REPUBLIC OF AZERBAIJAN

On the rights of the manuscript

ABSTRACT

of the dissertation for the degree of Doctor of Philosophy

ETIOLOGICAL STRUCTURE OF GENITOURINARY TRACT INFECTIONS CAUSED BY CONDITIONALLY PATHOGENIC MICROORGANISMS IN WOMEN

Specialty: 2414.01- Microbiology

Scientific field: Medicine

Applicant: **Matanat Burjali Aliyeva**

BAKU-2024

The dissertation work was performed at the Department of Medical Microbiology and Immunology of the Azerbaijan Medical University.

Scientific supervisor: Doctor of Medical Sciences, Prof.
Mehman Habib Aliyev

Official opponents: PhD in Medicine
Nigar Faig Mutallibova

PhD in Medicine
Rauf Fizuli Azizov

Doctor of Medical Sciences, Prof.
Sadraddin Abdulla Atakishizade

Dissertation Council FD 2.28 of the Supreme Attestation Commission under the President of the Republic of Azerbaijan operating at the Azerbaijan Medical University

Chairman of the Dissertation Council: Honored Scientist,
Doctor of Medical Sciences, Professor
İbadulla Aliagha Aghayev

Scientific secretary of the Dissertation Council: PhD in Medicine, Associate Professor
Shahla Murad Balayeva

Chairman of the Scientific Seminar: Doctor of Medical Sciences, Professor
Akif Irzakhan Gurbanov

İMZANI TƏSDİQ EDİRƏM
Azərbaycan Tibb Universitetinin
ELMİ KATİBİ
Tibb üzrə fəlsəfə doktoru, dosent
Rəşad Səmsəddin oğlu Talışınskiy
"12" 12 2024-ci il

GENERAL CHARACTERISTICS OF THE RESEARCH

Relevance of the topic: The vaginal microbiocenosis is a collection of microorganisms living in a specific biotope, usually consisting of lactobacilli. The state of the vaginal microbiocenosis is of great importance for reproductive health. Bacterial vaginosis (BV) and aerobic vaginitis (AV) are infectious, non-inflammatory diseases of the vaginal tract, which are caused by an imbalance between the physiological microflora of the vagina and conditionally pathogenic microflora[1].

Diseases of the genitourinary system are among the most common diseases in urological practice. Besides, the increase in the level of pathology occurs with a decrease in the quality of life and unfavorable environmental conditions[2].

A urinary tract infection is a condition in which microbes grow in the urinary tract, causing inflammation. The urinary tract includes the kidneys, ureters, bladder, and urethra. Urinary tract infections can occur in two forms: asymptomatic infection and symptomatic infection. Asymptomatic UTIs occur when bacteria remain persistently in a woman's urinary tract without any symptoms. Symptomatic UTIs are divided into two types, affecting the upper and lower urinary tracts[3].

---

1. Савичева, А.М. Бактериальный вагиноз и аэробный вагинит как основные нарушения баланса вагинальной микрофлоры. Особенности диагностики и терапии / А.М. Савичева, Н.И. Тапильская, Е.В. Шипицына [и др.] // Акушерство и Гинекология, – Москва: – 2017. № 5, – с. 24–31. DOI: https://dx.doi.org/10.18565/aig.2017.5.24-31
2. Дулов, А.А. Заболевания мочеполовой системы и обоснование организации оказания медицинской помощи больным на различных этапах: / автореферат дис. доктора медицинских наук. / – Воронеж, 2015. – 23 с.
3. Salari, N. Global prevalence of urinary tract infection in pregnant mothers: a systematic review and meta–analysis / N. Salari, Y. Khoshbakht, M. Hemmati [et al.] // Public Health, – 2023, 224, – p. 58–65/ DOI: 10.1016/j.puhe.2023.08.016

In recent years, the management of urinary tract infections has become increasingly difficult due to the emergence of multidrug-resistant pathogens. The use of narrow- and broad-spectrum antibiotics in treatment remains a topic of wide discussion in clinical trials[4].

BV is more common in females aged 15-24 (42%) than in older age groups (48-60%), but no significant association with age has been identified. The prevalence of BV is influenced by educational level, marital status, or hygienic aspects of behavior, such as daily changing of underwear and regular water procedures.

BV occurs in 51% of African-American women, 32% of Hispanic women, and 23% of Caucasians. However, these differences are largely due to the dietary and sexual behaviors of representatives of different races. In a study conducted among indigenous peoples living in the North, Khanty and Mansi, BV was found in only 7% of those surveyed. In recent years, an increase in urinary tract infections has been observed [5,6].

The human microbiome is very diverse and includes bacteria, fungi, protozoa, and viruses, having species that intensively colonize different tissue areas.

---

4. Əhmədzadə, C.İ. Sidik yolu infeksiyalarinda oral fosfomisinin istifadəsi: ədəbiyyata baxiş kəskin sistit müalicəsində urfocin´in © (fosfomycin) effektivliyi: şəxsi nəticəmiz / C.İ.Əhmədzadə, İ.S.Əhmədov // – Bakı: Urologiya və reproduktiv təbabət, – 2024. N 2, – s. 45–52.
5. Дикке, Г.Б. Бактериальный вагиноз: новые аспекты этиопатогенеза и выбора терапевтических стратегий. РМЖ // – Москва: Мать и дитя, – 2019. № 2, – с. 307–313. DOI: 10.32364/2618-8430-2019-2-4-307-313
6. Mancuso, G. Urinary Tract Infections: The Current Scenario and Future Prospects / G. Mancuso, A. Midiri, E. Gerace [et al.] // Pathogens, – 2023. 12 (4), – p. 1–17. doi: 10.3390/pathogens12040623

The nature of the symbiotic relationship between macroorganisms and mycoplasmas depends on the state of the body's immunobiological defense mechanisms and the pathogenic properties of the bacteria[7].

At the same time, mycoplasmas are carriers of a number of unique properties. In particular, in addition to participating in the formation of the inflammatory response, they can have an immunosuppressive effect, cause the synthesis of autoantibodies and autoimmune reactions, have pro- and/or antiapoptotic activity, provide genome instability, and cause chromosomal aberrations and transformations in the genetic apparatus of infected cells[8].

According to many authors, the main causative agents of UTIs are Escherichia coli (64.6%), Klebsiella spp. (9.5%), Enterococcus spp. (6.46%), and Staphylococcus spp. (5.1%). Depending on the nature of the infection, UTIs are divided into complicated and uncomplicated forms. Uncomplicated infections are observed in patients without obstructive uropathy and structural changes in the kidneys, as well as in patients without serious diseases. The high resistance of pathogens to antimicrobial drugs in the treatment of patients with recurrent UTIs causes certain difficulties[9].

---

7. Fierer, J. Nature and Pathogenicity of Micro–organisms / J. Fierer, D. Looney, J.C. Pechère // Infectious Diseases, – 2017. 4 (25), – p. 1–24. doi: 10.1016/B978-0-7020-6285-8.00002-2
8. Benedetti, F. Mycoplasmas–Host Interaction: Mechanisms of Inflammation and Association with Cellular Transformation / F.Benedetti, S. Curreli, D. Zella // Microorganisms, – 2020. 8 (9), – p. 1–21. doi: 10.3390/microorganisms8091351
9. Сурсякова, К.И. Некоторые эпидемиологические особенности инфекций мочевыводящих путей (обзор литературы) // К.И. Сурсякова, Т.В. Сафьянова // – Иркустк: Сибирский научный медицинский журнал, – 2017. №6, – с. 61–70.

The presence of microbial associations provides the most important adaptive abilities of microorganisms, leading to the strengthening of the pathogenicity of each microorganism in the association. The resistance factors of microorganisms in the mucous membrane of the genital tract include the prevention of the entry of antimicrobial substances into the body, the presence of metabolic activity and the high rate of division of individual microorganism cells in the population. Enzymes secreted by cells in the mucous membrane have the ability to degrade or modify antibiotics.

Sometimes, despite treatment, 30-50% of women with urinary tract infections experience recurrences within 6-12 months, and recurrent acute urinary tract infections negatively affect the quality of life of women of various reproductive ages, including premenopausal women[10,11].

Taking all this into account, it becomes clear that the study of bacterial vaginosis and infection of the urogenital tract with opportunistic pathogens in women is a relevant issue. In this regard, we considered it appropriate to conduct research to analyze the etiological structure of infection of the urogenital tract with opportunistic pathogens.

Purpose of the research: Microbiological evaluation of urogenital tract infections associated with opportunistic microorganisms.

10. Naber, K.G. RECAP group. Psychosocial burden of recurrent uncomplicated urinary tract infections / K.G. Naber, J. Tirán–Saucedo, F.M.E. Wagenlehner // GMS Infectious Diseases, – 2022. 24 (10), – p.1–9. DOI: 10.3205/id000078
11. Ennis, S.S. Premenopausal women with recurrent urinary tract infections have lower quality of life / S.S. Ennis, H. Guo, L. Raman [et al.] // International Journal of Urology, – 2018. 25 (7), – p. 684–689. DOI: 10.1111/iju.13698

Tasks of the research:

1. Study of the spread characteristics of various urogenital tract infections.
2. Evaluation of the nature and clinical course of chronic inflammatory processes in the urogenital tract of patients depending on the causative microorganisms.
3. Microbiological comparative assessment of the effectiveness of antibiotic resistance of various pathogens in mixed genitourinary tract infections.
4. Development of effective treatment methods by assessing the etiological and pathogenetic characteristics of the studied urogenital tract infections and the sensitivity of microorganisms.

Scientific novelty:

- The clinical microbiological characteristics of the spread of genitourinary tract inflammatory diseases associated with mixed infections have been studied.
- Complex optimal preventive and therapeutic measures aimed at anti-inflammatory and immune system restoration against the background of assessing the state of the microbiological system in patients have been developed.

Practical significance of the research:

1. Improvement of anti-inflammatory measures during various infectious processes of the genitourinary tract has created new opportunities for practicing doctors.
2. An extensive study of the genitourinary tract during infectious processes of various etiologies will increase the effectiveness of diagnostic measures.
3. Providing patients with immune therapy in addition to antibiotic therapy will create a foundation for increasing the effectiveness of treatment and preventive measures.

Main provisions of the dissertation presented for defense.

1. Anaerobic microorganisms have been found to cause abnormal vaginal discharge in addition to the involvement in the

bacterial complex that plays a role in the occurrence of infectious diseases of the genitourinary tract.

2. Extensive microbiological examinations play an important role in choosing treatment tactics for infectious pathologies of the genitourinary tract.
3. Complex treatment of bacterial vaginosis is carried out with the combined use of drugs that correct the microbiocenosis and affect the immune system.
4. Lactobacilli, which are the dominant bacteria in the local vaginal microbiocenosis, have the ability to colonize the mucous membrane and block the proliferation and reproduction of potentially pathogenic microorganisms in the vagina.

Publications. The results and fragments of the dissertation work are reflected in 5 theses and 8 articles.

Probation. The results of the research were discussed at the following conferences:

Current Problems of Medicine dedicated to the 100th anniversary of the Azerbaijan Democratic Republic, 2018. L International correspondence scientific and practical conference “International scientific review of the problems and prospects of modern science and education” (Boston. USA. October 22-23, 2018). V International Scientific Congress on "Modern Problems of Pharmacy", dedicated to the 90th anniversary of the establishment of the Azerbaijan Medical University and the 80th anniversary of Higher Pharmaceutical Education in Azerbaijan, 2021. The primary discussion of the dissertation was performed at the 18th meeting of the Department of Medical Microbiology and Immunology and the Department of Epidemiology of AMU on March 16, 2022. It was also discussed at the 14th scientific seminar of the BED 4.19 One-time Dissertation Council of AMU on May 31, 2024.

Structure and volume of the dissertation. The dissertation is 152 pages (203,851 characters) of computer typing and consists of Introduction (7,156 characters), Literature review (61,280 characters), Personal research and discussion (11,805 characters),

Chapter III (85,241 characters), Chapter IV (19,411 characters), Results (17,830 characters), Conclusions (1,070 characters), Practical recommendations (358 characters), and List of references. The list of references consists of 169 sources. The dissertation includes 15 tables and 15 diagrams.

MATERIALS AND METHODS OF THE RESEARCH

The study included 110 patients and 22 healthy women who applied for routine examinations at the Ganja City Women's Clinic. Out of 110 patients, 20 were diagnosed with chronic recurrent genital candidiasis, 29 with recurrent bacterial vaginosis, and 39 with both chronic recurrent genital candidiasis and bacterial vaginosis. In the first stage of the study, the number of annual recurrences of the corresponding diseases in the patients registered in 2017 (defined as 10-12 times, 6-9 times, and less than 6 times per year) was calculated in number and % for each group of patients. In the second stage of the study, the incidence and percentage of normomenorrhea, oligomenorrhea, hypomenorrhea, hypermenorrhea, algodysmenorrhea, and premenstrual syndrome were studied in each group of patients (groups composed of 20 patients with chronic recurrent genital candidiasis, 29 patients with recurrent bacterial vaginosis, and 39 patients with both chronic recurrent genital candidiasis and recurrent bacterial vaginosis) and a group of healthy individuals (22 persons).

The occurrence and colony-forming ability of S. epidermidis, S. aureus, E. coli, Enterococci, S. Saprophyticus, and C. albicans microorganisms in vaginal smears and vaginal fluid samples of the affected groups (39 patients – Group III, 29 patients - Group II, 20 patients – Group I), and 22 healthy women (Control group) who applied to the Ganja City Women's Counseling Center were determined and expressed in number and %, CFU. Both the patient and control groups included women in the age range of 30-35. The reason we chose such a narrow age range is to minimize the effects of age on the results of the study.

To compare the colonization capacity of various opportunistic microorganisms in vaginal smears during the complex application of immunotherapy and probiotic therapy in patients suffering from both recurrent genital candidiasis and bacterial vaginosis, women were included in the study, and it was found that the mentioned diseases were caused by mixed infection of microorganisms such as U.urealyticim, U.parvum, M.hominis, G.vaginalis, A.vaginae, and C.albicans. Then, the colonization indicators of conditionally pathogenic microorganisms, such as Enterococci, Streptococci, Staphylococci, Mycoplasma hominis, Ureaplasmas, and Candida species in vaginal smear samples taken from both groups of women (sick and healthy) when they visited the hospital were studied using CFU (Colony Forming Unit). Women with diseases were treated with Metronidazole for 7 days, according to the regimen recommended by the World Health Organization's Center for Disease Control and Prevention. After the end of treatment and the disappearance of clinical symptoms, their vaginal smears were again studied for the CFU indicators of the above-mentioned opportunistic microorganisms. Microbiological studies were carried out in the laboratory operating at the aforementioned clinic. The study was continued during the two subsequent relapses of the diseases of these patients who were taken under control. Thus, when the patients recovered after the “basis” treatment (which included Bactistatin and Sambovin) and after a certain period of time, the disease relapsed, we again qualitatively analyzed their vaginal smears for the specified conditionally pathogenic microorganisms.

The composition of Bactistatin is as follows:

- sterile culture medium of the Bacillus subtilis species -200 mg
- natural enterosorbent (zeolite) -195 mg
- soy flour hydrolyzate -100 mg
- calcium stearate or aerosil -5 mg

The sterilized culture solution contains biologically active substances consisting of natural microproducts of the microorganism - Bacillus subtilis (lysozyme, bacteriocins, catalases, etc.).

In addition to improving the functional state of the gastrointestinal system, as well as its positive properties, such as removing toxic substances from the body, etc., bactistatin can also be used in the following pathologies:

- Acute and chronic diseases of the gastrointestinal tract accompanied by dysbacteriosis,
- As part of complex therapy in cases accompanied by a violation of the microflora of various organs and systems,
- As a component of complex therapy in mycoses,
- In combined therapy for allergic diseases (eczema, etc.),
- As part of combined treatment in dermatological diseases (neurodermatitis, psoriasis, etc.),
- In combined treatment when poisoning with toxins and heavy metal salts,
- During exhaustion, etc.

The preparatory measures for taking a smear are as follows:

- Administration of antibiotics or antifungals is stopped 2 weeks before the examination. If it is impossible to stop these drugs, the doctor taking the smear is informed.
- The patient should not have sexual intercourse for 3 days before the examination.
- Vaginal suppositories, tablets, creams, and other local therapeutic agents should not be used 2 days before the examination.
- The vagina should not be douched before the examination, but it is possible to wash the perineum with ordinary warm water and simple soap.

The swab is placed on the visible part of the inflammation or, if it is not there, on the posterior arch of the vagina. The material is evenly transferred to a glass slide, air-dried, fixed with ethanol (2-3 drops per glass), marked with a marker, placed in a closed container, and sent to the laboratory. During bacteriological examination, the swab is placed in a test tube and immediately sent to the laboratory. Although bacterial vaginosis is diagnosed only by native smear, its cultivation is necessary for the multiplication of *Gardnerella*

vaginalis. For this purpose, the material taken from the vagina is cultured in a special nutrient medium, Colombia agar, and incubated for 48 hours in a 5-10% CO_2 medium. Preparations are prepared by the Gram method from *G. vaginalis* colonies that give beta hemolysis in 48 hours on Colombia agar prepared with human blood. Gram-negative coccobacilli and hippurate-hydrolyzing bacteria are identified as *G. vaginalis*.

In patients suspected of having soft chancre (chancroid), the culture of H. ducreyi is obtained on chocolate agar with and without antibiotics. These bacteria require 5-10% CO_2 and factor X for growth. They grow at 35^0C for 3-9 days.

When a fungal infection is suspected, Saburo agar is used for cultivation. Gram stains are prepared from the culture medium in which growth is observed. When Gram-positive, budding yeast-like cells are detected, a germ tube test is performed to differentiate *Candida albicans* from other *Candida* species. Thus, the majority of yeasts with a positive germ tube test are *C. albicans*.

Cultivation is a sensitive method for *T.vaginalis*. Diamond's medium is used for this purpose. The result is obtained within 3-4 days. The culture medium is incubated at 37^0C for 1-2 hours, then cultivation is performed. A sample of genital secretions taken with a swab is placed in the culture medium and shaken well. A urine sample is centrifuged and then cultured from the sediment. It is incubated at 37^0C. It is necessary to wait 96 hours to report a negative result.

The following criteria are used to determine the degree of contamination with microorganisms:

- when about 10 microbes are detected in the field of view – minimal (+);
- 11-100 cells – moderate (++);
- 100-1000 cells – large quantities (+++);
- more than 1000 cells – massive quantities (++++).

A qualitative analysis is also performed to determine what types of microorganisms are present in the smear. Therefore, it is stained in various ways - Gram or Romanowsky-Giemsa methods.

The number of the detected microorganisms has been noted in the summary.

A phenotypic test is used to detect induced resistance in *S. aureus* strains. This is also done in the form of two discs. The examined *S.aureus* strain is inoculated and a 2 μg antibiotic disc and a 15 μg erythromycin disc are placed on Muller Hinton agar medium approximately 15 mm apart. Incubation is performed in the thermostat at 37°C, for one day, and then assessed. When induced resistance is determined by the examination of S.aureus strains, the sterile zone in the part facing the erythromycin disc is reduced, and this area takes on a shape similar to the letter D. The synthesis of extended-spectrum beta-lactamase in E.coli strains is determined by a phenotypic test - using two discs.

For this purpose, an amoxicillin+clavulanic acid disc was placed directly next to the antibiotic disc on the surface of the solid nutrient medium in which the bacterial strain under examination was inoculated. The result was evaluated after one day of incubation in a thermostat at 37^0C. If the bacterial strain is synthesized, the sterile zone around the antibiotic disc extends towards the disc. The synthesis of the inducible beta-lactamase enzyme in Pseudomonas strains was determined by a phenotypic test - using two discs.

Screening of the antimicrobial activity of the studied antibiotics against conditionally pathogenic Gram-positive and Gram-negative bacteria and yeasts was carried out by the agar diffusion method. General methods accepted in microbiology were used to inoculate microbial cultures and maintain working cultures. The resazurin staining method was used to determine the minimum inhibitory concentration of substances against Gram-positive and Gram-negative bacteria. The effect of the substance, which shows high antimicrobial properties against Gram-positive and Gram-negative conditionally pathogenic bacteria, on the development and reproduction (growth curve) of bacteria was studied in a liquid nutrient medium. In this case, the amount of cells was determined by optical density using a photoelectric colorimeter. The bacteriostatic

and bactericidal effect of the substances was determined based on the time of cell death in a liquid nutrient medium.

The obtained numerical data were processed using statistical methods taking into account modern requirements. The mean values (M) for group indicators, their standard error (m), the minimum (min) and maximum (max) values of the series, as well as the frequency of occurrence of quality indicators in groups were determined.

The parametric Student's t-test method was used for the primary assessment of differences in the variational series. Then, the obtained quantitative indicators are checked and refined. For this, one of the non-parametric criteria - Wilcoxon (Mann-Whitney) U-criterion is applied. Correlation analysis is carried out in order to determine the strength of the relationship between the studied indicators. A computer program was used for statistical processing of the results obtained during the study. The obtained quantitative indicators are displayed in tables and diagrams.

The results of the research were processed using standard methods of variation statistics by the programs "Microsoft Excel 2010", "BioStat 6.0", and "Statistica 10.0". The normality of the distribution of variables was checked using the Kolmogorov-Smirnov criterion. All continuous variables were presented as the mean and standard error of the mean $M \pm m$, as well as the width of the 95% confidence interval (95% CI), and categorical variables were expressed as numbers (in percentages). Due to the prevalence of parameters with a non-normal distribution, statistical analysis of study results comparing groups on quantitative parameters was performed using non-parametric criteria of analysis: two independent groups were compared using the Mann-Whitney U test.

Spearman's rank correlation coefficient (r) was used to analyze the relationship between variables (indicators). Based on the examination of the strength indicators of the relationship between the correlation coefficients and the determination of the dependency of one trait on the change in another (regression coefficient), the quantitative characteristics of the dependence of the examined traits

were provided. Differences between the compared variation orders were considered statistically significant at the p <0.05 level.

RESULTS OF THE PERSONAL RESEARCH

Out of a total of 110 people who applied to the Women's Clinic – in a group of 20 women with chronic recurrent genital candidiasis, nine patients experienced 10–12 recurrences in 2017, which covered 45.0±11.12% of the relevant group. However, 17 out of 29 patients with recurrent bacterial vaginosis, 58.6±9.15% of the group, and 14 out of 39 people with both chronic recurrent genital candidiasis and bacterial vaginosis, 35.9±7.68% of the group, experienced 10–12 recurrence of the disease during the mentioned year. Seven individuals with a diagnosis of chronic recurrent genital candidiasis experienced 6-9 recurrences of the disease during the year, accounting for 35.0±10.67% of the corresponding group of patients (Table 1). Seven patients in the group with recurrent bacterial vaginosis, comprising 24.1±7.95% of patients with vaginosis, and 21 patients with both chronic recurrent genital candidiasis and bacterial vaginosis, comprising 35.9±7.68% of the corresponding group, experienced relapses during this time interval.

Table 1.

Characteristics of disease recurrence in women with genital candidiasis and bacterial vaginosis

Number of relapses per year	Group I, n=20		Group II, n=29		Group III, n=39	
	n	%	n	%	n	%
10–12	9	45.0±11.12	17	58.6±9.15	14	35.9±7.68
6–9	7	35.0±10.67	7	24.1±7.95	21	53.8±7.98
<6	4	20.0±8.94	5	17.2±7.01	4	10.3±4.86

Four patients in the group of 20 patients with chronic recurrent genital candidiasis, five in the group of 29 women with recurrent bacterial vaginosis, and four in the group of 39 women with both chronic recurrent genital candidiasis and recurrent vaginosis experienced less than six recurrences annually. These cases accounted for 20.0±8.94%, 17.2±7.01%, and 10.3±4.86%, respectively, in the corresponding groups. When analyzing the numerical indicators, we observed that the most undesirable situation in terms of the number of relapses of the disease was in women with recurrent bacterial vaginosis (58.6±9.15% of cases, 10-12 relapses per year). In a group of 39 patients suffering from both chronic recurrent genital candidiasis and bacterial vaginosis, thyroid disease was observed in 9 patients (23.1% of the group) and gastritis in 7 patients (17.9% of the group). In this group, cholecystitis was observed in 8 patients, intestinal dysbacteriosis in another 8 patients, and cystitis in 16 individuals, which accounted for 20.5%, 20.5%, and 41% of the corresponding group, respectively. The study revealed that incidences of various somatic pathologies and menstrual cycle disorders in patients with problems in the genital tract were relatively higher than in healthy women. During the study of quantitative indicators of various microorganism types in vaginal smears of patients with bacterial urogenital infections, in a group of 39 women suffering from both chronic recurrent genital candidiasis and bacterial vaginosis, the S. epidermidis microorganism was found in 16 vaginal smear samples, which was 41% in the group. In this group, the S. aureus microorganism was found in 8 biological samples, which was 20.5% of this group. The E. coli microorganism was found in 13 samples, which was 33.3% of this group (Table 2).

In this group, Enterococcus species were found in 3 vaginal smear samples accounting for 7.69% of the group. S.saprophyticus microorganisms in vaginal smear samples in this group occurred also in 3 samples, covering 7.69%. Candida fungi were found in 16 samples and covered 41.03% of the group.

Table 2.

Incidences of various microorganisms in vaginal smears of patients with mixed etiology of bacterial urogenital infections

Microorganisms	Indicators	Control n = 22	Women with mixed genital infections		
			Group III, n=39	Group II, n=29	Group I, n=20
S. epidermidis	n	4	16	10	6
	%	18.2	41.0	34.5	30.0
	p		> 0.05	> 0.05	> 0.05
S. aureus	n	0	8	5	5
	%	0.0	20.5	17.2	25.0
	p		<0.05	<0.05	<0.05
E. coli	n	0	13	8	10
	%	0.0	33.3	27.59	50.00
	p		<0.01	<0.01	<0.001
Enterococcus spp.	n	1	3	2	1
	%	4.55	7.69	6.90	5.00
	p		> 0.05	> 0.05	> 0.05
S.saprophyticus	n	1	3	2	1
	%	4.55	7.69	6.90	5.00
	p		> 0.05	> 0.05	> 0.05
Candida albicans	n	0	16	1	1
	%	0.0	41.03	3.45	5.00
	p		<0.001	> 0.05	> 0.05

Note: *p – statistical significance according to the control group.*

The study of quantitative indicators of various microorganism types in vaginal smears of patients with mixed etiology of bacterial urogenital infections revealed the occurrence of S. epidermidis species in 10 biological samples taken from a group of 29 women suffering from bacterial vaginosis, which covered 34.5% of the group. In these patients, the S.aureus species was observed in 5 of the biological samples, the E.coli species in 8 samples, Enterococci in 2 samples, S.saprophyticus in 2 samples, and Candida strains in only 1 sample, which is accounted for 17.5%, 27.59%, 6.9%, 6.9%, and 3.45% of the women in the respective groups, suffering from this nosological unit.

Quantitative indicators of various microorganism types were studied in vaginal smears of patients with mixed etiology of bacterial

urogenital infections during associations with various microorganisms. In biological material samples taken from a group of 20 women diagnosed with chronic recurrent genital candidiasis, S. epidermidis was found in 6 smear samples (30% of the total group), S. aureus in 5 samples (25%), E. coli in 10 samples (50%), and Enterococcus species, S. saprophyticus species, and C. albicans fungi were found in only 1 sample each (5% of the total samples in the group).

During the microbiological examination of vaginal smear samples taken from 22 women who were accepted as a control group and who were not diagnosed with any genital infection and simply applied to the antenatal clinic for routine prophylactic examinations, S. epidermidis was found in 4 samples or 18.2% of the corresponding group, and Enterococci and S. saprophyticus were found in 1 sample each, accounting for 4.55% of each group. However, Candida albicans, E. coli, and S. aureus were not found in the biological samples of these women. The qualitative indicators of various microorganism types in vaginal smears of patients with mixed etiology of bacterial urogenital infections were studied in association with various opportunistic microorganisms. In a group of 39 women suffering from both chronic recurrent genital candidiasis and bacterial vaginosis, the colony-forming units of S. epidermidis, S. aureus, E. coli, Enterococci, S. saprophyticus, and C. albicans were 7.88±0.07 CFU, 2.22±0.16 CFU, 4.11±0.12 CFU, 0.68±0.04 CFU, 2.02±0.34 CFU, and 4.34±0.23 CFU, respectively (Table 3).

In vaginal smear samples taken from the group of 29 women with recurrent bacterial vaginosis, the average CFU of microorganisms such as S. epidermidis, S. aureus, E. coli, Enterococci, and S. saprophyticus were 3.08±0.11, 2.09±0.12, 3.14±0.2, 1.03±0.17, and 2.20±0.45, respectively, while Candida fungi were not found in vaginal smears. In a group of 20 women suffering from chronic recurrent genital candidiasis, the average group CFU of S. epidermidis, S. aureus, E. coli, Enterococci, S. saprophyticus, and C. albicans species was 6.06±0.20, 7.86±0.31, 5.46±0.35, 0.74±0.08, 2.27±0.17, and 1.00±0.12, respectively.

Table 3.

Colony-forming unit (CFU) of various microorganism species in patients with mixed etiology of bacterial urogenital infections

Microorganisms	CFU	Control n = 22	Women with mixed genital infections		
			Group III, n=39	Group II, n=29	Group I, n=20
S. epidermidis	M±m	1.04±0.044	7.88±0.07	3.08±0.11	6.06±0.20
	p		<0.001	<0.001	<0.05
S. aureus	M±m	-	2.22±0.16	2.09±0.12	7.86±0.31
	p		<0.001	<0.001	<0.05
E. coli	M±m	-	4.11±0.12	3.14±0.20	5.46±0.35
	p		-	-	-
Enterococcus spp.	M±m	0.026±0.006	0.68±0.04	1.03±0.17	0.74±0.08
	p		-	-	-
S.saprophyticus	M±m	0.012	2.02±0.34	2.20±0.45	2.27±0.17
	p		-	-	-
Candida albicans	M±m	-	4.34±0.23	-	1.00±0.12
	p		<0.001	-	-

Note: *p – statistical significance according to the control group.*

In a group of 22 healthy women without any genital problems, when studying the colonization abilities of the above-mentioned microorganisms, the colony-forming units of S. epidermidis, Enterococci, and S. saprophyticus were 1.04±0.044, 0.026±0.006 and 0.012, respectively, while the corresponding indicators of S. aureus, E. coli and C. albicans were 0. The qualitative characteristics of lactobacilli and bifidobacteria (with CFU) and the nature of vaginal discharge were studied in vaginal fluid samples from patients with mixed-etiology of bacterial

urogenital infections during associations with various opportunistic microorganisms. While the colonization capacity of lactobacteria in a group of 39 women suffering from both chronic recurrent genital candidiasis and bacterial vaginosis was 3.78±0.19 CFU, in a group of 29 women suffering from recurrent bacterial vaginosis it was 1.17±0.16 CFU, and in a group of 20 women suffering from chronic recurrent genital candidiasis, the average value of this indicator was estimated to be 3.08±0.27 CFU. In the group of 22 women who visited the women's clinic for a routine preventive examination, which we accepted as a control group, this indicator was found to be 6.07±0.21 CFU on average. As a result of the study of the colonization ability of lactobacilli, the highest indicator was recorded in the healthy women who made up the control group, and the lowest in the group of patients with vaginosis. The average colony-forming ability of bifidobacteria in the group of women suffering from both bacterial vaginosis and chronic recurrent genital candidiasis was found to be 1.87±0.08 CFU, in the group of 29 women suffering from recurrent bacterial vaginosis, it was 0.31±0.03 CFU, and in the group of women suffering from chronic recurrent genital candidiasis, it was 1.24±0.05 CFU.

In the second phase, the nature of the vaginal discharge of patients with mixed etiology of bacterial urogenital infections was studied in association with various opportunistic microorganisms. In a group of 39 women suffering from both chronic recurrent genital candidiasis and bacterial vaginosis, white vaginal discharge was recorded in 18 women, which accounts for 46.2% of the corresponding group. In this group, mucous vaginal secretion was observed in 20 patients, accounting for 51.3% of the group. Purulent vaginal discharge was detected in 4 patients, accounting for 10.3% of the group (Table 3.9). Yellow secretion was observed in 6 patients, accounting for 15.4% of the group. No cases of absence of vaginal discharge were recorded in this patient group.

In a group of 29 women with recurrent bacterial vaginosis, 12 women complained of white vaginal secretion, which accounted for 41.4% of the group. In this group, mucous vaginal secretion was observed in 11 women, which accounted for 37.9% of the group. In 2 patients in the group, that is, 6.9% of the group, purulent vaginal discharge was recorded. Yellow vaginal fluid was recorded in 5 patients in this group, covering 17.2% of the corresponding group. Vaginal discharge was recorded in all patients in this group.

In a group of 20 patients suffering from chronic recurrent genital candidiasis, cases of white vaginal discharge were observed in 4 women, which covered 20% of the corresponding group. Mucous vaginal discharge was observed in 12 women in this group, which covered 60% of the group. Purulent vaginal discharge was observed in 5 people in the group, which accounted for 25% of the group. In this group, yellow vaginal discharge was also recorded in 5 people (25%). In this group, no vaginal discharge was observed. Of the 22 healthy women we accepted as the control group, only 1 (4.5%) had vaginal discharge, which was white. In the remaining 21 women (95.5%), no vaginal discharge was observed.

Although the average group CFU for Streptococcus species in the group of healthy women was calculated to be 1.13±0.082, in the biological samples taken before treatment in the group of 39 women suffering from both bacterial vaginosis and recurrent genital candidiasis, this indicator was found to be 4.17±0.090 CFU.

During the study, the average colonization capacity of streptococcal species in the group of healthy women was calculated to be 1.13±0.082 CFU. However, in the biological samples taken before treatment in a group of 39 women suffering from both bacterial vaginosis and recurrent genital candidiasis, this indicator was found to be 4.17±0.090 CFU. The treatment of these patients was accepted as a “basis”, and after the measures detailed above were implemented, the average CFU of

streptococci in vaginal smear samples was calculated to be 3.92±0.103 for the group. After the patients were treated with "basis" treatment + bactistatin (probiotic), the CFU of streptococci was slightly lower, 3.66±0.107, in vaginal smear samples. After the next relapse of the patients included in this group, for the patients treated according to the "basis" treatment + bactistatin (probiotic) + sambovin (immunomodulator) scheme, the qualitative indicator of the mentioned opportunistic microorganism in the biological samples was calculated to be 2.86±0.093 CFU, which is closer to the corresponding indicators of healthy individuals compared to the indicators resulting from other treatment measures. While the average quality index of staphylococcal species in vaginal smear samples obtained from a group of 22 healthy women involved in the study was calculated to be 2.38±0.109 CFU, the average quality index of the microorganism species in vaginal smear samples taken from a group of 39 women suffering from both recurrent chronic genital candidiasis and bacterial vaginosis was calculated to be 5.36±0.138 CFU.

After the basis treatment was applied in patients, this indicator was slightly higher, 6.42±0.229 CFU, than the indicator before treatment. When the next relapse occurred after the disease was cured, the treatment according to the "basis" + bactistatin (probiotic) scheme was applied and this indicator was slightly lower than the indicator before treatment, 5.15±0.295 CFU.

During relapses after patients recovered from treatment with a combination of "basis" treatment and parabiotics, treatment was performed according to the "basis" treatment + bactistatin + sambovin scheme. Then after the patients were recovered, vaginal swab samples were taken again and analyzed for the mentioned opportunistic pathogen, and as a result, the CFU of that microorganism was found to be lower, 4.17±0.234, than that of after other treatment measures.

Table 4.

Colony-forming unit (CFU) of various opportunistic microorganisms in vaginal smears of patients in Group III

Microorganisms	Indicators	Healthy women (n=22)	Before treatment	Treatment measures		
				Basis treatment	Basis+bactistatin +sambovin	Basis +bactistatin
Enterobac-teriaceae	M±m	2.06±0.069	5.14±0.086	4.82±0.133	3.11±0.088	4.54±0.112
	p			$p_1 > 0.05$	$p_1 < 0.001$ $p_2 < 0.001$	$p_1 < 0.001$ $p_2 > 0.05$
Strepto-coccus	M±m	1.13±0.082	4.17±0.090	3.92±0.103	2.86±0.093	3.66±0.107
	p			$p_1 > 0.05$	$p_1 < 0.001$ $p_2 < 0.001$	$p_1 < 0.01$ $p_2 > 0.05$
Staphylo-coccus	M±m	2.38±0.109	5.36±0.138	6.42±0.229	4.17±0.234	5.15±0.295
	p			$p_1 < 0.01$	$p_1 < 0.01$ $p_2 < 0.001$	$p_1 > 0.05$ $p_2 < 0.01$
Candida albicans	M±m	0.30±0.080	5.38±0.119	4.25±0.214	2.28±0.235	4.26±0.270
	p			$p_1 < 0.05$	$p_1 < 0.01$ $p_2 < 0.01$	$p_1 < 0.05$ $p_2 > 0.05$

Note: *p1 – statistical significance according to pre-treatment data, p2 – statistical significance according to "basis" treatment.*

The recorded CFU for Candida albicans in vaginal smear samples taken from 22 women who visited the women's clinic for periodic preventive examinations was 0.30±0.08. In a study of 39 patients suffering from both bacterial vaginosis and recurrent genital candidiasis, this indicator was found to be ten times higher, 5.38±0.119 CFU, than that of healthy women, before any treatment was performed. After the "basis" treatment was applied, the average CFU for the fungal species in the biological samples was found to be 4.25±0.214 per group, which is significantly lower than the indicator before treatment. In the patients who recovered with the help of "basis" treatment, but later relapsed, the colony-forming ability of C. albicans in biological samples obtained after treatment with the "basis" treatment + bactistatin scheme was almost at the same level

as the corresponding indicator in women who recovered with only "basis" treatment, 4.26±0.270 CFU. However, the corresponding indicator was calculated to be significantly lower, 2.28±0.235 CFU, as a result of treatment of patients with the "basis" treatment + bactistatin + sambovin regimen during their next relapse. According to the results, those with bacterial vaginosis and recurrent vaginal candidiasis would benefit more from using probiotics and immunological preparations in addition to generally accepted treatment measures.

The average scores of the indices were compared with those of other illnesses, such as asthma and chronic obstructive pulmonary disease, in order to examine the quality of life of women who had chronic infections. The study revealed that these women have a worsening psycho-emotional state, low self-esteem, social and physical activity. Deterioration of the vaginal microflora, the presence of discharge and bad odor cause discomfort, and a feeling of shame in front of their sexual partner. Thus, when studying the diagnostic markers of bacterial vaginitis using traditional and modern examination methods, it becomes clear that for the correct diagnosis, it is necessary to start with the determination of clinical and anamnestic indicators. Although microscopic and microbiological examinations are considered the gold standard for the examination of BV, new molecular examinations also attract attention as they are more effective and convenient. It became evident from the literature review that identifying the main clinical symptoms is also crucial for BV evaluation.

To study the sensitivity of microorganisms to various antibiotics in biological samples taken from patients suffering from both bacterial vaginosis and chronic recurrent genital candidiasis, smear samples were sent to the microbiology laboratory of the same medical institution. In these samples, the sensitivity of E. coli, Staphylococcus spp., Enterococcus faecalis, Ureaplasma urealyticum, and Mycoplasma hominis species to antibacterial drugs such as Azithromycin, Macropen, Amoxicillin, Cefataxime, Ceftriaxone, Ampicillin, Oxacillin, Doxycycline, Amikacin, and

Gentamicin was determined by applying antibiotic "disks" in terms of number of incidences and percentage.

As a result of the study, sensitivity to Azithromycin was recorded in 77 out of 88 samples, which is 87.5±3.53% of the total samples. The sensitivity of E. coli to Amoxicillin was recorded in 42 samples, to Cefotaxime in 21 samples, to Oxacillin in 5 samples, to Doxocillin in 20 samples, to Amikacin in 9 samples, and to Gentamicin in 45 samples, which accounted for 47.7±5.32%, 23.9±4.54%, 5.7±2.47%, 22.7±4.47%, 10.2±3.23%, and 51.1±5.33% of the total samples, respectively. However, no significant susceptibility of E. coli to Macropen, Ceftriaxone, and Ampicillin was observed in any sample (Table 5).

As a result of the study, 55 out of 88 samples (62.5±5.16% of the total samples) showed sensitivity of Staphylococci to Azithromycin, 75 samples (85.2±3.78%) to Amoxicillin, 3 samples (3.4±1.93%) to Cefotaxime, 25 samples (28.4±4.81%) to Doxycycline, and 19 samples (21.6±4.39%) to Gentamicin. Staphylococci were not sensitive to Macropen, Ceftriaxone, Ampicillin, Oxacillin, and Amikacin in the samples.

As a result of the study, Streptococci were found to be susceptible to Azithromycin in 70 samples, which accounts for 79.5±4.30% of the total samples. The susceptibility of this microorganism to Amoxicillin was found in 83 samples, to Cefotaxime and Ceftriaxone in 5 samples each, and to Gentamicin in 10 samples, which accounted for 94.3±2.47%, 5.7±2.47%, 5.7±2.47% and 11.4±3.38% of the total samples, respectively. Streptococci were not found to be susceptible to Macropen, Ampicillin, Oxacillin, Doxycycline, and Amikacin in any of the samples.

Enterococcus faecalis was found to be susceptible to Azithromycin in 8 samples, to Amoxicillin in 7 samples, to Doxocillin in 4 samples, and to Gentamicin in 8 samples, which accounted for 9.1±3.06%, 8.0±2.88%, 4.5±2.22%, and 9.1±3.06% of the total samples, respectively. None of the total samples showed

significant susceptibility of Enterococcus faecalis to Macropen, Cefotaxime, Ceftriaxone, Ampicillin, Oxacillin, and Amikacin.

Table 5

Sensitivity of microorganisms to various antibiotics in biological samples taken from patients

Preparation	E.coli	Staphylococcus spp.	Streptococcus spp.	Enterococcus faecalis
Azithromycin	77 87.5±3.53%	55 62.5±5.16%	70 79.5±4.30%	8 9.1±3.06%
Makropen	0	0	0	0
Amoxicillin	42 47.7±5.32%	75 85.2±3.78%	83 94.3±2.47%	7 8.0±2.88%
Cefotaxime	21 23.9±4.54%	3 3.4±1.93%	5 5.7±2.47%	0
Ceftriaxone	0	0	5 5.7±2.47%	0
Ampicillin	0	0	0	0
Oxacillin	5 5.7±2.47%	0	0	0
Doxycycline	20 22.7±4.47%	25 28.4±4.81%	0	4 4.5±2.22%
Amikacin	9 10.2±3.23%	0	0	0
Gentamicin	45 51.1±5.33%	19 21.6±4.39%	10 11.4±3.38%	8 9.1±3.06%

As a result of the study, the sensitivity of Ureaplasma urealyticum to Azithromycin was recorded in 51 samples, sensitivity to Macropen in 14 samples, and sensitivity to Doxocillin in 63 samples, which is 58.0±5.26%, 15.9±3.90%, and 71.6±4.81% of the total samples, respectively. The sensitivity of Ureaplasma urealyticum to Amoxicillin, Cefotaxime, Ceftriaxone, Ampicillin, Oxacillin, Amikacin, and Gentamicin was not recorded.

CONCLUSIONS

1. As a result of the study, in a group of 20 women with chronic recurrent genital candidiasis, cases of 10-12 recurrences of the disease during 2017 were recorded in 9 patients, which is 45.0±11.12% of the corresponding group.
2. During the microbiological examination of vaginal smear samples taken from 22 women who were accepted as a control group and who were not diagnosed with any genital infection, S. epidermidis was found in 4 samples, in 18.2% of the corresponding group, and Enterococci and S. saprophyticus were found in 1 sample each, each accounting for 4.55% of the group. However, Candida albicans, E. coli, and S. aureus were not found in the biological samples of these women.
3. No significant sensitivity of E. coli to Macropen, Ceftriaxone, and Ampicillin; Staphylococci to Macropen, Ceftriaxone, Ampicillin, Oxacillin, and Amikacin; and Enterococcus faecalis to Macropen, Cefotaxime, Ceftriaxone, Ampicillin, Oxacillin and Amikacin was recorded in any sample.
4. The results obtained showed that it is more beneficial to apply probiotics and immune preparations in addition to generally accepted treatment measures in individuals suffering from both bacterial vaginosis and recurrent genital candidiasis.

PRACTICAL RECOMMENDATIONS

1. Based on the obtained data, a basis has been established in medical practice for conducting more correctly directed microbiological examinations in cases of chronic microbiocenosis pathologies of various types in genital tracts.
2. In addition to the gold standard, microscopic and microbiological examinations, identification of the main symptoms may also be of particular importance in the diagnosis of BV.

3. More effective results can be achieved when treating vaginosis clinically by providing medications based on the “basis” treatment + bactistatin + sambovin scheme.

Publications

1. Bakterial vaginozun mikrobiologiyasına müasir yanaşmalar. //Sağlamlıq, № 5, 2017, səh.25-29/M.B.Əliyeva
2. Müxtəlif mikroorqanizmlərlə assosasiyalar zamanı qarışıq etiologiyalı bakterial urogenital infeksiyalı xəstələrdə müxtəlif mikroorqanizm növlərinin kəmiyyət və keyfiyyət göstəriciləri.//Azərbaycan Təbabətinin Müasir Nailiyyətləri, № 3, 2018, səh.148-152/M.B.Əliyeva
3. Qadınlarda genital infeksiyaların residivləşmə və digər somatik xəstəliklərin rastgəlmə xüsusiyyətləri.//Azərbaycan Təbabətinin Müasir Nailiyyətləri, № 4, 2018, səh.275-279 /M.B.Əliyeva
4. Qarışıq etiologiyalı bakterial urogenital infeksiyalı xəstələrdə laktobasil və bifidobakteriyaların kəmiyyət göstəriciləri.//Sağlamlıq.№5, 2018 səh.80-84/ M.B.Əliyeva
5. Mikroorqanizmlərlə assosasiyalar zamanı bakterial urogenital infeksiyalı xəstələrdə müxtəlif mikroorqanizmlərin kəmiyyət və keyfiyyət göstəriciləri.//Sağlamlıq, №5, 2018, səh.199-202.//M.N Əliyev, M.B.Əliyeva
6. Cinsiyyət yolları infeksiyalarının etiopatogenetik mexanizmləri.//Azərbaycan Xalq Cümhuriyyətinin 100 illik yubileyinə həsr olunmuş Təbabətin Aktual Problemləri, 2018, səh. 175 /M.B.Əliyeva
7. Частота рецидивов генитальных инфекций и других соматических заболеваний у женщин.The recurrence rate of genital infections and other somatic diseases in women.// bichик проблем биологii i медицини jurnali buraxiliş № 3 (145) 2018. səh.76-80/Алиева М.Б
8. Somatic disease and menstrual disorders in women with genital pathology. Соматические заболевания и нарушения менструального цикла у женщин с генитальными

патологиями.// L interna-tional correspondence scientific and practical conference « inter-national scientific review of the problems and prospects of modern science and education. (Boston.USA. October 22-23, 2018) səh 76-78 / Алиева М.Б.
9. Genital infeksiyalı xəstələrdə müxtəlif şərti – patogen mikroorqanizmlərin koloniyalaşma qabiliyyəti.// Azərbaycan Tibb Jurnalı, № 1, 2019. Səh. 120-126//M.N Əliyev, M.B.Əliyeva.
10. Qarışıq etiologiyalı bakterial urogenital infeksiyalı xəstələrdə laktobasil və bifidobakteriyaların keyfiyyət göstəriciləri .//Sağlamlıq, № 1, 2019, səh.76-81/M.B.Əliyeva.
11. Микробиологический мониторинг и сравнение частоты встречаемости различных микроорганизмов у больных с соматическими заболеваниями. // Биомедицина № 1, 2019, ст. 21-26/ Алиев М.Х., Алиева М.Б., Ибаева Ш.А.
12. Qadınlarda urogenital mikst-infeksiyaların gedişi və terapiyasının mikrobioloji xarakteristikası.//Azərbaycan Tibb Jurnalı, №2, 2020, səh.97-106 //M.N Əliyev, M.B.Əliyeva.
13. Genital infeksiyalı xəstələrdə probiotikoterapiyanın tətbiqi zamanı müxtəlif mikroorqanizmlərin kəmiyyət və keyfiyyət göstəriciləri.// Azərbaycan Tibb Universitetinin yaradılmasının 90, Azərbaycanda Ali Əczaçılıq Təhsilinin 80 illik yubileylərinə həsr edilmiş "Əczaçılığın müasir problemləri" mövzusunda V Beynəlxalq Elmi Konqres, 2021, səh. 379-380/M.B.Əliyeva.

The defense of the dissertation will be held at the FD 2.28 Dissertation Council meeting on "15" January 2025 at "14⁰⁰".

Address: 14 A. Gasimzade St., (conference hall), AZ 1022, Baku city.

The dissertation is accessible at the library of the Azerbaijan Medical University.

Electronic versions of the dissertation and abstract are available on the official website of the Azerbaijan Medical University (amu.edu.az).

The abstract was sent to the required addresses on 12 december 2024.

Signed for print: 06.12.2024
Paper format: 60 x 84 1/16
Volume: 37,987 characters
Order: 100
Number of hard copies: 30
"Tabib" Publishing House