recent years radioimmunological methods have been used increasingly rare and are being replaced by ELISA to avoid work with isotopes and avoid the use of expensive radioactivity counters.

Multiplex immunoassay (MIA), which allows using monoclonal antibodies to simultaneously determine up to 100 proteins using fluorescent and other labels, including using technology of microparticles or microbeads coated with antibodies, as well as microchips (Bioplex, Multiplex analysis, Biochip Array Technology).

In this option, several cytokines are analyzed using one biochip at the same time, which significantly saves time and reagents, but the cost of equipment for carrying out such an analysis is still too high.

Cytofluorimetric methods for analyzing the expression of cell surface molecules (membrane forms of cytokines and cytokine receptors) and determining cytokines in the cytoplasm of cells.

Cytokines play a key role in the regulation of innate immunity. Cytokines are no less important in the formation and development of specific immune reactions, primarily associated with the regulation of proliferation and differentiation of T and B lymphocytes. Now there is no doubt about the division of the T-lymphocyte population into 2 fundamentally different classes: T-helpers and cytotoxic T-lymphocytes. Thanks to further studies of the functions of antigen-activated helper T-lymphocytes, it became clear that this

population of cells is also heterogeneous and can be divided into subclasses according to functional activity[20].

The cytokine balance and its disturbances during the development and course of CSO have not been fully studied, but it is reliably known that the protective functions of pro-inflammatory cytokines are stronger locally, i.e. at the point of application - at the site of inflammation, but an increase in their overall secretion is not identical to an increase in local efficiency [7].

The main component of the fight against etiological microbial factors of chronic salpingoophoritis (CSO) is generally recognized as the T-cell component of the immune response, the coordinators of which are cytokines that regulate the strength and effectiveness of the immune reaction, which determines the course and strength of inflammation [1].

Cytokines program the regulatory and effector functionality of immune cells. Normally, a balance is maintained with pro- and anti-inflammatory cytokines [15]. Infectious inflammation stimulates a local increase in their secretion, limiting inflammation and leveling existing damage [5].Healthy immunity does not allow inflammatory mediators to be secreted in any quantities, regulating both their secretion and utilization, which determines the adequacy of the body's response to infectious agents [12, 13].

The degree of lymphopenia, a decrease in T-lymphocytes (CD3), T-helper cells (CD4), immunoregulatory index (CD4/CD8), B-lymphocytes, an imbalance of antibody formation and nonspecific protection varies greatly depending on the severity of CSO and the duration of the disease, and also changes in and beyond exacerbation of pathology [9, 11, 21,].

Hypersecretion and generalization of proinflammatory cytokines causes the formation of dysfunction of various organs and systems [17].

Exacerbations of CSO introduce an imbalance in the population of lymphocytes, increasing the number of B-lymphocytes, increasing the concentration of IgA, M, G, reducing the activity of T-lymphocytes in the blood serum [3, 4]. Women with infertility due to CSO are characterized by a significant suppression of the secretion of CD3+, CD4+ and CD8+, as well as their ratios, as well as CD19+ and CD22+, as well as natural killer cells – CD57+ [6].

The basis of the immune pathogenesis of CSO are inflammatory mediators produced by the body as a response to factors of microbial aggression [14,18].

In case of CSO, the levels of IL-1P, IL-6 and TNF-a significantly increase ($p<0.001$) relative to healthy women, and the anti-inflammatory IL-4 significantly decreases ($p<0.05$). Pro-inflammatory cytokines in CSO are significantly higher than normal, anti-inflammatory cytokines are significantly lower, i.e. an imbalance of cytokine status was stated [16, 248-251]. The maximum concentration of IL-6 was found during exacerbation of CSO [6].

Substance P is a neuropeptide from the tachykinin family. Also called substance P. SP (substance P) is secreted by cells and neurons of various organs and systems of the body. In the digestive system, SP is present in enterochromaffin-like cells of the stomach and neurons of the duodenum, as well as, to a lesser extent, in the proximal jejunum, esophagus and colon. SP is similar in structure and properties to neurokinin A and consists of 11 amino acid residues: Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met. SP has a very wide spectrum of biological activity. In the digestive system, SP stimulates the motility of the esophagus, stomach, small and large intestines, and the mechanical and electrical activity of the digestive tract. SP stimulates pancreatic and salivary gland secretions. Inhibits the secretion of bile and somatostatin. SP also has a vasodilating effect, affects blood pressure, increases capillary permeability, promotes degranulation of mast cells, is a chemoattractant for leukocytes, activates the synthesis and release of inflammatory mediators, causes contraction of smooth muscles, has a secretogenic effect, stimulates release of prolactin.

SP was also detected in the cell bodies and processes of the Auerbach and Meissner intramural nerve plexuses of the intestine, cells of the mucous membrane of the pyloric part of the stomach and colon, as well as in endocrine cells and neuroepithelial bodies of the lungs, hypothalamus and substantia nigra (I.M. Pichugina et al.2022.).

SP is a protein with a wide spectrum of activity that directly and indirectly regulates the course of inflammation and the vascular bed. SP directly affects the growth and differentiation of endothelial cells, enhancing angiogenesis, dilating the vascular bed and increasing capillary permeability, which enhances the reaction of post-capillary venules responsible for the adhesion of lymphocytes and flow into the inflammatory focus [28]. SP increases the expression of endothelial cell adhesion IcAM-1 and VcAM-1, which increases the influx of lymphocytes and eosinophils into the inflammation site, the secretion of superoxide by eosinophils, the recruitment of natural killer cells and the enhancement of their cytotoxic effect [26].

The indirect effect of SP is the activation of mast cells, eosinophils, macrophages, and lymphocytes. SP promotes an increase in the secretion of: "IL-1, -2, -4, -6, -10, -12, leukotrienes, prostaglandins, histamine, FHO-a, nerve growth factor (NGF)" [22, 24]. IL-1 and FHO-a determine the secretion of neuropeptides from sensory nerves, increasing the concentration of SP [27]. SP усиливает следующие звенья: «индуцированную антигенами, митогенам и или Т-клеточным рецептором пролиферацию Т-лимфоцитов и увеличивает пролиферацию кератиноцитов,фибробластов и эндотелиальных клеток, созревание и рост функциональности дендритных клеток» [25].

Thus, SP, produced by sensory nerve endings, is a primary inflammatory mediator, enhancing the secretion and functionality of kinins and prostaglandins, serotonin and histamine, activating T cells and the secretion of interleukins, enhancing vascular reactions [25]. And the sensory nerve endings themselves demonstrate an increase in activity under the influence of inflammatory mediators, which ultimately develops into a kind of vicious circle, increasing the strength and volume of the source of inflammation [34].

The purpose of this work is to study the characteristics of changes in cytokines and SP in the blood in women with chronic urinary syndrome with and without CPPS and to determine the connection of these changes with the manifestations of pain in these patients. To achieve the goal of our work, we have set ourselves the following tasks:

1. Determination of cytokines and SP in the blood serum of patients with CSO with and without CPPS before and after treatment.
2. Determination of the relationship between changes in cytokines and SP in the blood serum of women with exacerbations of CSO with and without CPPS.

Based on literature data, ., that SP is: "a neurotransmitter of noncholinergic nerves and a major mediator of neurogenic inflammation, causing pain, mucosal edema and mucus hypersecretion" [27, 36]. An increase in the serum level of substance P is interrelated with the activity of the pathology and the severity of inflammation [22, 35].

Material and research methods.

During the period 2018-2022. We examined 136 women of reproductive age 21-35 years old (the average age of the examined patients was 27.8±2.89 CSO with and without CPPS who applied to the gynecological department.

In the main group (MG) we included 54 patients with CSO with and without CPPS in the anamnesis lasting 5-10 years with disturbances of the biocenosis of the female body; MG aged from 21 to 35 years, the average age was 28.2±1.34 years.

The comparison group (ComG) included 52 women with a history of CSO lasting up to 10 years with CPPS without disruption of the biocenosis of the female body, aged from 23 to 35 years, the average age was 28.8±1.17 years.

The control group (ConG) consisted of 30 women with CSO lasting up to 5 years without a history of CPPS, aged from 23 to 35 years, the average age was 28.6±1.25 years.

During a comprehensive examination of patients with CSO, attention was paid to complaints and an active history of the disease was collected in detail, laboratory, general clinical and instrumental examination methods were carried out. We especially carefully and actively collected the women's life history and obstetric-gynecological history, paying special attention to the time of onset, the dynamics of changes and the nature of clinical complaints and symptoms of CSO. We noted all diseases (in the prepubertal and pubertal periods) in the history of each patient, focusing on inflammatory and non-inflammatory pathologies of the genitals.

The patients were examined in accordance with international recommendations. All stages of the clinical examination of patients were carried out in accordance with the rules and regulations of gynecological diagnostics, and the clinical course of CSO was assessed according to gradations of the severity of CPPS.

In clinical symptoms, all (100%) patients with CSO had pain in the lower abdomen, moderate aching pain in 71.70%, acute severe pain in 28.30%. Fever was detected in 71.70% of women, of which 54.72% had low-grade fever. The pain in these patients was not associated with menstruation. The peculiarities of exacerbations of CSO among our patients led us to the idea of identifying and separating variants of the course of exacerbation of CSO and studying them separately, so of all the women studied, 55.21% of women had SM - a bright current exacerbation, and 47.49% - ATO - a slow exacerbation

To determine the nature of CPPS in our patients with CSO, especially with HTO CSO, we examined the serum concentration of SP - Table 1.

Table 1.

Serum levels of SP in patients of the study groups (M±m).

Group	Type of exacerbation	CP concentration, pg/ml
MG (n=54)	SM(n=26)	249,17±15,46
	OTA(n=28)	2219,34±66,98
	Average	1069,42±61,97
ComG (n=52)	SM(n=23)	241,75±13,08
	OTA(n=29)	2103,28±65,45
	Average	1052,36±60,18
Cong (n=30)	SM(n=22)	134,65±5,47
	OTA(n=8)	187,82±10,08
	Average	161,41±7,04
Norm		130,5±5,05

In the subgroups of OTA CSO of the studied groups, a large concentration of SP is explained by the presence of pain, which correlates with the literature data, as well as vasodilation and edema [31]. The concentration of SP is associated with the severity of pain, but not the severity of inflammation [23] .

On the one hand, SP is a neurotransmitter, on the other, an inflammatory mediator. SP is considered as a mediator of afferent innervation of immune system organs [27].

To determine the concentration of SP at which pain does not depend on inflammation, but is neurogenic, we used the CRO analysis. This parameter had the maximum area under the SP curve (0.933±0.0197) ($p<0.0001$). The study of the relationship between sensitivity and specificity of SP made it possible to determine a certain point of informativeness of the level of serum SP, above which pain is predominantly neurogenic in nature - 1997.98 pg/ml (rounded 2000 pg/ml). At this point, the sensitivity of the technique reached 98.16% with a specificity of 88.12%.

In patients with OH and ComG with SP levels ≤ 2000 pg/ml, we found a moderate direct correlation between the concentration of SP and the severity of exacerbation (r=0.479; p=0.001), so moderate pain (SP<2000 pg/ml) corresponded to the severity inflammation during exacerbation of chronic vomiting.

Severe pain in patients MG and ComG with SP >2000 pg/ml was characterized by a very weak positive relation ship between SP concentration and severity infla memation (r=0.0078; p=0.911), i.e. atypical pain with SP ≥2000 pg/ml is not associated with the severity of inflammation during exacerbation of CSO. The addition of a neurogenic component during exacerbations of CSO is expressed by sharp acute pain, which led to pronounced CPPS even with minor clinical symptoms of exacerbation of CSO.

We hypothesized that the difference in the severity of pain and symptoms of exacerbation of CSO in our patients may be various changes in the immune status of the women studied.

Analysis of the serum cytokine profile in patients with SM and OTA demonstrated no differences in pro-inflammatory IL-1β and anti-inflammatory IL-4 with normal ConG values (p>0.05).

Table 2.

Serum concentrations of TNFα, IL-1β, IL-6, IL-4 in patients with exacerbation of CSO (pg/ml).

Tsitokini	MG (n=54)	ComG (n=52)	ConG (n=30)
ΦНОα	4,62±0,46^^	4,22±0,71*	2,07±0,38
IL-1β	1,92±0,75^	1,62±0,39	1,06±0,53
IL-6	4,04±0,41^^	4,41±0,51**	2,16±0,35
IL-4	2,14±0,32	1,98±0,31	1,46±0,19

Note: * - statistically significant difference from the ConG indicators at p<0.05; ** - at p<0.001.^ - statistically significant difference between the MG and ComG indicators at p<0.05; ^^ - at p<0.001.

The serum concentration of proinflammatory cytokines TNFα and IL-6 in patients with CPPS CSO was found to be two times higher than the reference values (p<0.001) and in patients with CPPS CSO (p<0.001), which proves an increase in serum levels of proinflammatory cytokines in severe and prolonged inflammation in exacerbation of CSO.

During HTO CSO, serum levels of TNFα and IL-6 did not have significant differences with the ConG indicators (p>0.05), which proves the suppression of the response to inflammation. Most likely, the increase in spontaneous production of cytokines in vitro during HTO CSO characterizes the activation of immune-competent cells, but the lack of increase in the serum level of cytokines occurs due to prolonged low-intensity stimulation of the immune system and an increase in the consumption of cytokines by the cells of the macroorganism[13].

In SM CSO in both groups, the results were multidirectional - the spontaneous concentration of IL-4 was found to be two times lower relative to those below the ConG (p<0.05); spontaneous concentration of TNFα is two times higher than the results of ConG (p<0.05), which indicates an excess of the level of pro-inflammatory cytokines over anti-inflammatory cytokines. The levels of IL-1β and IL-6 were statistically insignificantly lower compared to the ConG (p>0.05).

Compared to the indicators of patients with HTO CSO, the spontaneous concentrations of all the cytokines we studied in HTO CSO were lower, so the levels of TNFα and IL-6 were twofold, IL-1β and IL-4 were 2.5 times lower (p<0.05)(table 3).

Presumably, this characterizes the inhibition of the functional activity and reserves of immunocompetent cells in these patients, since they have practically used up their physiological potential, which suggests the idea of studying their reserve potential.

Table 3.

Spontaneous levels of TNFα, IL-1β, IL-6, IL-4 in vitro in patients with exacerbation of chronic obstructive syndrome (pg/ml)

Tsitokine	MG (n=54)	ComG (n=52)	ConG (n=30)
FNOα	269,21±54,27**^	123,66±16,23*	56,76±2,37
IL-1β	22,13±4,64*^	9,79±3,00	10,56±4,41
IL-6	100,74±9,16**^^	52,10±3,98	65,14±6,32
IL-4	14,42±1,38*^	5,68±1,71*	11,83±0,37

Note: * - statistically significant difference from the ConG indicators at p<0.05; ** - at p<0.001.^ - statistically significant difference between the MG and ComG indicators at p<0.05; ^^ - at p<0.001

Having analyzed the clinical symptoms of exacerbations of chronic toxicity with the obtained immunological parameters, we stated that exacerbation of chronic toxicity in the gastrointestinal tract had more severe inflammation, which caused a decrease in induced concentrations of IL-1β - twofold, IL-6 - one and a half times (p<0.05) with a clear tendency to decrease in IL-4 (p>0.05). This is due to the limits of the reserves of immunocompetent cells due to the massive production of inflammatory mediators and the acceleration of the consumption of cytokines by the cells of the macroorganism against the background of prolonged low-intensity stimulation of immunocompetent cells by CSO.

When comparing the results of a study of patients with HTO CSO in the MG and ComG, we found a one and a half-fold decrease in the induced concentration of IL-1β (p<0.05) against the background of a constant content of IL-6, IL-4 and TNFα in comparison with the ConG, which is characteristic of some the remainder of the reserves of cells that secrete them and a weak response to inflammation (Table 4).

Table 4.

In vitro-induced levels of TNFα, IL-1β, IL-6, IL-4 in patients with exacerbation of chronic obstructive syndrome (pg/ml- u.u.)

Tsitokini	MG (n=54)	ComG(n=52)	ConG (n=30)
ΦНОα	2571,46±29,73	2541,00±33,97	2319,90±7,93
IL-1β	207,50±11,96*	188,14±22,21**	330,67±31,17
IL-6	398,93±27,64^	280,24±33,14*	416,64±8,66
IL-4	28,04±6,66	20,42±5,59	27,11±1,26

Note: * - statistically significant difference from the ComG indicators at p<0.05; ** - at p<0.001.^ - statistically significant difference between the MG and ComG indicators at p<0.05.

Calculation of the index of stimulation of cells secreting proinflammatory cytokines TNFα, IL-1β, IL-6 in patients of both groups revealed its inhibition relative to ConG. The stimulation index (SI) of TNFα in the ConG was 0.99±0.005 u.u., while in the MG it was 0.92±0.04 u.u. (p<0.001), and in ComG – 0.96±0.03 u.u. (p>0.05). IL-1β SI in MG was 0.92±0.03 u.u. (p<0.05), in ComG – 0.93±0.04 u.u. (p>0.05), and in the ConG – 0.95±0.03 u.u. The index of IL-6 in the MG was 0.74±0.06 u.u. (p<0.001), in ComG – 0.83±0.05 u.u. (p<0.05), in the ConG – 0.94±0.04 u.u.

Thus, in the studied patients with CSO, IS was found to be significantly lower compared to the ConG, but in the ConG we recorded a less significant decrease. Most likely, this characterizes an increase in the reactivity of immunocompetent cells, which in general will have a beneficial effect on counteracting inflammation, but indicates its long duration and predicts the prolongation of active CSO therapy.

The difference in the IS of IL-4 was also important, so in case of nuclear therapy it was recorded 2.2 times less in comparison with the ConG – 0.32±0.08 u.u. (p<0.001) versus 0.57±0.04 u.u. In patients with HTO, the MVR was found to be significantly greater compared to the ConG (p<0.001) – 0.78±0.08 u.u., (p<0.05). Consequently, the strong activity of inflammation in CSO causes the inhibition of SP

in the secretion of anti-inflammatory cytokines, and CPPS CSO is characterized by the preservation of cell reserves and immunity to the secretion of anti-inflammatory IL-4.

The effectiveness of the treatment was assessed by us based on the dynamics of serum concentration of SP and is presented in (Table 5).

Table 5.

Dynamics of SP level before and after therapy (M±m)

Gruppy		SP, пг/мл
Do lechenia	MG (n=54)	1069,42±61,97
	ComG (n=52)	1052,36±60,18
Cherez is 2 weeks old	MG A (n=27)	986,23±52,48
	MG B(n=27)	953,28±53,87
	Com A (n=26)	962,34±58,33
	ComB(n=26)	926,89±51,38
Cherez 3 leaves	MG A (n=27)	654,32±36,48*#
	MG B(n=27)	516,37±46,39*#
	ComGA (n=26)	412,38±36,61*#
	ComGB(n=26)	381,52±32,15*#

Note: * - the difference is statistically significant relative to its subgroup at the P≤0.05 level, # - the difference is statistically significant relative to another subgroup at the P≤0.05 level.

As follows from the presented data, SP concentrations after treatment decreased in both subgroups, this was especially clearly stated 3 months after treatment (P≤0.05). We recorded a greater tendency towards normalization in the B subgroups, and in ComG B it was maximally manifested.

Based on these data, we can make a preliminary conclusion about the better effect of complex therapy on the body of the studied patients, which confirms its greater effectiveness.

In the studied patients with CSO, we assessed the dynamics of spontaneous and induced in vitro serum levels of the following laboratory parameters: TNFα, IL-1β, IL-6, IL-4.

Table 6.

Dynamics of serum levels of pro-inflammatory cytokines in the studied patients with CSR in the dynamics of observation (M±m)

Gruppy		ΦНОα	IL-1β	IL-6	IL-4
ConG (n=30)		2,07±0,38	1,06±0,53	2,16±0,35	1,46±0,19
Do lechenia	MG (n=54)	4,62±0,46*	1,92±0,75	4,04±0,41*	2,14±0,32
	ConG (n=52)	4,22±0,71*	1,62±0,39	4,41±0,51*	1,98±0,31
Cherez is 2 weeks old	ОГ A (n=27)	3,86±0,52	1,83±0,68	3,82±0,53	1,98±0,66
	MG B(n=27)	3,77±0,41	1,69±0,71	3,79±0,47	1,93±0,61
	ConG A (n=26)	3,49±0,48	1,56±0,41	3,51±0,58	1,74±0,43
	ConG(n=26)	3,26±0,51	1,37±0,32	3,29±0,57	1,69±0,42
Cherez 3 leaves	MG A (n=27)	3,13±0,53	1,54±0,67	3,08±0,54	1,74±0,54
	MG B(n=27)	2,98±0,56	1,46±0,59	2,94±0,52	1,64±0,53
	ConG A (n=26)	2,51±0,42	1,40±0,31	2,48±0,45	1,56±0,31
	ConG B(n=26)	2,18±0,43	1,19±0,29	2,26±0,39	1,49±0,29

Note: * - statistically significant difference from the ConG indicators at p<0.05;
- statistically significant difference between MG and ComG indicators at p<0.05

According to the results of this study, a slight decrease in the immune response following a surge in immune activity allows us to predict the leveling of pathogens, however, the duration of inflammation requires repeated courses of complex treatment.

At the same time, it is worth noting a better normalization of the levels of pro-inflammatory

cytokines in patients of the B subgroups, especially HS B group.

Conclusion

The results of a comprehensive clinical-anamnestic and laboratory-instrumental study of the etiopathogenetic mechanisms of the formation and persistence of CSO that we analyzed will enable the world community to study this medical and social problem of global scale in more detail and in depth.

It should be understood that the course of CSO is determined by genital and extragenital pathology, colonization resistance of the mucous membrane of the vagina and genital cervix, as well as the characteristics of the microbiocenosis of the reproductive tract, and the hyperproduction of substance P by its secreting immunocompetent cells. Increases in serum SP levels are associated with the activity of pathology and the severity of inflammation. The addition of a neurogenic component during exacerbations of CSO is expressed by sharp acute pain, which led to pronounced CPPS even with minor clinical symptoms of exacerbation of CSO(36).

In CSO, the study of serum levels of cytokines does not make it possible to fully and comprehensively study cellular immunity, which is due to the short half-life of cytokines. An increase in spontaneous production of cytokines in vitro during HTO CSO characterizes the activation of immune-competent cells, but the absence of an increase in the serum level of cytokines occurs due to prolonged low-intensity stimulation of the immune system and an increase in the consumption of cytokines by the cells of the macroorganism.

Having analyzed the clinical symptoms of exacerbations of chronic toxicity with the obtained immunological and indicators, we stated that the exacerbation of chronic toxicity in the gastrointestinal tract had more severe inflammation, which led to a decrease in the induced concentrations of IL-1β - twofold, IL-6 - one and a half times ($p<0.05$) with a clear trend to a decrease in IL-4 ($p>0.05$). This is due to the limits of the reserves of immunocompetent cells due to the massive production of inflammatory mediators SP and the acceleration of the consumption of cytokines by the cells of the macroorganism against the background of prolonged low-intensity stimulation of immunocompetent cells by CSO.

When comparing the results of a study of patients with HTO CSO in the MG and ComG, we found a one and a half-fold decrease in the induced concentration of IL-1β ($p<0.05$) against the background of a constant content of IL-6, IL-4 and TNFα in comparison with the ConG, which is characteristic of some the remainder of the reserves of cells that secrete them and a weak response to inflammation.

Thus, in the studied patients with CSO, IS was found to be significantly lower compared to the ConG, but in the ConG we recorded a less significant decrease.

Most likely, this characterizes an increase in the reactivity of immunocompetent cells, which in general will have a beneficial effect on counteracting inflammation, but indicates its long duration and predicts the prolongation of active CSO therapy.

The strong activity of inflammation in CSO causes inhibition of the ability to secrete anti-inflammatory cytokines, and a sluggish exacerbation of CSO is characterized by the preservation of immune cell reserves for the secretion of anti-inflammatory IL-4. The results of determining the levels of SP in the blood plasma in groups of patients with CSO indicated the existence of a close relationship between the levels of these neuropeptides, cytokines and pain in the uterine appendages. This, in our opinion, serves as indirect confirmation of the theory of vein-specific inflammation that occurs against the background of varicose vein transformation and is accompanied by hypoxia of the vein wall, which should be considered as a damaging agent contributing to the formation of neurogenic inflammation in the vein wall. Possibly, blood stagnation causes activation of a cascade of pathological reactions, accompanied by activation of endothelial cells and degranulation of mast cells. It is possible that these processes generate an increase in the synthesis of SP in the perivascular ganglia, which are responsible for the development of neurogenic inflammation and the occurrence of CSO.

Conclusions:

1. When studying the indicators of cytokines and SP in the blood serum of patients with CSO with CPPS and without CPPS, it was found that during exacerbation of CSO with CPPS, there is a significant increase in the number of SP and pro-inflammatory cytokines.

2. Repeated exacerbation of CSO had more severe inflammation, which caused a decrease in induced concentrations of IL-1β - twofold, IL-6 - one and a half times, with a clear tendency to decrease IL-4, which indicates the limits of the reserves of immunocompetent cells due to the massive production of inflammatory mediators SP and acceleration consumption of cytokines by the cells of the macroorganism against the background of long-term low-intensity stimulation of immunocompetent cells with CSO.

3. Characteristic features of the dynamics of the pain peptide SP and cytokine status in women with CSO were identified, which made it possible to put forward a hypothesis about the non-inflammatory genesis of pain in this group of patients.

Литература

1. Airiyants I.R., Yagubov M.I. Chronic pelvic pain in sexological ractice.//Social and clinical psychiatry. - 2020, - t. 30 No. 3.- P. 93-98;
2. Akhtamova N.A., Zokirova N.I., Musindzhanova P.S./Innovative method of treating pelvic pain.//Re-Heald Journal, 2021.1(9), 10-14 p.
3. Kira E.F. Bacterial vaginosis // M.: MIA. – 2012. - p. 472.;
4. Kira E.F., Rogovskaya S.I., Artymuk N.V., Savicheva A.M. Results of a study of the effectiveness and safety of vaginal use of ornidazole in the treatment of bacterial vaginosis // Journal of Obstetrics and Gynecology. – 2015. - No. 8. - With. 89-95
5. Kiseleva T.V. Features of the state of nonspecific immunity in various clinical forms of inflammation of the uterus and appendages: Dissertation...doctor of medical sciences. - Novosibirsk, 2004. - 228 p.;
6. Klenina N.M., Rykalina E.B. Pharmaco-epidemiological analysis of antimicrobial therapy for salpingo-oophoritis in a hospital in the Saratov region // Bulletin. honey. Internet conf. – 2014. – No. 4. - P. 799-806.;
7. Konoval A.A. Features of the immune status of women with infertility caused by chronic salpingo-oophoritis // Scientific bulletins. Medicine series. Pharmacy. 2013. No. 25 (168). Issue 24 – p. 84-89
8. Hemp A.A. Correction of disturbances in immune, cytokine and antioxidant status in patients with chronic salpingoophoritis // Medical Immunology. - 2006. - T. 8, No. 1. - P. 97-100.
9. Kuznetsova D.E., Prokopenko S.V., Makarenko T.A. Features of the vegetative status and quality of life in patients with external genital endometriosis and chronic pelvic pain syndrome // Journal of Obstetrics and Women's Diseases. – 2019. – No. 3. – P. 12–21.;
10. Kuzmin V.N., Murrieva G.A. The importance of polymorphism and expression of cytokine genes in predicting the risk of premature birth // Attending physician. - 2013. - No. 11. - P. 26-29
11. Musakhodzhaeva D.A. Immunobiological parameters in women outside and during pregnancy, normal and under the influence of unfavorable factors. Abstract of thesis. Diss. doc. biol. Sci. - Tashkent. - 2010. – 34 p.;
12. Nazhmutdinova D.K., Kim D.K. The importance of immune mechanisms in the formation of premature abruption of a normally located placenta // Bulletin of NSU. Series: Biology, clinical medicine.- 2011.-T.9.-Iss. 3.-P.107-109.
13. Орадова А.Ш., Садуакасова К.З., Лесова С.Д. / Лабораторная диагностика цитокинов (обзорная статья)// Вестник КазНМУ №2-2017. С.200-203.
14. Parashchuk Yu.S. Kalinovskaya O.I. Infertility in the family (causes, methods of diagnosis and treatment): method. rec. for students and medical interns. - Kharkov: KhNMU, 2012. - 44 s.

15. Pitirimova L.N. Immunological and genetic predictors of recurrent miscarriage // Abstract of thesis. diss...doctor of medical sciences. St. Petersburg, - 2014. – 36 p.
16. Popkov V.M., Chesnokova N.P., Zakharova N.B. Cytokines: biological role in the development of adaptation and damage reactions under normal conditions and pathologies of various origins. – M.: Sarat Publishing House. state honey. University, 2016. – 448 p.
17. Popkova S.M., Rakova E.B., Khramova E.E., et al. Microecological combinations of vaginal and intestinal biotopes in women with inflammatory diseases of the lower floor of the reproductive tract and adolescent girls with ovarian dysfunction // Bulletin of the Siberian Branch of the Russian Academy of Medical Sciences . - 2013. - T. 33. - No. 4. - P. 77–83.
18. Russian Society of Obstetricians and Gynecologists. Diagnosis and treatment of diseases accompanied by pathological discharge from the female genital tract // Clinical recommendations. - M.: GOETAR. – Media. - 2013. –47 p.
19. Samorodskaya I.V., Rylov A.L. Chronic pelvic pain // Medical Bulletin. – 2014. - No. 30 (679) – pp. 9-10.
20. Simbirtsev A.S., Totolyan A.A. / cytokines in the laboratory diagnostics *.// Infectious diseases: news, opinions, training No. 2 2015.p.82-98.
21. Shatunova E.P., Lineva O.I., Kaganova M.A. Immune profile and optimization of treatment of patients with chronic salpingo-oophoritis // Russian Bulletin of Obstetrician-Gynecologist, 2016 - No. 6. – p.83-88.
22. Gavrilov SG, Vasilieva GY, Vasiliev IM, Efremova OI. Calcitonin Gene-Related Peptide and Substance P As Predictors of Venous Pelvic Pain. // Acta Naturae. 2019 - No. 11(4). – p.88-92.
23. Hua F, Wang HR, Bai YF, Sun JP. Substance P promotes epidural fibrosis via induction of type 2 macrophages. // Neural Regen Res. 2023 - №18(10). – p.2252-2259
24. Jin Z, Kim JB, Jin YH. Effect of endogenous substance P on visceral afferent signal integration in the nucleus tractus solitaries of rat brainstem slices. // IBRO Neurosci Rep.
25. Liu Q, Yu M, Liao M, Ran Z, Tang X, Hu J, Su B, Fu G, Wu Q. The ratio of alpha-calcitonin gene-related peptide to substance P is associated with the transition of bone metabolic states during aging and healing. // J Mol Histol. 2023 - №54(6) – p.689-702
26. Methods In Medicine CAM. Retracted: Electroacupuncture Treats Myocardial Infarction by Influencing the Regulation of Substance P in the Neurovascular to Modulate PGI2/TXA2 Metabolic Homeostasis via PI3K/AKT Pathway: A Bioinformatics-Based Multiomics and Experimental Study. // Comput Math Methods Med. 2023 - №13 p.979-988
27. Muñoz M, Pavon A. Long-term Use of Angiotensin-converting Enzyme Inhibitors Increases Substance P Levels, Which is Involved in the Risk of Lung Cancer. // Arch Bronconeumol. 2023 - №59(12) – p.848-849
28. Park DS, Oh S, Jin YJ, Na MH. et al Preliminary Investigation on Efficacy and Safety of Substance P-Coated Stent for Promoting Re-Endothelialization: A Porcine Coronary Artery Restenosis Model. // Tissue Eng Regen Med. 2024 - №21(1) p.53-
29. Pierce J, Harte SE, Afari N, Bradley CS, Griffith JW. MAPP Research Network. Mediators of the association between childhood trauma and pain sensitivity in adulthood: a Multidisciplinary Approach to the Study of Chronic Pelvic Pain Research Network analysis. // Pain. 2023 - №5 – p.12-19.
30. Schrepf AD, Mawla I, Naliboff BD, Gallop B. Neurobiology and long-term impact of bladder-filling pain in humans: a Multidisciplinary Approach to the Study of Chronic Pelvic Pain (MAPP) research network study. // Pain. 2023 - №6 – p. 29-44
31. Tempest N., Efstathiou E., Petros Z., Hapangama O.K. Laparoscopic outcomes after normal clinical and ultrasound findings in young women with chronic

pelvic pain: A cross-sec-tional study // J. Clin. Med. -2020. -Vol. 9. - №8. - P. 2593-2601

32. Velho RV, Sehouli J, Mechsner S. Mechanisms of peripheral sensitization in endometriosis patients with peritoneal lesions and acyclical pain. // Arch Gynecol Obstet. 2023 - №308(4). – p.1327-1340.
33. Zaks N, Batuure A, Lin E, Rommel AS. Association Between Mental Health and Reproductive System Disorders in Women: A Systematic Review and Meta-analysis. // JAMA Netw Open. 2023 - №6(4) – p. e238-245.
34. Zhang J, Zhang C, Miao L, Meng Z, Song G. Stigmasterol alleviates al lergic airway inflammation and airway hyperresponsiveness in asthma mice through inhibiting substance-P receptor.//Pharm Biol.2023- №61(1) – p.449-458.
35. Zhou Z, Sui X, Cao Z, Li X, Qing L, Tang J. Substance P promote macrophage M2 polarization to attenuate secondary lymphedema by regulating NF-kB/NLRP3 signaling pathway. // Peptides. 2023 - №168. – p.171-185.
36. Ziyaeva E.R., Ruzieva N.Kh., Sobirova M.R./Chronic pelvic pain syndrome in chronic salpingo-oophoritis - a modern view of pathogenesis.// Re-Health Journal 2022 №. 2. 4-9 s.

Хакида маълумот -

Зияева Эътиборхон Расулжановна Андижон Давлат тиббиёт институти

1-Акушерлик ва гинекология кафедраси ассистенти.