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ТИББИЁТДА ЯНГИ КУН
НОВЫЙ ДЕНЬ В МЕДИЦИНЕ
NEW DAY IN MEDICINE

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METHODS FOR DIAGNOSING DISEASES OF THE UTERINE CERVIX

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✓ ***Resume***

Goal: was - the study of the state of the vaginal microbiocenosis and determination of the presence of diseases of the cervix, using the PCR method (Femoflor-16) - for the differential diagnosis of dysbiotic disorders and infectious and inflammatory diseases of genitalia in women.

Methods: Clinical and laboratory examination of 105 patients with cervical diseases was carried out. All patients underwent the necessary diagnostic standard: examination of the cervix in the mirrors, cytological examination of smears from the ecto- and endocervix, polymerase chain reaction (PCR) for HPV, pH-metry, extended colposcopy, histological examination of cervical biopsies. Material for the study of vaginal microbiocenosis was collected from the posterolateral wall of the vagina, for HPV detection - from the cervical canal. The scraping was placed in an Eppendorf tube containing 1 ml of saline; storage and transportation of the material was carried out in accordance with the current regulatory documents. DNK was isolated using a PROBA-GS reagent kit (Standard Diagnostics, Bukhara). The study was carried out by PCR with detection of results in real time (RT-PCR) using Femoflor-16 reagents (Standard Diagnostics, Bukhara) in a detecting amplifier DT-96, according to the manufacturer's instructions, in the laboratory (Standard Diagnostics, g . Bukhara).

Keywords: colposcopy, base-line and pre-cancer diseases of cervix of the the uterus, diagnostics of diseases of cervix of the uterus, femoflor-16.

МЕТОДЫ ДИАГНОСТИКИ ЗАБОЛЕВАНИЙ ШЕЙКИ МАТКИ

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✓ ***Резюме***

Цель: изучение состояния микробиоценоза влагалища и определение наличия заболеваний шейки матки, с использованием метода ПЦР (Фемофлор-16) - для дифференциальной диагностики дисбиотических нарушений и инфекционно-воспалительных заболеваний гениталий у женщин.

Методы: Проведено клинико-лабораторное обследование 105 пациенток с заболеваниями шейки матки. Всем пациенткам проводился необходимый стандарт диагностики: осмотр шейки матки в зеркалах, цитологическое исследование мазков из экто- и эндоцервикса, полимеразная цепная реакция (ПЦР) на ВПЧ, pH-метрия, расширенная кольпоскопия, гистологическое исследование биоптатов шейки матки. Материал для исследования

микробиоценоза влагалища забирали из заднебоковой стенки влагалища, для выявления ВПЧ – из цервикального канала. Соскоб помещали в пробирку Эппендорфа, содержащую 1 мл физиологического раствора, хранение и транспортировку материала осуществляли в соответствии с действующими нормативными документами. ДНК выделяли с помощью набора реагентов ПРОБА-ГС (Стандарт Диагностикс, Бухара). Исследование проводили методом ПЦР с детекцией результатов в реальном времени (ПЦР-РВ) с использованием реагентов Фемофлор-16 (Стандарт Диагностика, г. Бухара) на детектирующем амплификаторе ДТ-96, согласно инструкции производителя, в лабораторных условиях (Стандарт Диагностика, г. Бухара).

Ключевые слова: кольпоскопия, предраковые заболевания шейки матки, диагностика заболеваний шейки матки, фемофлор-16.

BACHADON BO'YNI KASALLIKLARINI TASHHIS QO'YISH USULLARI

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✓ ***Resume***

Maqsad: vaginal mikrobiosenoz holatini o`rganish va ayollarda jinsiy a'zolarning yuqumli va yallig`lanish kasalliklarini differensial tashxis qilish uchun PZR usuli (Femoflor-16) yordamida bachadon bo`yni kasalliklarini aniqlash.

Bachadon bo'yni kasalliklari bilan og'rigan 105 nafar bemor klinik-laborator tekshiruvdan o'tkazildi. Barcha bemorlarga zarur diagnostika standarti o'tkazildi: bachadon bo'yni ko'zgularda tekshirish, ekto- va endoserviksdan sitologik tekshirish, OPV uchun polimeraza zanjiri reaksiyasi (PZR), pH-metriya, kengaytirilgan kolposkopiya, bachadon bo'yni biopsiyalarining gistologik tekshiruvi. Qin mikrobiosenozni o`rganish uchun material qinning orqayon devoridan, OPVni aniqlash uchun – servikal kanaldan yig`ilgan. Surtmani 1 ml fiziologik eritmaga ega bo`lgan Eppendorf-probirkasiga joylashtirildi; materialni saqlash va tashish amaldagi me'yoriy hujjatlarga muvofiq amalga oshirildi. DNKni GS-sinamasining reaktiv to`plami ("Standart diagnostika", Buxoro shahri) yordamida aniqlandi. O`rganishlarni real vaqtda PZR usuli (PZR-RV) yordamida Femoflor-16 reagentlaridan foydalangan holda ("Standart diagnostika", Buxoro shahri) DT-96 detektor kuchaytirgichida ishlab chiqaruvchining ko`rsatmalariga muvofiq ("Standart diagnostika", Buxoro shahri) o`tkazildi.

Kalit so'zlar: kolposkopiya, bachadon bo'yni saraton oldi kasalliklari, bachadon bo'yni kasalliklari diagnostikasi, femoflor-16.

Relevance

Pathology of the cervix is one of the most common diseases in the world and requires a lot of attention. This is due to the fact that they leave their mark on the reproductive activity of women. But the main problem is different: all background diseases at some point can lead to malignancy of the formations. Meanwhile, cervical cancer ranks third in terms of frequency of occurrence among diseases of the reproductive system [8,9].

There are various types of cervical pathologies, among which there are three main groups: background diseases, precancerous and cancerous. The former include cervical erosion, ectopia, polyps, and leukoplakia. The group of precancerous diseases is mainly represented by different types of cervical dysplasia [4,13].

The significant frequency of cervical diseases and their adverse effect on the reproductive health of women determine the need to improve knowledge about the features of etiopathogenesis, about early diagnosis and prevention of this pathology.

As a rule, the pathology of the cervix is accompanied by urogenital and or viral infection [5,6]. Diseases of the cervix of the uterus of infectious origin are one of the most common reasons for women to seek medical attention and represent an important medical and social problem due to the possibility of developing malignant pathology [2,7]. The urgency of the problem of diagnosis and prevention of cervical diseases is primarily due to the ability of this pathogen to initiate oncological transformation. Cervical cancer is the second most common cancer of the reproductive system in the world and the first leading cause of cancer death in women in developing countries.

Chronic inflammation is one of the etiological factors in the development of tumor and precancerous diseases of the cervical epithelium [1,2]. Currently, a correlation has been established between bacterial vaginosis (BV) and persistence of human papillomavirus (HPV) in the cervical canal [3,4]. It has been shown that an increase in vaginal pH increases the risk of infection with several types of HPV and the development of LSIL in women under the age of 35 and over 65 years [5]. According to foreign researchers, BV is one of the cofactors of cervical neoplasia [6,8]. At the same time, according to J.M. Klomp et al. [13-53], among anaerobes in cervical neoplasia, Gardnerella vaginalis is the most common.

Since opportunistic bacteria, when they reach high concentrations in the vagina and cervix, have the potential ability to induce and maintain a dysplastic process [8,10], the study of vaginal microflora in cervical intraepithelial neoplasias and HPV persistence is important for understanding the mechanisms of tumor transformation of the cervical epithelium [11].

In most cases, the development of invasive squamous cell carcinoma of the cervix is preceded by cervical intraepithelial neoplasia (CIN), which is divided into 3 degrees: CIN1 corresponds to mild stratified squamous epithelium dysplasia (MPE), CIN2 - moderate dysplasia and CIN3 - severe in dysplasia and carcinoma. Essentially, this morphological classification of Richard [1] corresponds to the stages of development of a malignant process [2]. The combination of severe dysplasia and carcinoma in situ into one group of CIN3 is due to the fact that differential morphological diagnosis between them is difficult, while the treatment tactics are the same.

Histological and cytological criteria for diagnosing CIN differ. During histological examination, the main criterion for determining the degree of CIN is the degree of involvement of the MBE layer in the pathological process. With CIN1, atypical cells occupy the lower 1/3 of the layer, with CIN2 - up to 2/3 of the layer, with CIN3 - almost the entire epithelial layer. In CIN1, there are few such cells and they differ slightly from normal cells of the corresponding MBE layer; in CIN2, there are more of them and the degree of atypia is more pronounced; in CIN3, there can be many such cells and it is difficult to distinguish them from cancer cells. Such unclear criteria in a number of cases create difficulties in the differential morphological diagnosis of various degrees of CIN and are the reason for the discrepancy in diagnoses between different specialists. This was the reason for the development of a new cytological classification of Bethesda [3] to include CIN2 together with CIN3 in one group - pronounced atypical squamous cell changes (HSIL). The use of such a classification is quite justified when conducting cytological screening for cervical cancer, since it allows not to miss cases with severe atypia of the epithelium and to send patients for in-depth examination and treatment to the appropriate specialized institutions. However, with an in-depth examination, it is desirable to more accurately determine the stage of development of the neoplastic process, since the choice of treatment tactics depends on this. From this point of view, it seems controversial to switch from a three-stage classification of CIN to a two-stage system (LSIL-HSIL), similar to Bethesda's classification in the modern WHO International Histological Classification of Cervical Tumors [4], despite the authors' recommendations to indicate CIN2 or CIN3 in brackets when diagnosing HSIL. Combining lesions with different biological potential into one group is controversial [5, 6]. This obliges doctors to carry out more radical treatment for such patients - conization of the cervix. In many cases, this is probably justified, although such treatment in young women can cause complications during subsequent pregnancy [7, 8-51]. It has also been shown that various methods of conservative treatment in patients with CIN 1-2 can be successful and lead to a complete regression of atypical changes in the cervical epithelium. Therefore, a more accurate definition of the stage of development of the malignant process remains very relevant.

Femoflor-16 is designed to detect the DNA of pathogenic and opportunistic microorganisms in order to assess the state of not only the microflora of the vagina and urogenital tract in women by PCR with

detection of results in real time. Femoflor allows the study of difficult to cultivate anaerobic microorganisms and at the same time has a high sensitivity and specificity. An express test for assessing the state of microbiocenosis and inflammation is carried out using microscopic analysis. This method is still one of the most popular because it is fast, convenient and cheap. However, the use of microscopy is associated with low sensitivity of the method, subjective results and approximate quantitative assessment. The causative agents of female genital infections, as a kind of exogenous factors, can also affect the vaginal microbiocenosis, most likely, both directly and indirectly through endocrine-immune mechanisms [14].

Investigation of the biocenosis of the vagina by the molecular biological method using the laboratory test "Femoflor-16"

The material for the study was scrapings of epithelial cells of the cervical canal of the cervix (exocervix). The material was placed in an Eppendorf tube containing 0.5 ml of saline; storage and transportation of the material was carried out in accordance with the current regulatory documents. DNK was isolated using the Proba-GS reagent kit (Standard Diagnostics, Bukhara). The study of vaginal biocenosis was carried out by RT-PCR using Femoflor reagents ("Standard diagnostics" of Bukhara) in a detecting amplifier DT-96 according to the manufacturer's instructions ("Standard diagnostics" of Bukhara). The study of microbiocenosis is complex, taking into account the biota of the site as a whole. The number of epithelial cells in the taken material was assessed by the results of the analysis of human genomic DNK in each sample. Using specialized software, the amount (in genome equivalents / ml (ge / ml)) of the total bacterial mass (TBM), lactobacilli and various groups of opportunistic microorganisms (facultative and obligate anaerobic MOs, mycoplasmas, and yeast-like fungi) was calculated. The proportion of normal flora, facultative anaerobic microorganisms and anaerobic microorganisms as a percentage among all identified bacteria was estimated. In accordance with the data of clinical approbation of 82 tests "Femoflor-16", the following classification of types of biocenosis was proposed:

1. normocenosis (absolute normocenosis) - a variant of biocenosis, in which the proportion of normal flora in its composition was more than 80% relative to the total bacterial mass (BMM), the amount of Ureaplasma spp., Mycoplasma spp., Candida spp. less than 104 ge / ml;
2. relative normocenosis - a variant of biocenosis, in which the proportion of normal flora in its composition was more than 80% relative to MBP, the amount of Ureaplasma spp., Mycoplasma spp., Candida spp. more than 104 ge / ml;
3. moderate (aerobic or anaerobic) dysbiosis - a variant of biocenosis, in which the proportion of lactobacilli is determined in the range of 20-80% relative to MBP and the proportion of aerobes or anaerobes is increased;
4. pronounced (aerobic, anaerobic or mixed) dysbiosis is a variant of biocenosis, in which the proportion of aerobes or anaerobes reaches 80% relative to MBP, and the proportion of lactobacilli decreases by less than 20% relative to MBP.

The introduction of modern molecular biological diagnostic methods made it possible to significantly expand the understanding of the species composition of opportunistic microorganisms living in the vagina. A qualitative and quantitative assessment of the main significant participants in vaginal microbiocenosis creates the preconditions for the development of a differentiated approach to the treatment of identified dysbiotic disorders in patients with cervical neoplasias before destruction.

Goal: was - the study of the state of the vaginal microbiocenosis and determination of the presence of diseases of the cervix, using the PCR method (Femoflor-16) - for the differential diagnosis of dysbiotic disorders and infectious and inflammatory diseases of genitalia in women.

Materials and methods

Clinical and laboratory examination of 105 patients with cervical diseases was carried out. All patients underwent the necessary diagnostic standard: examination of the cervix in the mirrors, cytological examination of smears from the ecto- and endocervix, polymerase chain reaction (PCR) for HPV, pH-metry, extended colposcopy, histological examination of cervical biopsies. Material for the study of vaginal microbiocenosis was collected from the posterolateral wall of the vagina, for HPV detection - from the cervical canal. The scraping was placed in an Eppendorf tube containing 1 ml of saline; storage and transportation of the material was carried out in accordance with the current regulatory documents. DNK was isolated using a PROBA-GS reagent kit (Standard Diagnostics, Bukhara). The study was carried out by PCR with detection of results in real time (RT-PCR) using

Femoflor-16 reagents (Standard Diagnostics, Bukhara) in a detecting amplifier DT-96, according to the manufacturer's instructions, in the laboratory (Standard Diagnostics, ct. Bukhara).

In accordance with the results of histological examination and testing for HPV, all examined patients were divided into 4 groups: Group 1 consisted of 48 women with underlying cervical disease, Group 2 consisted of 19 HPV-negative patients with LSIL, Group 3 included 21 HPV-positive patients with LSIL, Group 4 consisted of 17 patients with histologically confirmed diagnosed with HSIL, all patients in this group were HPV-positive. The average age of all surveyed women was 34.9± 3.2 years.

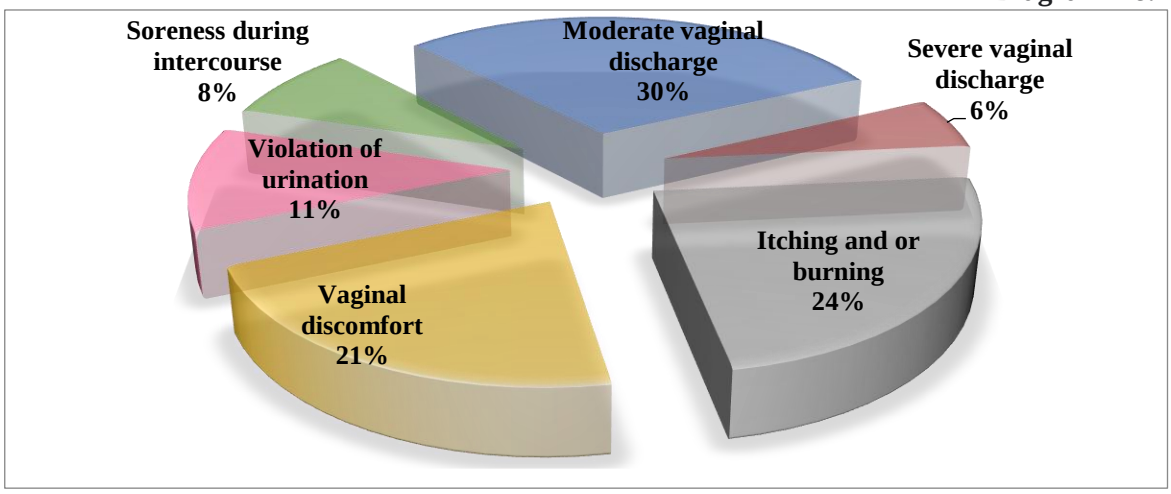
All the obtained research data were subjected to statistical processing on a personal computer using the Microsoft Office Excel – 2010 and IBM SPSS Statistic 20 software package in the Windows 10 Pro environment. We used the methods of variational parametric and nonparametric statistics: the arithmetic mean (M), the standard deviation (σ), the standard error of the mean (m), and relative values в (frequency,%) were calculated. When comparing the mean values, we calculated the Student's test (t) with the error probability (P) with a normal distribution and F — Fisher's test.

To assess the statistical reliability of the calculated criteria, indicators and tables of critical values for acceptable levels of significance (P) were used. For statistically significant changes, four main levels of significance were taken: high - P <0.001, medium - P <0.01, low (marginal) - P <0.05, insignificant (unreliable) - P> 0.05.

Result and discussions

In our study, in women with complaints of minor and moderate vaginal discharge - 41.7%, severe vaginal discharge disturbed 7.8% of patients. Complaints of itching and or burning were reported by 33% of patients, vaginal discomfort - 30%, urination disorder - 15.5%, pain during intercourse - 11.6% (Diagram No. 1).

Diagram No. 1



When analyzing the structure of the vaginal microbiocenosis, significant differences were revealed between the groups of examined women, depending on the state of the cervical epithelium and HPV infection. In 95.5% of clinically healthy women (group 5), the state of the vaginal microbiota, according to RT-PCR, met the criteria for normocenosis; including in 15 (68.2%) patients absolute normocenosis was revealed, in 6 (27.3%) - conditional normocenosis due to the presence of Ureaplasma spp. and Candida spp. in the amount of more than 104 GE / ml. Dysbiosis, according to RT-PCR, was detected in only 1 women.

In patients of group 1 (background diseases of the cervix), normocenosis was revealed statistically significantly less frequently than in the control group: the composition of the microbiota corresponded to the criteria of absolute normocenosis in 41 (41%), and conditional normocenosis - in 33 (33%) examined. In 26 (26%) patients, the state of microbiocenosis met the criteria of dysbiosis, including moderate - in 9%, severe - in 17%.

In patients of the 2 and group (LSIL, HPV-negative) did not differ significantly from the indicators, the composition of the microbiota corresponded to the criteria of absolute normocenosis in 23 (47.9%),

and conditional normocenosis - in 17 (35.4%) examined. In 8 (16.7%) patients, the state of microbiocenosis corresponded to the criteria of dysbiosis, including moderate - in 10.4%, severe - in 6.3%. The structure of the vaginal microbiocenosis in patients of the third group (LSIL, HPV-positive) did not differ significantly from the indicators of the 2 and group.

Thus, in women with LSIL, regardless of the presence of HPV, dysbiotic disorders were statistically significantly more frequent than in the control group. On the one hand, this observation may indicate the significance of abnormalities in the vaginal microbiocenosis in the occurrence of dysplastic lesions of the cervix at the initial stage, even without the influence of the virus. On the other hand, it cannot be ruled out that HPV-negative patients at the time of examination were infected with the virus in the past, which was the starting point for the development of dysplasia. In the case of the subsequent spontaneous elimination of HPV, the virus, of course, cannot be determined at the present time, and the accompanying dysbiotic process contributes to the preservation of changes in epithelial cells characteristic of LSIL.

Among the patients of the fourth group (HSIL), normocenosis was detected only in every fourth group, including absolute normocenosis - in 2 (11.8%), conditional - in 3 (17.6%) examined. In the majority of women with HSIL, the state of the vaginal microbiota met the criteria for dysbiosis, including severe dysbiosis (53%) of the examined. The frequency of detection of dysbiosis in patients of the fourth group was statistically significantly higher than in conditionally healthy women (5th group), as well as in patients with LSIL (2nd and 3rd groups), which may support the theory of the role of viral-bacterial associations in the progression of the dysplastic process in the cervix (table No. 1).

Table No. 1

The structure of the vaginal microbiocenosis in patients with cervical diseases and clinically healthy women

№	View biocenosis	1st group (background CD; n = 48)	2nd group (LSIL, HPV-; n = 19)	3rd group (LSIL, HPV +; n = 21)	4th group (HSIL; n = 17)	5th group (norm; n = 22)
1	Absolute normocenosis	23(47,9[a])	7(36,8[b,g])	8(36,4[c,h])	2(11,8[g,d,h])	15(68,2[a,b,d])
2	Conditional normocenosis	17(35,4[e])	36(31,6[g])	6(33,3[c,h])	3(17,6[g,d,h])	6(27,3)
3	Moderate dysbiosis	5(10,4[a])	2(10,5[b])	3(14,3[c])	3(17,6[d])	1(4,5[a,b,c,d])
4	Severe dysbiosis	3(6,3[a])	4 (21,1[c,h])	4(18,2[c,h])	9(53[g,d,h])	0(0)

Note. Here and in table 2: HPV- - HPV-negative, HPV + - HPV-positive; statistically significant differences: a - between the 1st and 5th groups, b - between the 2nd and 5th, c - between the 3rd and 5th, d - between the 4th and 5th, e - between 1st and 3rd, f - between 2nd and 3rd, g - between 2nd and 4th, h - between 3rd and 4th.

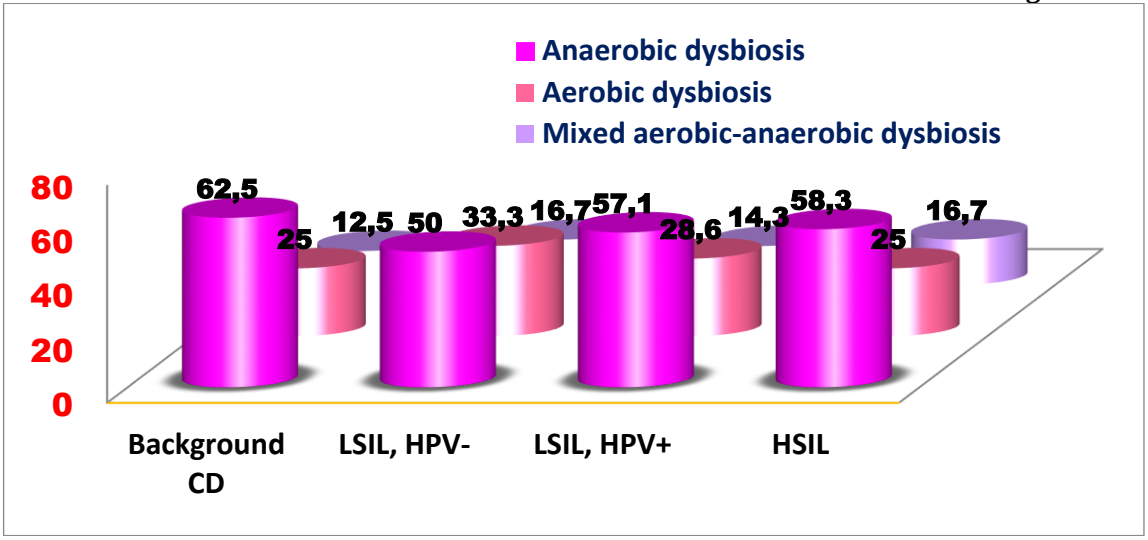
In the structure and moderate severe dysbiosis in patients of the 1st, 2nd, 3rd and 4th groups, anaerobic dysbiosis prevailed - 5 (62.5%), 3 (50%), 4 (57.1%) and 7 (58.3%) cases, respectively, aerobic dysbiosis was detected much less frequently - in 2 (25%), 2 (33.3%), 2 (28.6%) and 3 (25%) women, respectively. Mixed aerobic-anaerobic dysbiosis was diagnosed in 1 (12.5%), 1 (16.7%), 1 (14.3%) and 2 (16.7%) patients, respectively. The data obtained are in good agreement with the results of previous studies, indicating a correlation between CIN and BV (Diagram no. 2).

Table No. 2

The quantitative composition of the vaginal microbiocenosis in women with cervical diseases

№	Study title	1st group (background CD; n = 48)	2nd group (LSIL, HPV-; n = 19)	3rd group (LSIL, HPV +; n = 21)	4th group (HSIL; n = 17)	5th group (norm; n = 22)
	Material taking control	7,6	6,8	6,8	5,4	7,8
1	Total bacterial mass	7,4(7,2-7,7)[d,e]	7,6(7,2-7,9)[f,g]	7,7(7,4-8,2)[f,h]	8,0(7,4-8,4)[g,h]	7,8 (7,3–8,1)
	NORMOFLORA					
2	Lactobacillus spp.	7,4(6,8–7,8)[a,e]	7,5(6,8–7,8)[b,g]	7,6 (7,1–8,0)[h]	7,0(5,37,6)[d,g,,h]	7,8(7,3-8,1)[a,b,d]
	OPTIONAL ANAEROBIC MICROORGANISMS					
3	сем.Enterobacterium spp.	3,2 (2,7–3,5)	3,1 (2,6–3,4)	2,9 (2,5–3,3)	3,1 (2,6–3,5)[d]	2,8 (2,4–3,2)[d]
4	Streptococcus spp.	2,3 (1,6-3,5)[a]	2,6 (1,7–3,6)[b]	2,5 (1,9-3,7)[c]	2,5 (2,1–3,9)[d]	2,1 (1,6-3,1)[a,b,c,d]
5	Staphylococcus spp.	2,9 (2,3–3,6)	3,0 (2,0–3,5)	2,9 (2,3–3,6)	3,1 (2,3–4,0) [d]	2,9 (2,4–3,5) [d]
	OBLIGATE-ANAEROBIC MICROORGANISMS					
6	Gardnerella vaginalis + Prevotella bivia + Porphyromonas spp.	3,8 (3,5–5,6)[a]	4,5 (3,5–6,1)[b,g]	3,9 (3,2-6,2)[c,h]	7,2 (6,1–7,6)[d,g,h]	3,5 (3,1-4,3)[a,b,c,d]
7	Eubacterium spp.	4,5 (3,6–5,7)[a]	4,5 (3,6-5,7)[b,g]	4,4 (3,2–5,7)[c,h]	6,4 (5,5–6,8)[d,g,h]	3,6 (3,1–4,8)[a,b,c,d]
8	Sneathia spp. + Leptotrichia spp. + Fusobacterium spp.	2,1 (1,2–2,9)	2,1 (0–3,3)[h]	2,1 (1,4–2,9)[h]	3,4 (2,1–6,9)[d,g,h]	2,0 (1,3–2,6)[g]
9	Megasphaera spp. + Veillonella spp. + Dialister spp.	3,2 (2,5–4,7)[a]	3,5 (2,6–4,7)[b,g]	3,3 (2,3–4,8)[c,h]	5,8 (4,1-7,2)[d,g,h]	3,0 (2,3–3,8)[a,b,c,d]
10	Lachnobacterium spp./ Clostridium spp.	2,7 (2,2–3,8)[a]	3,4 (2,2–4,4)[b,g]	2,8 (2,2–4,1)[h]	4,1 (2,9–5,1) [d,g,h]	2,7 (2,2–3,5)[a,b,d]
11	Mobiluncus spp./ Corynebacterium spp.	3,1 (2,5–3,9)[a,e]	3,5 (2,8–4,1)[b,f,g]	3,1 (2,5–3,9)[f,h]	3,9 (3,0–4,6) [d,g,h]	3,1 (2,5–3,8) [a,b,d]
12	Peptostreptococcus spp.	3,3 (2,3–4,4)[a]	3,3 (2,3–4,4)[b,g]	2,9 (2,3–4,0)[c,h]	4,4 (2,9–6,2) [d,g,h]	2,6 (2,2–3,3)[a,b,c,d]
13	Atopobium vaginae	3,1 (2,6–5,1)[a,e]	3,1 (2,6–5,1)[b,f,g]	2,5 (2,0–3,8)[f,h]	6,6 (2,8–7,5) [d,g,h]	2,5 (2,0–3,1)[a,b,d]
	YEAST-LIKE MUSHROOMS					
14	Candida spp.	2,5 (2,3–3,9)	2,7 (2,4–3,3)	2,6 (2,4–2,9)	2,6 (2,4–3,0)	2,5 (2,3–2,8)
	MYCOPLASM					
15	Mycoplasma hммmominis	2,4 (2,2–3,9)	2,6 (2,4–3,3)	2,5 (2,3–2,9)	2,5 (2,3–3,0)	Not found
16	Ureaplasma spp.	3,0 (0,0–4,2)[a]	3,0 (0,0–4,2)[b]	1,7 (0,0–4,8)	2,9 (0,0–4,9)[d]	1,3 (0,0–3,8)[a,b,d]
	PATHOGENIC MICROORGANISMS					
17	Mycoplasma genitalium	Not found	Not found	Not found	Not found	Not found

Diagram no. 2



The level of the total bacterial mass of the vaginal biotope was the highest in the patients of the 4th group - 108 GE / ml, while the amount of lactoflora was statistically significantly lower than in the women of the comparison group (5thgroup) and patients of the 2nd and 3rd groups (table 2). In addition, in the patients of the 4th group, the absolute content of obligate and facultative anaerobes was statistically significantly higher compared to both the indicators of women in the 5th group (norm) and in the 2nd, 3rd groups. Particular attention should be paid to microorganisms of the groups Gardnerella vaginalis / Prevotella bivia / Porphyromonas spp., Atopobium vaginae, Eubacterium spp. and Megasphaera spp./Veillonella spp./Dialister spp., the number of which was 1000–10,000 times higher than that in patients of other groups and amounted to 107.2, 106.6, 106.4 and 105.8 GE / ml, respectively. The number of obligate anaerobes in patients of group 4 was also significantly increased compared to the norm (group 5), but the excess was within 10–100 times. There was also a statistically significant increase in the number of facultative anaerobes in patients with HSIL compared to the comparison group, however, the difference with similar indicators was moderate and did not exceed one order of magnitude (10 times).

Thus, the qualitative and quantitative composition of the vaginal microbiocenosis in patients with cervical dysplasia and in healthy women is significantly different. The data obtained indicate the relationship between the severity of dysplasia and the degree of dysbiosis in patients with cervical pathology: patients with HSIL are characterized by more pronounced changes in the number and composition of the vaginal microflora with the dominance of obligate anaerobes compared to LSIL. The question remains open, what is primary in this case: is the dysbiotic process in the vagina a factor contributing to the formation of more pronounced pathomorphological changes in the cervical epithelium infected with HPV, or the persistence of HPV creates a favorable background for the proliferation of opportunistic obligate anaerobic microflora in the vagina with the development of severe dysbiosis. Given the high frequency of dysbiotic disorders in patients with cervical pathology, especially with HSIL, it is advisable to conduct a comprehensive study of the vaginal microbiota and, if necessary, individual correction of dysbiosis in this category of patients.

Conclusions

1. The development of cervical intraepithelial neoplasias in HPV-negative women is accompanied by a violation of the vaginal microbiocenosis. In this case, both anaerobic and mixed aerobic-anaerobic dysbiosis are important. Among anaerobes, Gardnerella vaginalis was most often identified in association with Eubacterium spp., Megasphaera spp. and Mobiluncus spp.

2. HPV-associated cervical neoplasias are accompanied by the development of pronounced dysbiotic processes in the vagina with the predominant participation of obligate anaerobes. Among anaerobes, the most important are Gardnerella vaginalis in association with Atopobium vaginae, Megasphaera spp./Veillonella spp./Dialister spp. and Eubacterium spp.

3. The variety of leading pathogens dictates the need for a comprehensive study of the vaginal microbiocenosis in patients with precancerous pathology of the cervix, which allows, with high accuracy and specificity, to give a quantitative and qualitative assessment of the main participants in the studied biotope for the purpose of individual therapy.

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