

PATHOGENETIC MECHANISMS OF DEVELOPMENT OF ADHESIVE PROCESS IN THE PELVIS AFTER GYNECOLOGICAL INTERVENTIONS TAKING INTO ACCOUNT THE LEVEL OF MATRIX METAL PROTEINASE AND TUMOR NECROSIS FACTOR

Zufarova Shakhnoza Alimdzhonovna, Eshdavlatov Ilkhom Eshniyozovich

Tashkent Pediatric Medical Institute, Samarkand State Medical University

<https://doi.org/10.5281/zenodo.14212520>

Abstract: The aim of this study was to evaluate the role of tumor necrosis factor α (TNF α) and matrix metalloproteinases (MMP-2 and MMP-9) in the development of adhesive disease during surgical interventions in gynecology.

Materials and methods. A comprehensive comparative immunological study of TNF- α and matrix metalloproteinases (MMP-2 and MMP-9) was conducted in 60 patients after gynecological operations in order to evaluate their role as predictors in the development of adhesive processes, of which 30 patients had an adhesive process in the small pelvis (N73.6 according to ICD-10; main group) and 30 women without an adhesive process (comparison group). The control group consisted of 20 women belonging to health group I.

Results. The average age of patients in the main group was 31.6 ± 1.9 , in the comparison group – 30.7 ± 2.1 ($p > 0.05$). Most women were in active reproductive age – 20 (66.7) and 21 (70.0%), respectively ($p > 0.05$). When studying the somatic history, a statistically significant higher level of comorbidity was established in patients in the main group: extragenital pathology was recorded 1.6 times more often in relation to the comparison group ($p = 0.001$). In 16.2% and 10% ($p = 0.048$) of cases, respectively, combined damage to several organs and systems was observed. It should be noted that statistically significant higher proportion of cardiovascular (arterial hypertension, chronic venous diseases) and endocrine diseases (obesity, hyper- and hypothyroidism) was observed in patients of the main group 2.0 ($p = 0.038$) times more often compared to the comparison group.

Conclusions. An increase in the content of matrix metalloproteinases-2 and 9 was established, which indicates a destructive effect on the extracellular matrix, creating conditions for the development of adhesions due to the growth of fibrous tissue. The range of MMP-2 levels > 400 pg/ml and MMP-9 > 350 pg/ml in determining the risk of developing adhesive processes after gynecological surgeries was established. A pathogenetic relationship between high TNF- α levels in the development of adhesive process in women who underwent gynecological surgeries was proven, a predictor content in the range of over 45 pg/ml in the development of adhesive disease was established.

Keywords: women, adhesive disease, pathogenesis, matrix metalloproteinases, tumor necrosis factor.

ПАТОГЕНЕТИЧЕСКИЕ МЕХАНИЗМЫ РАЗВИТИЯ СПАЕЧНОГО ПРОЦЕССА В МАЛОМ ТАЗУ ПОСЛЕ ГЕНИКОЛОГИЧЕСКИХ ВМЕШАТЕЛЬСТВ С УЧЕТОМ УРОВНЯ МАТРИКСНОЙ МЕТАЛОПРОТЕИНАЗЫ И ФАКТОРА НЕКРОЗА ОПУХОЛИ

Аннотация: Цель работы оценить роль уровня фактора некроза опухолей α (ФНО α) и матриксных металлопротеиназ (ММП-2 и ММП-9) в развитии спаечной болезни при хирургических вмешательствах в гинекологии.

Материалы и методы исследования. Проведено комплексное сравнительное иммуногическое исследование ФНО- α и матричных металлопротеиназ (ММР-2 и ММР-9) у 60 пациенток после проведенных гинекологических операций с целью оценки их роли в качестве предикторов в развитии спаечных процессов, из них у 30 пациенток установлен спаечный процесс в малом тазу (N73.6 по МКБ-10; основная группа) и 30 женщин без спаечного процесса (группа сравнения). Группу контроля при этом составили 20 женщин, относящиеся к I группе здоровья.

Результаты. средний возраст пациенток основной группы составил – $31,6 \pm 1,9$, в группе сравнения – $30,7 \pm 2,1$ ($p > 0,05$). Большинство женщин находились в активном репродуктивном возрасте – 20 (66,7) и 21 (70,0%) соответственно ($p > 0,05$). При изучении соматического анамнеза, у пациенток основной группы установлен статистически значимый более высокий уровень коморбидности: экстрагенитальная патология регистрировалась в 1,6 раза чаще по отношению к группе сравнения ($p = 0,001$). В 16,2% и 10% ($p = 0,048$) случаев соответственно наблюдалось сочетанное поражение нескольких органов и систем. Необходимо отметить статистически значимый более высокий удельный вес заболеваний кардиоваскулярной (артериальная гипертензия, хронические заболевания вен) и эндокринной системы (ожирение, гипер- и гипотиреоз) отмечался у пациенток основной группы в 2,0 ($p = 0,038$) раза чаще по отношению к группе сравнения.

Выводы. Установлено повышение содержания уровня матричных металлопротеиназ-2 и 9, что свидетельствует о разрушающем воздействии на внеклеточный матрикс, создающим условия для развития спаечного процесса за счет нарастания фиброзной ткани. Установлен диапазон уровней ММР-2 > 400 пкг/мл и ММР-9 > 350 пкг/мл в определении риска развития спаечных процессов после осуществленных гинекологических операций. Доказана патогенетическая связь высоких показателей ФНО- α в развитии спаечного процесса у женщин, перенесших гинекологические хирургические вмешательства, установлено предикторное содержание в диапазоне свыше 45 пкг/мл в развитии спаечной болезни.

Ключевые слова: женщины, спаечная болезнь, патогенез, матричные металлопротеиназы, фактор некроза опухоли.

INTRODUCTION.

Postoperative adhesions (PAP) of the pelvic organs is one of the most common causes of tubal- peritoneal infertility, chronic pelvic pain and ectopic pregnancy [6, 10, 14, 15], which causes a significant decrease in the quality of life (QOL) of patients [1, 3, 8] and leads to significant economic costs for the healthcare system [9, 11, 13].

It is indicated that the frequency of adhesion development after various types of gynecological surgical interventions can reach 87%, with preventive anti-adhesion measures - 51%. After gynecological surgical interventions, the predominant location of adhesions is the ovaries - in 70% of cases, less often (in 20% of cases) the fallopian tubes are involved in the adhesion process, in 8% - the body of the uterus and in 7% - the retro-uterine space [2, 4, 5].

In recent decades, there has been a real boom in the scientific community in the field of studying the so-called "epithelial- mesenchymal transitions" (EMT). EMT is considered one of the mechanisms of development of a number of diseases accompanied by organ fibrosis. As a result of EMT type 2, epithelial cells (cells of the renal tubules, alveolar epithelium, hepatocytes) are transformed into fibroblasts and myofibroblasts, which subsequently form fibers of the

extracellular matrix [12]. It is believed that chronic inflammation, which results in organ fibrosis, is a stimulator of EMT.

In an adult organism, signs of EMT are normally observed during the healing of acute wounds in keratinocytes migrating from the edge of damage. In this case, proinflammatory cytokines participate in the induction of EMT, in particular tumor necrosis factor α (TNF α) by increasing the level of bone morphogenetic protein-2. Persistent inflammation promotes EMT during the formation of hypertrophic scars. Reactivation of the EMT process can also occur as an attempt to control the inflammatory response and healing of damaged tissues, but the complexity of EMT regulation and its relationship with chronic inflammation often leads to EMT inadequacy and, as a consequence, the development of pathological fibrosis. During epithelial- mesenchymal transformation, epithelial cells separate from the basal plate and begin to destroy collagen IV, laminin, nidogen, of which it is built. [7,12] For this purpose, cells produce matrix metalloproteinases MMP-2 and MMP-9.

MAIN PART

The aim of the study: to assess the role of the level of tumor necrosis factor α (TNF α) and matrix metalloproteinases (MMP-2 and MMP-9) in the development of adhesive disease during surgical interventions in gynecology.

Materials and methods of the study: a comprehensive comparative immunological study of TNF- α and matrix metalloproteinases (MMP-2 and MMP-9) was conducted in 60 patients after gynecological surgeries in order to assess their role as predictors in the development of adhesive processes, of which 30 patients had an adhesive process in the small pelvis (N73.6 according to ICD-10; main group) and 30 women without an adhesive process (comparison group). The control group consisted of 20 women belonging to health group I.

Clinical examination of patients in the main group and comparison group included a study of complaints, obstetric and gynecological history, bimanual examination (deviation of the uterus from the midline, condition of the appendages).

Ultrasound (US) screening of the pelvic organs was performed according to a unified protocol on the Philips Affinity 30 (Japan) device, which allows visualization of tissues and organs in a real time continuum. The study was carried out in B-mode using transabdominal and transvaginal convex sensors (frequency 3.5-5 MHz).

Using ELISA, risk factors for the development of adhesions after gynecological OV were determined: TNF-alpha, MMP-2, MMP-9. Type of study - heterogeneous ELISA (enzyme-linked immunosorbent assay, ELISA).

Results of the study: the average age of patients in the main group was 31.6 ± 1.9 , in the comparison group – 30.7 ± 2.1 ($p > 0.05$). Most women were in active reproductive age – 20 (66.7) and 21 (70.0%), respectively ($p > 0.05$).

When studying the somatic history, a statistically significant higher level of comorbidity was established in patients of the main group: extragenital pathology was registered 1.6 times more often in relation to the comparison group ($p = 0.001$). In 16.2% and 10% ($p = 0.048$) of cases, respectively, combined damage to several organs and systems was observed. It should be noted that a statistically significant higher proportion of cardiovascular (arterial hypertension, chronic venous diseases) and endocrine diseases (obesity, hyper - and hypothyroidism) was noted in patients of the main group 2.0 ($p = 0.038$) times more often in relation to the comparison group.

It should be noted that the pathologies under consideration, regardless of the specific nosology, are characterized by a universal mechanism of damage (oxidative stress, endothelial

dysfunction, initiation of excessive production of proinflammatory cytokines and, as a consequence, the development of a local hyperergic inflammatory reaction).

When analyzing the content of MMP-2 and -9, as well as TNF- α in the blood serum, it was found (Table 1) that in patients with the presence of adhesive processes after gynecological interventions, a significant increase in the level of MMP -2 was noted in the blood serum, amounting to 495.5 ± 25.65 pg / ml, which was 1.5 times higher than the comparison group, where this indicator was 332.00 ± 47.35 pg / ml and 1.9 times higher than the control group (262.51 ± 8.28 pg / ml).

Table 1. The content of matrix metalloproteinases types 2 and 9 (MMP-2, MMP-9) and TNF- α in the blood serum of the examined women

Groups		MMP-9, ng /ml	MMP-2, pg /ml	TNF- α , pkg /ml
Main n=30	avg.	423.94 \pm 17.71	495.50 \pm 25.65	56.2 \pm 1.7
	min.	237,00	217,00	23.7
	Max.	740,00	765,00	61.4
Reliability of data to control indicators, P<		0,001	0.01	0,001
Comparison group n=30	avg.	329.26 \pm 45.65*	332.00 \pm 47.35**	45.7 \pm 1.3**
	min.	243.26	196.3	35.2
	Max.	448,0	414.1	51
Reliability of data to control indicators, P<		0,001	0.01	0,001
Control n=20	avg.	162.70 \pm 3.35	262.51 \pm 8.28	12.7 \pm 2.1
	min.	120,00	201,00	8.0
	Max.	269,00	538,00	17.0

Note: reliability of data between the main and comparison groups (* - P<0.05; ** - P<0.01)

MMP -9 values was also significantly higher in patients of the main group with the presence of adhesions after gynecological interventions, amounting to 423.94 ± 17.71 ng / ml , which was 1.28 times higher than the comparison group, where this indicator was 329.26 ± 45.65 ng /ml and 2.6 times higher than the control group (162.70 ± 3.35 ng /ml).

When analyzing the parameters of TNF- α in the study groups, it was determined that the highest content of TNF- α was observed in the first main group of patients, amounting to 56.2 ± 1.7 pg /ml, which was 1.23 times higher than in the second comparison group, where the average content of TNF- α in the serum of patients was 45.7 ± 1.3 pg /ml and almost 4.5 times higher than in the control group, the average value of which was 12.7 ± 2.1 pg /ml.

To establish the etiopathogenetic role of MMP -2 in the development of the adhesive process after gynecological intervention in women, it is proposed to calculate such indicators as the association coefficient (Cass), Pearson chi-square (χ^2), relative risk (RR), sensitivity (Se) and specificity (Sp), the calculation results of which are presented in Table 2.

Table 2. Diagnostic assessment of MMP -2 levels in blood serum

Quantities	MMP-2		
	<350 pg /ml	350-400 pkg /ml	>400 pg /ml

K_{ass}	0.61, $p < 0.01$	0.69, $p < 0.01$	0.96, $p < 0.001$
χ^2	5.28, $p < 0.05$	5.52, $p < 0.05$	28.1, $p < 0.001$
R.R.	3.0	4.8	12.2*
S_e (%)	62.3	73.5	89.2
S_p (%)	72.1	62.3	81.1

Moderate specificity ($S_p = 72.1\%$) and low sensitivity ($S_e = 62.3\%$) of the risk of developing adhesions after gynecological surgeries had MMP -2 levels <350 pg /ml, but with a relative risk $RR = 3.0$. The diagnostic level of MMP-2 = 350-400 pg /ml in the development of the adhesion process after surgical intervention had reliable indicators of the association coefficient ($K_{ass} = 0.69$, $p < 0.01$), contingency ($\chi^2 = 5.22$, $p < 0.05$) and relative risk ($RR = 4.8$). However, the sensitivity ($S_p = 73.5\%$) and specificity ($S_p = 62.3\%$) for this test were less than moderate.

High level of association ($K_{ass} = 0.96$, $p < 0.001$) and contingency ($\chi^2 = 28.1$, $p < 0.001$), and therefore high $RR = 12.2$ was inherent in the factor - MMP -2 > 400 pg / ml among women after gynecological operations in whom the presence of comorbid risk factors for the development of adhesions was noted. At the same time, this level had higher sensitivity ($S_e = 89.2\%$) and specificity ($S_p = 81.1\%$), i.e. in 89.2% of cases, positive test results will allow to correctly diagnose the risk of adhesions after gynecological operations.

Thus, the diagnostically significant level of MMP -2 for predicting the risk of developing adhesions after gynecological surgeries was MMP -2 >400 pg /ml.

Study of the etiopathogenetic role of the influence of MMP-9 indicators in the development of the adhesion process after gynecological intervention in women showed that MMP -9 values <350 pg /ml had moderate specificity ($S_p = 71.4\%$) and low sensitivity ($S_e = 65.1\%$), but with a relative risk of $RR = 3.9$ (Table 3)

Table 3. Diagnostic assessment of MMP-9 levels in blood serum

Quantities	MMP-9		
	<250 pg /ml	250 - 350 pg /ml	>350 pg /ml
K_{ass}	0.77, $p < 0.001$	0.81, $p < 0.01$	0.92, $p < 0.001$
χ^2	8.85, $p < 0.01$	11.78, $p < 0.01$	14.2, $p < 0.001$
R.R.	3.9	5.4	11.5
S_e (%)	65.1	70.1	91.5
S_p (%)	71.4	77.2	86.4

The diagnostic level of MMP-9 in the range of 250-350 pg /ml in the development of the adhesion process after the surgical intervention had reliable indicators of the association coefficient ($K_{ass} = 0.81$, $p < 0.01$), contingency ($\chi^2 = 11.78$, $p < 0.01$) and relative risk ($RR = 5.4$). However, the sensitivity ($S_e = 70.1\%$) and specificity ($S_p = 77.2\%$) for this test were less than high.

High level of association ($K_{ass} = 0.92$, $p < 0.001$) and contingency ($\chi^2 = 14.2$, $p < 0.001$), and therefore high $RR = 11.5$ was inherent in the factor - MMP -9 > 350 pg / ml among women after gynecological operations in whom the presence of comorbid risk factors for the development of adhesions was noted. At the same time, this level had higher sensitivity ($S_e =$

91.5%) and specificity ($S_p = 86.4\%$), i.e. in 91.5% of cases, positive test results will allow to correctly diagnose the risk of adhesions after gynecological operations.

As a result, we found that the diagnostically significant level of MMP -9 for predicting the risk of developing adhesions after gynecological surgeries was $MMP -9 > 350$ pg /ml.

TNF- α indicators in the development of the adhesion process after gynecological intervention in women showed that that TNF- α values < 35 pg /ml had moderate specificity ($S_p = 69.1\%$) and low sensitivity ($S_e = 64.5\%$), but with a relative risk $RR = 2.9$ (Table 4)

Table 4. Diagnostic assessment of MMP-9 levels in blood serum

Quantities	TNF- α		
	< 35 pg /ml	$35-45$ pg /ml	> 45 pg /ml
K_{asse}	0.71, $p < 0.001$	0.75, $p < 0.01$	0.85, $p < 0.001$
χ^2	7.95, $p < 0.01$	9.71, $p < 0.01$	12.1, $p < 0.001$
R.R.	2.9	6.1	10.8
S_e (%)	64.5	68.9	85.6
S_p (%)	69.1	74.8	83.5

The diagnostic level of TNF- α in the range of $35-45$ pg /ml in the development of the adhesion process after surgical intervention had reliable indicators of the association coefficient ($K_{ass} = 0.75$, $p < 0.01$), contingency ($\chi^2 = 9.71$, $p < 0.01$) and relative risk ($RR = 6.1$). However, the sensitivity ($S_e = 68.9\%$) and specificity ($S_p = 74.8\%$) for this test were less than high.

High level of association ($K_{ass} = 0.85$, $p < 0.001$) and contingency ($\chi^2 = 12.1$, $p < 0.001$), and therefore high $RR = 10.8$ was inherent in the factor - TNF- $\alpha > 45$ pg / ml among women after gynecological operations in whom the presence of comorbid risk factors for the development of adhesions was noted. At the same time, this level had a higher sensitivity ($S_e = 85.6\%$) and specificity ($S_p = 83.5\%$), i.e. in 85.6% of cases, positive test results will allow to correctly diagnose the risk of adhesions after gynecological operations.

As a result, we found that the diagnostically significant level of TNF- α for predicting the risk of developing adhesions after gynecological surgeries was $TNF-\alpha > 45$ pg /ml .

Thus, these results indicate the presence of a correlation between elevated levels of MMP -2, MMP -9 and TNF- α ($MMP-2 > 400$ pg /ml; $MMP -9 > 350$ pg /ml; $TNF-\alpha > 45$ pg /ml) , which are diagnostically significant indicators in determining the risk of developing adhesions after gynecological surgeries, which is associated with both the inflammatory process and other factors in the anamnesis.

CONCLUSIONS

1. An increase in the level of matrix metalloproteinases-2 and 9 was established, which indicates a destructive effect on the extracellular matrix, creating conditions for the development of the adhesive process due to the growth of fibrous tissue. The range of MMP-2 levels > 400 pg / ml and $MMP-9 > 350$ pg / ml was established in determining the risk of developing adhesive processes after gynecological operations.

2. A pathogenetic link between high TNF- α levels and the development of adhesions in women who have undergone gynecological surgery has been proven; a predictive content in the range of over 45 pg /ml has been established in the development of adhesive disease.

Literatures

1. Verkhuletskiy, I. E. Aspects of morphology and classification of the adhesive process of abdominal organs / I. E. Verkhuletskiy // Ukrainian journal of surgery. - 2019. - No. 3. - P. 30 - 33.
2. Gasparov A. S. Pelvic peritoneal adhesions: etiology, pathogenesis, diagnostics, prevention / A. S. Gasparov, E. D. Dubinskaya. - M.: OOO "Medical Information Agency", 2018. - 168 p.
3. Klykova E. S. Adhesive pelvic peritonitis: risk factors and features of cesarean section / E. S. Klykova, E. I. Novikov, I. V. Fomina // Youth Innovation Bulletin. - 2019. - Vol. 8, No. 2. - P. 137-139.
4. Magomedov M.A. Intraoperative prevention of adhesions in the abdominal cavity / M.A. Magomedov, V.S. Abdulgadiev, D.M. Damadaev // Modern problems of science and education. - 2017. - No. 3. - Access mode: <https://science-education.ru/ru/article/view?id=26528>
5. Peritoneal pelvic adhesions in women of reproductive age: diagnostics and treatment tactics / A. N. Rybalka, A. N. Sulima, S. S. Anikin, A. A. Alekseev // Tavricheskiy Mediko-Biologicheskii Vestnik. - 2015. - Vol. 18, No. 1(69). - P. 113-116.
6. Prevention of adhesions after surgical interventions on the ovaries and fallopian tubes / I. B. Manukhin, A. A. Kolesov, L. K. Bekmurzaeva, E. A. Petrovich. M., 2013. - Access mode: <http://www.petrovax.ru/medication/articles/117>
7. Puchinskaya M. V. Epithelial- mesenchymal transition in norm and pathology // Archives of Pathology. 2015. No. 1. P. 75–83.
8. Tikhomirov A.L. Risks of adhesions during surgical interventions in gynecology and their prevention / A. L. Tikhomirov, M. A. Gevorkyan, S. I. Sarsaniya // Problems of reproduction. - 2016. - V. 22, No. 6. -P. 66 - 73.
9. Consensus recommendations on adhesions (version 2014) for the ESGE Adhesions Research Working Group (European Society for Gynecological Endoscopy): an expert opinion / RL De Wilde, EA Bakkum, H. Brolmann H [et al.] // Arch Gynecol. - 2014. - Vol. 290. - P. 581 - 582.
10. González Quintero, VH Preventing adhesions in obstetric and gynecologic surgical procedures / VH González Quintero, FE Cruz Pachano // Reviews in Obstetrics and Gynecology. - 2019. - Vol. 2, No. 1. - P. 38 - 45.,
11. Is patient education about adhesions a requirement in abdominopelvic surgery? / A. Hirschelmann, C.W. Wallwiener, M. Wallwiener [et al.] // Geburtshilfe Frauenheilkd. - 2022. - Vol. 72. - P. 299 - 304.
12. Lee K., Nelson CM New insights into the regulation of epithelialmesenchymal transition and tissue fibrosis. Int. Rev. Cell Mol. Biol. 2012; 294: 171–221.
13. Peritoneal adhesions in human and veterinary medicine: from pathogenesis to therapy. A review / A. Rizzo, M. Spedicato, M. Mutinati [et al.] // Immunopharmacol Immunotoxicol. - 2018. - 32, No. 3. - R. 481 - 494.,
14. Peritoneal response to abdominal surgery: The role of equine abdominal adhesions and current prophylactic strategies / JDM Alonso, ALG Alves, M. Watanabe [et al.] // Vet. Med. Int. - 2014. - Vol. 2017. - P. 1 - 8.
15. Wiseman, DM Disorders of adhesions or adhesion-related disorder: monolithic entites or part of something bigger - CAPPS? / DM Wiseman // Seminars in Reproductive Medicine. - 2018. - Vol. 26, No. 4. - P. 356 - 368.