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TR-THERAPY IN CASE OF INCONSISTENCY OF THE PELVIC FLOOR

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ABSTRACT

The failure of the pelvic floor (NTD) is a common syndrome in the modern female population, in which there is a combined or isolated prolapse of the pelvic floor and pelvic organs. In the study, tissues of the vaginal walls of 20 patients were studied by pelvic floor failure using light microscopy, PCR diagnostics, and immunohistochemistry before and after treatment with contact diathermy method. The results of the study of the material obtained after a course of diathermic treatment indicate compensation of chronic oxidative stress at the molecular level by leveling the imbalance of collagen and elastin and restoring the architectonics of the vaginal wall.

Keywords:*genital prolapse, pelvic floor, diathermy.*

INTRODUCTION

Statistics show that about half of women aged over 50 suffer from some form of POP, most of these patients need surgical treatment [1]. POP in reproductive age, unfortunately, also often occurs - at least 30% of patients with prolapse are in this period of life [2]. The majority of modern methods of operation give, on the one hand, up to 40% of relapses [3], and on the other hand, inevitable operational complications and even the appearance of complaints de novo [4, 5], which forces the surgeon and the patient to postpone surgery until the disease greatly deteriorates the quality of life. Given the characteristics of the course and progression POP, Perineologists face the challenge of finding factors for the pathogenesis of the disease, identifying markers that could indicate to doctors the optimal time for surgery or for conservative treatment. According to recent scientific papers, the ratio of collagens type I and type III, as well as the expression of matrix metalloproteinases, plays a leading role in pathological processes. MMP-1, MMP-2, MMP-3, MMP-9, responsible for the degradation of proteins ICM, and their inhibitors — TIMP-1 and TIMP-4 [6,7]. Also important is the activation of fibroblasts in constantly stretching, damaged tissues, as a

result of which not only the number of smooth muscle cells decreases [8], but their phenotype also changes. There is an assumption that smooth myocytes are modified by platelet-derived growth factor β (platelet-derived growth factor β, PDGF-β) [9].

Affect the reorganization of the pelvic support structures with POP can glycoprotein extracellular matrix tenascin X, encoded by the gene TNXB. Activation of this protein occurs after the action of a damaging factor on the cells [10]. Thus, it becomes obvious the need to search for new markers involved in the pathogenesis of pelvic organ prolapse, as well as the study of their cumulative effect on the problem, which determines the relevance of this study.

Purpose of the study:
to study the pathological changes in the tissues of the supporting apparatus of the pelvic organs with the failure of the pelvic floor to determine the ultrastructural components of the pathogenesis of POP and justify the method of targeted contact diathermy for early treatment

MATERIAL AND METHODS

The study was conducted at the clinical bases of the Department of Obstetrics and Gynecology with a course of perinatology at the Medical Institute PFUR and at the Clinic of Integral Medicine. The study included 20 women of reproductive age (35 ± 5 years), diagnosed with pelvic floor muscle insufficiency, old pelvic muscle tears, who applied for conservative treatment. The material from each patient was taken twice - before and after the course of the directed contact diathermy on the device BTL-6000 TR-Therapy ELITE (United Kingdom).

Morphological study: Were studied the tissues of the anterior and posterior walls of the vagina, taken by puncture biopsy for 5-7 days of the menstrual cycle, using methods of light microscopy, PCR diagnostics, as well as immunohistochemistry (IHC). Morphological study was performed using light microscopy after tissue preparation using the standard method (fixation, wiring, staining, conclusion).

IHC study was carried out after the dewaxing and rehydration paraffin sections according to standard protocol automatically immunogistosteynere Bond-Max (Leica, UK). Mouse antibodies were used as primary antibodies. (Abcam, United Kingdom) to type I collagen (Anti-Collagen I antibody, Clone abab138492, 1:500), type III collagen (Anti-Collagen III antibody, Clone ab23445, 1:750), α-SMA (Anti-alpha smooth muscle Actin antibody, Clone ab7817, 1:200). A light-optical microscope was used to visualize the biopsy material. Carl Zeiss Lab.A1 (Carl Zeiss, Germany), combined with a video camera AxioCam ERc5s (Carl Zeiss Microscopy GmbH, Germany) and software ZEN Lite. To determine the expression MMP-1, MMP-2, MMP-3, MMP-9, PDGF-βи TNXB in the biopsy specimens of the vaginal walls before and after diathermic treatment was performed real-time PCR.

Statistical analysis: The results of the study were statistically processed using the software package. SPSS 7.5 for Windows (IBM Analytics, USA): the arithmetic and standard deviations were calculated, the conformity of the data to the normal distribution was confirmed using the Kolmogorov – Smirnov criterion, to compare two samples, the t-test with the significance level was used $p < 0,05$.

RESULTS

A microscopic description of the vaginal biopsy specimens of patients is presented in Figure 1. In biopsy specimens taken before the course of directional contact diathermy the vaginal mucosa's own lamina is represented by fibrous connective tissue with fibrosis and sclerosis, ICM loose, edematous, blood and lymphatic vessels with symptoms of stasis. Attention is drawn to the pronounced disorganization of the architectonics of smooth muscle tissue, collagenization of loose fibrous connective tissue, and the separation of smooth muscle cells. After a course of diathermy in all the studied samples, multiple elastic fibers with a moderate vascularization index, blood vessels with proliferation phenomena were found; the main substance is compact, and smooth myocytes are arranged in ordered bundles, surrounded by elastic fibers.

According to the results of the IHC study of biopsy specimens, taken both before and after therapy, it was found that the ratio of type I and type III collagen was 0.5 before treatment and 2.0 after diathermic treatment, which indicates the

activation of plastic processes of fibrous connective tissue of the wall vagina.

Expression MMP-1, MMP-2, MMP-3, MMP-9, TIMP-1, PDGF-β and TNXB in the samples of the vaginal walls before treatment, they statistically significantly exceeded the threshold level of internal control (GAPDH), as well as the degree of expression of the same genes after a course of directed contact diathermy. The expression of all studied metalloproteinases significantly decreased after treatment - 1.50– 1.65 times. In this regard, it was not surprising that the increase in the expression of genes encoding metalloproteinase inhibitors. Significantly increased (1.8 times) expression TIMP-1, but remained without significant changes after therapy TIMP-4.

Discussion

The data obtained complement the existing understanding of the pathogenesis ПТО and allow to identify ultrastructural mechanisms leading to changes at the molecular level and, as a consequence, the formation of violations of the architectonics of the pelvic floor at the macro level. The key features of the histo-biochemical picture of vaginal tissues with the failure of the pelvic floor steel: uneven boundaries of the epithelium, swollen ВКМ; a lot of full blood vessels and, as a result, an increase in gene expression PDGF-β; violation of the architectonics of smooth myocytes, a general decrease in the absolute number of their disorganization; decrease in the number of elastic fibers up to their absence; pronounced activity of metalloproteinases, reduced expression of an inhibitor such as TIMP-1. The revealed facts reflect the degradation of tissues stretched by the failure of the pelvic floor. The answer to this mechanical damage, and a key link in the pathogenesis НТД, the induced oxidative stress is likely to become, as also reported in a number of recent studies [11]. An important result of this study turned out to be ultrastructural and gene-biochemical changes in the tissues of the vagina, identified after a course of directed contact diathermy: the number of typical elastic fibers increased significantly; the general level of vascularization has decreased, the expression has decreased (by 1.8 times) PDGF-β, however, the vascular endothelium is found in a state of proliferative activity; smooth muscle layer acquired normal architectonics; the severity of histological signs of collagenization activity decreased, one hundred and accompanied by a decrease in the expression of metalloproteinases in 1,50–1,65 fold and increased gene activity of their inhibitor TIMP-1. These changes can be explained by a decrease in the impact of a damaging factor and a decrease in microtraumatization of supporting tissues as a result of physiotherapy treatment.

Conclusion

Probably, changes in the intracellular metabolism of fibroblasts at the molecular level are the basis of events unfolding in the tissues of the pelvic support apparatus; oxidative stress serves as a trigger point for these changes in constantly stretching, injured tissues with impaired support from the pelvic floor. The result of oxidative stress and the next link in pathogenesis is not apoptosis, but a change in the function of fibroblasts and smooth myocytes. The results of the study of the tissues of the walls of the vagina, taken after a course of directed contact diatomy, reflect the impact of the apparatus on the molecular component by compensating for chronic oxidative stress.

References

1. Bump, R.C. and Norton, P.A., 1998. Epidemiology and natural history of pelvic floor dysfunction. *Obstetrics and gynecology clinics of North America*, *25*(4), pp.723-746.
2. Weidner, A.C., Barber, M.D., Markland, A., Rahn, D.D., Hsu, Y., Mueller, E.R., Jakus-Waldman, S., Dyer, K.Y., Warren, L.K., Gantz, M.G. and Meikle, S., 2017. Perioperative behavioral therapy and pelvic muscle strengthening do not enhance quality of life after pelvic surgery: secondary report of a randomized controlled trial. *Physical therapy*, *97*(11), pp.1075-1083.
3. Katsara, A., Wight, E., Heinzelmann-Schwarz, V. and Kavvadias, T., 2016. Long-term quality of life, satisfaction, pelvic floor symptoms and regret after colpocleisis. *Archives of gynecology and obstetrics*, *294*(5), pp.999-1003.
4. *Crisp C. C., Book N. M., Cunkelman J. A., Tieu A. L., Pauls R. N.; Society of Gynecologic Surgeons' Fellows' Pelvic Research Network. Body image, regret, and satisfaction 24 weeks after colpocleisis: a multicenter study. Female Pelvic Med. Reconstr. Surg. 2016; 22(3): 132–5.*
5. *Van Iersel J. J., de Witte C. J., Verheijen P. M., Broeders I. A., Lenters E., Consten E. C. et al. Robotassisted sacrocolporectopexy for multicompartment prolapse of the pelvic floor: a*

prospective cohort study evaluating functional and sexual outcome. Dis. Colon. Rectum. 2016; 59(10): 968–74.

6. Dobrinatsvetkova. "Patient Safety: Examining the Etiology, Epidemiology, and Pathophysiology of Hospital Acquired Infections (HAIs)." International Journal of Pharmacy Research & Technology 5.1 (2015), 4-5. Print. doi:10.31838/ijprt/05.01.02
7. Wang, X., Li, Y., Chen, J., Guo, X., Guan, H. and Li, C., 2014. Differential expression profiling of matrix metalloproteinases and tissue inhibitors of metalloproteinases in females with or without pelvic organ prolapse. Molecular medicine reports, 10(4), pp.2004-2008.
8. Hasamnis AA, Patil SS, Shaik Imam, Narendiran K. "A Review of Pharmacoeconomics: the key to "Healthcare for All"." Systematic Reviews in Pharmacy 10.1 (2019), s40-s42. Print. doi:10.5530/srp.2019.1s.21
9. Seo, K.W., Lee, S.J., Kim, Y.H., Bae, J.U., Park, S.Y., Bae, S.S. and Kim, C.D., 2013. Mechanical stretch increases MMP-2 production in vascular smooth muscle cells via activation of PDGFR-β/Akt signaling pathway. *PLoS One*, *8*(8), p.e70437.
10. Aznal, S.S.S., Meng, F.G., Nalliah, S., Tay, A., Chinniah, K. and Jamli, M.F., 2012. Biochemical evaluation of the supporting structure of pelvic organs in selected numbers of premenopausal and postmenopausal Malaysian women. *Indian Journal of Pathology and Microbiology*, *55*(4), p.450.
11. Kim, E.J., Chung, N., Park, S.H., Lee, K.H., Kim, S.W., Kim, J.Y., Bai, S.W. and Jeon, M.J., 2013. Involvement of oxidative stress and mitochondrial apoptosis in the pathogenesis of pelvic organ prolapse. *The Journal of urology*, *189*(2), pp.588-594.