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### PCR DIAGNOSTIC OF HUMAN PAPILLOMAVIRUS AT WOMENS WITH ENDOMETRIOSIS

*The relationship of endometriosis and infertility for many years attracted the attention of clinicians and researchers. In the structure of gynecological disease endometriosis takes the third place after inflammatory diseases of the genital and uterine fibroids, affecting 17-80% of women of reproductive age. Infertility rate in couples of reproductive age is from 10% to 20% and does not tend to decrease. Frequency of occurrence indicator of endometriosis patients with infertility, ranging from 20 to 47.8%.  
**Keywords:** Endometriosis, reproductive age, polymerase chain reaction real time (PCR-RT), sexually transmitted infection (STI).*

#### **Methods.**

Total DNA was isolated was extracted from vaginal samples at 96 (65, 8 %) women at the time of the address it was diagnosed endometriosis and at 50 (34, 2%) - a normal state of microflora of a vagina (control group). The analysis of products of PCR of amplification carried out by PCR in real time.

#### **Results.**

It is revealed that associations endometriosis less often met three infectious agents in comparison with one and two (Candida albicans at 32 (59, 3±7, 1 % patients, and HPV 16, 18 - at 26 (48, 1± 6, 7 %).

#### **Conclusion.**

Our study demonstrates that, using a panel of DNA by means of PCR in real time the identification of endometriosis patients with minimally invasive techniques.

#### **Introduction.**

Endometriosis is one of the most urgent problems of modern gynecology. Despite more than a hundred years since the first reports of endometriosis, certain aspects of the etiology, pathogenesis, clinical, morphological and functional, immunological, biochemical, genetic variants of the disease continues to be the subject of scientific research. Although many issues have been resolved, the relevance of studying the problem of endometriosis is not reduced [1].

Endometriosis is one of the most common gynecological diseases in women during the reproductive age [2, 3]. Despite of this, endometriosis is one of the human diseases where a very long time interval exists between insurgence of the symptoms and final diagnosis, making it one of the most under-diagnosed and under-treated disease [4-7]. Therefore, the clinical value of a non-invasive diagnostic test for endometriosis would be enormous, because it could allow to immediately identify among women with sub-fertility, those suffering of endometriosis and to rapidly perform laparoscopic surgery, that has been reported to increase fertility [8, 9].

In recent years the importance of sexually transmitted infection (STI) sharply increased. Steady growth of STI at persons of young age, women of fertility age who are often accompanied by the complications leading to disability, to infertility, a pre-natal infection is observed, causing fruit and newborn diseases.

Despite ambiguity of opinions of researchers in an pathogenic role of Ureaplasma spp. and Mycoplasma hominis, in etiological classification of World Health Organization and sindromaly classification of Centers for Disease Control and Prevention [10] these microorganisms are allocated as possible etiological agents nonspecific not gonokokkal urethritis, inflammatory diseases of bodies of a small basin [11]. The question of what conditions are decisive for realization of pathogenic potential of opportunistic mycoplasmas, so far remains obscure. Numerous researches testify that it is possible to judge an etiological role of the specified activators with this or that share of probability only by results of the quantitative analysis. The standardized approaches to laboratory verification of the diagnosis of a urogenital infection are developed insufficiently.

The aim of this study is to determine the sensitivity, specificity and the predictive value of the endometriosis and to observe the risk factors associated in the study of 96 women with endometriosis in association with pathogenic and/or opportunistic causative agents of infections of a urogenital path (Candida albicans, HPV 16, 18 types).

#### **Methods.**

This enrollment began conducted in molecular-biological group of scientific clinical diagnostic laboratory at Atchabarov Research Institute of Fundamental and Applied Medicine of Kazakh National Medical University named by S.D. Asfendiyarov, Almaty, Kazakhstan, from January 2015 to December 2015. Funded by Kazakh National Medical University named by S.D. Asfendiyarov, Almaty, Kazakhstan, and grant: № of state registration 0115PK00259. Women that were eligible for project included Russian-speaking women between the ages of 18 and 45 years who were able and willing to give informed consent. Informed consent was received for all participants who were 18 years old.

As a result of the conducted complex laboratory testing at 96 (65, 8 %) women at the time of the address it was diagnosed endometriosis and at 50 (34, 2%) - a normal state of microflora of a vagina (control group). 96 patients with endometriosis were divided into 3 groups:

Group I - 30 (31, 3%) women with endometriosis;

Group II - 12 (12, 5%) women with endometriosis in association with Candidiasis vulvovaginitis;

Group III - 54 (56, 3%) women with endometriosis in association with Human Papilloma Virus 16 and 18 types (HPV).

Selection criteria in group of patient's endometriosis were: reproductive age; lack of pregnancy and lactation; clinical-morphological confirmation of the diagnosis endometriosis; existence of complaints.

Selection criteria of patients in control group were: reproductive age; lack of pregnancy and lactation; clinical-morphological confirmation of a normal state of vaginal microflora and excluded STD; absence of complaints.

At the baseline and follow-up visits, a trained nurse conducted a physical examination (including a pelvic examination) and collected endocervical specimens for testing for sexually transmitted infections. Women with bacterial or protozoan STIs were treated according to CDC guidelines [12].

Total DNA from vaginal samples was isolated using the DNA sorb-AM nucleic acid extraction kit (AmpliSens) according to the manufacturer's guidelines. DNA was allocated on an amplificatory "Rotor-Gene 6000" (Corbett Research, Australia) by set of reagents for DNA identification in a clinical material a method of PCR with gibridization-fluorescent detection of «Amplisens® Candida albicans/HPV 16,18-FL».

#### **Results.**

In studied group of women were observed moderated (66, 7±7, 5%), ochroleucous color (68, 5±8, 3%), homogeneous (70, 4±11, 2 in %), viscous (70,4±11,2 in %) allocation from sexual ways more often. At survey of mucous membranes of genitals the vagina hyperemia (48, 1±6, 7 %) and uterus necks (59, 3±7, 1 %) more often came to light, and also contact bleeding of a neck of a uterus (50, 0±6,4 by %) was noted. At bimanual survey more often in this group of women morbidity and increase in appendages of a uterus, existence of adhesive process in a small basin (51, 9±6, 6 by % and 70, 4±11, 2 in %, respectively), ( $p < 0, 05$ ).

In the control group of women in all 50 samples (100%) by PCR real-time detection of Human Papilloma Virus 16 and 18 types (HPV) not defined. All 50 samples (100%) of control group by PCR real-time detection of Candida albicans was less than  $10^3$  copies/ml, which corresponds to the performance standards.

#### **Discussion.**

Apparently from the presented data, in studied group of women the PCR method found Candida albicans at 32 (59, 3±7, 1 %) patients, and HPV 16, 18 – at 26 (48, 1±6, 7 %).

Thus, the association endometriosis with one infectious agent in the studied group of women was observed in 21 (38, 9 ± 6, 3 by %) a case. Association with two infectious agents was at 23 (42, 6 ± 6, 5 the %) patients.

#### **Conclusion**

Recently often applied method of diagnostics of causative agents of urogenital infections is PCR, allowing identifying them in liquids and organism fabrics. The method is based on the analysis of nucleotide sequence and it is considered the most sensitive (94-100 %) and specific (97-100 %). The main problem in use of PCR is connected with their exclusively high sensitivity of a method that demands observance of rigid rules of work. Besides, at interpretation of results, it is necessary to consider that PCR reveals only a small part of a genome of a microorganism and, therefore, isn't criterion of its viability.

We are demonstrated an association between increased severity of endometriosis and the future incidence of contracting an STI. Although the use of the scoring systems used in this article may not be practical in the hospital, the studies' conclusions are most helpful in counseling the patient, as having all more severe cases of BV puts that patient at risk for future STIs.

Thus, endometriosis now it is necessary to consider not only as frequent independent nozological unit, but also as a background for additional development of STI. In this regard the importance has careful laboratory inspection of each patient endometriosis on STI, including carrying out screening on existence of the extra genital centers of a chlamydia infection. Attracts attention, what even in the absence of STI, at patient's endometriosis, besides vagina defeat, signs cervicitis and/or urethritis take place. It can be caused by realization of pathogenic properties of opportunistic microorganisms.

#### REFERENCES

- 1 Colli E, Bertulesi C, Landoni M, Parazzini F. Endometriosis in pregnancy and preterm birth: evidence from the literature // J Int Med Res. – 1996. - №24. – P. 317–324.
- 2 S.E. Bulun, Endometriosis // N. Engl. J. Med. – 2009. - №360. – P. 268–279.
- 3 G. Benagiano, I. Brosens, History of adenomyosis // Best Pract. Res. Clin. Obstet. Gynaecol. - 2006. - №20. – P. 449–463.
- 4 A.Baldi, M. Campioni, P.G. Signorile, Endometriosis: pathogenesis, diagnosis, therapy and association with cancer // Oncol. Rep. – 2008. - №19. – P. 843–846.
- 5 L.C. Giudice, L.C. Kao, Endometriosis // Lancet. – 2004. – №364. – P. 1789–1799.
- 6 P.G. Signorile, A. Baldi, Endometriosis: new concepts in the pathogenesis // Int. J. Biochem. Cell Biol. – 2010. - №42. – P. 778–780.
- 7 P.G. Signorile, A. Baldi, New evidence in endometriosis // Int. J. Biochem. Cell Biol. – 2015. - №60. – P. 19–22.
- 8 R. Hadfield, H. Mardon, D. Barlow, S. Kennedy, Delay in the diagnosis of endometriosis: a survey of women from the USA and the UK // Hum. Reprod. – 1996. - №11. – P. 878–880.
- 9 T.M. D'Hooghe, A.M. Mihalyi, P. Sisma, C.K. Kyama, K. Peeraer, P. De Loecker, et al., Why we need a noninvasive diagnostic test for minimal to mild endometriosis with high sensitivity // Gynecol. Obstet. Investig. – 2006. - №62. – P. 136–138.
- 10 Kisina V. I. Centers for Control and Prevention // STD Guidelines. - 2006. - №2. – P. 88-94.
- 11 Prilepsky B.H., Sokolovsky E.V. Debated questions of clinical value of genital mycoplasmas // Clinical dermatology and venereology. - 2007. - №1. – P. 71-77.
- 12 Centers for Disease Control and Prevention. 1998 Guidelines for treatment of sexually transmitted diseases. MMWR // Recomm Rep. – 1998. - №47. – P. 91–111.
- 13 Kiselyov V. I., Dmitriev G. A., Latypov M. F. Polymerase chain reaction in diagnostics of urogenital infections. – M.: 2000. – 166 p.

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### **ЭНДОМЕТРИОЗ БАР ӘЙЕЛДЕРДІҢ АДАМ ПАПИЛЛОМАСЫ ВИРУСЫНА ПТР-ДИАГНОСТИКАСЫ**

**Түйін:** Көптеген жылдар бойы эндометриоз және бедеулік қатынасы дәрігерлер мен зерттеушілердің назарын аударды. гинекологиялық бұзылуы эндометриоздың құрылымында репродуктивті жастағы әйелдердің 17-80% қозғайтын, жыныс және миомы қабыну аурулары кейін үшінші орын алады. ұрпақты болу жасындағы жұп бедеулікті деңгейі 10% -дан 20% -ға дейін болып табылады және азайту үшін ешқандай үрдісі бар. Бедеу науқастарда эндометриоздың жиілігі анықтау 20 47,8% -ға дейін болып табылады.

**Түйінді сөздер:** Эндометриоз, ұрпақты болу жасы, нақты уақытта полимеразды тізбекті реакция (РТ-ПТР), жыныстық жолмен берілетін инфекциялар (ЖЖБИ)

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### **ПЦР ДИАГНОСТИКА ВИРУСА ПАПИЛОМЫ ЧЕЛОВЕКА У ЖЕНЩИН С ЭНДОМЕТРИОЗОМ**

**Резюме:** Отношения эндометриоза и бесплодия на протяжении многих лет привлекали внимание медиков и исследователей. В структуре гинекологических заболеваний эндометриоз занимает третье место после воспалительных заболеваний генитальной и миомы матки, поражая 17-80% женщин репродуктивного возраста. Уровень бесплодия у пар репродуктивного возраста составляет от 10% до 20% и не имеет тенденции к снижению. Частота выявления случаев эндометриоза у пациентов с бесплодием составляет от 20 до 47,8%.

**Ключевые слова:** Эндометриоз, репродуктивный возраст, полимеразная цепная реакция в реальном времени (ПЦР-РВ), инфекция, передаваемая половым путем (ИППП)