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Analysis of Vagina Microecology and Investigation of Common Infection in Border Area of Chongzuo City

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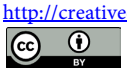

Abstract

Objective: To investigate the microecology and common infection of the female vagina in border area of Chongzuo City. **Methods:** Female patients in the outpatient department and inpatient department of gynecology of our hospital from April 1, 2018 to August 23, 2021 were selected as the research objects. The reproductive tract secretions of all patients were examined by microecological examination, morphological examination, chemical analysis and microscopic examination, and the results of various indicators were statistically analyzed to draw relevant conclusions. **Results:** Among the 1498 women with vaginal microecological imbalances, cleanliness was determined to be 534 cases of degree II, 868 cases of degree III, and 96 cases of degree IV; Nugent scores were 9 - 10 in 9 cases, 8 in 122 cases, 7 in 30 cases, 6 in 18 cases, 5 in 8 cases, 4 in 717 cases, 3 in 248 cases, 2 in 209 cases, 1 in 118 cases, and 0 in 19 cases. AV scores were 8 in 1 case, 7 in 1 case, 6 in 2 cases, 5 in 9 cases, 4 in 84 cases, 3 in 207 cases, 2 in 850 cases, 1 in 284 cases, and 0 in 60 cases. Among the pathogen infections, 99 cases were mycoplasma, 361 cases were candida, 199 cases were bacterial and 8 cases were trichomonas; In chemical analysis, there were 138 cases with pH 5.1, 326 cases with pH 4.8, 775 cases with pH 4.6, 217 cases with pH 4.4, and 42 cases with pH 4.1; meanwhile, 1390 cases were positive for hydrogen peroxide, 505 cases were positive for glucosidase, 487 cases were positive for acetylglucosaminidase, 184 cases were positive for sialidase, 290 cases were positive for proline aminopeptidase, 501 cases were positive for coagulase, and 1064 cases were positive for leukocyte esterase. **Conclusion:** Female reproductive tract microecology is more complex, and the pathogens of infection are also diverse. A systematic examination can clarify the occurrence and development of the disease, and then provide reliable laboratory data for clinical treatment and guide clinical medication.

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Keywords

Border Area, Chongzuo City, Female, Reproductive Tract, Microecology, Infection

1. Introduction

A variety of microorganisms are parasitized in the female vagina. Under normal circumstances, the micro-ecology maintains the stable balance; when the micro-ecological balance is disrupted, pathogenic microorganisms will continue to breed and cause vagina-related diseases [1] [2] [3]. In recent years, the examination of vaginal secretions is mainly based on microscopy which has high experience requirements and strong subjectivity; and the inspection process is affected by the quality of the microscope and the uniformity of the smear; moreover, diagnosis only by morphological identification has certain defects in sensitivity and specificity, which is difficult to meet the needs of clinical diagnosis [4] [5] [6]. This situation further leads to the low cure rate, high recurrence rate and low rational use rate of antibiotics in the clinical treatment of gynecological infection, which brings many unnecessary troubles to doctors and patients [7] [8]. Vaginal microecology analysis can assist in the diagnosis of women's vaginal health, including microecological environment, vaginitis, bacterial vaginosis (BV), trichomonas vaginitis, vulvovaginal candidiasis; vaginal microecology analysis can also be used for vaginal microecology assessment of women, auxiliary diagnosis of suspicious vaginal infections, finding asymptomatic infections, investigating cases of drug resistance caused by conventional treatment, helping clinical selection of treatment options, judging the recovery, and carrying out prevalence survey of vaginal infection, etc [9] [10] [11] [12]. In order to understand the microecology and common infection of the female reproductive tract in Chongzuo City, the enrolled patients were analyzed. The results are reported as follows.

2. Materials and Methods

2.1. Research Objects

A total of 1498 female patients were enrolled from April 1, 2018 to August 23, 2021 in the outpatient department and inpatient department of gynecology of our hospital. They were between 7 and 74 years old, with an average age of 33.45 ± 9.22 years. All cases felt vaginal discomfort and came to see a doctor, excluding other related diseases. The cases used in this study were signed by the patients themselves and approved by the medical ethics committee of our hospital.

2.2. Method

2.2.1. Specimen Collection

Collected by the clinician, a sterile swab or cotton swab is dipped in sterile normal saline and inserted into the patient's vagina, and the dome is rotated 3 - 5

times, and placed in a sterile plastic test tube for inspection.

2.2.2. Instruments and Reagents

The equipment uses the Shanghai Xingyao company ACT-2000 ultra-high magnification microscopy system, Antu AutowoMO automatic genital secretion workstation, Deere heater, Deere automatic staining machine DL-DYE-GR-8, sterile swab or cotton swabs and supporting reagents are provided by Antu Company.

2.2.3. Detection Method

1) Take the kit out of the refrigerated environment and place it at room temperature (18˚C - 25˚C) to equilibrate for at least 30 minutes; 2) Sample processing: Take a sample adding test tube, drop the joint test II diluent to between the two graduation lines at the bottom of the test tube, place the collected vaginal secretion swab in the test tube, wash it thoroughly and gently squeeze it with your fingers On the test tube wall, make the liquid absorbed by the swab flow back to the test tube, discard the swab, and get the sample liquid: A. Suck the sample solution through a straw, add 1 drop of sample solution (about 30 ul) to a clean slide, make thin slices, dry them in a Deere heater at 75˚C, and then use DL-Dye-GR-8 automatic Deere dyeing machine for Gram staining; After that, the microecological evaluation system indexes (epithelial cells, density of bacteria, diversity of bacteria, proportion of Gram-positive slender bacillus and proportion of other bacteria) were completed in ultrahigh magnification microscopic system; B. Use a straw to absorb the sample solution, add 1 drop of sample solution (about 30 ul) to the clean glass slide to make the wet slide, and complete the morphological examination of the wet slide (red blood cells, clue cells, cleanliness, white blood cells, pus cells, Trichomonas, mycoplasma, chlamydia, intracellular G-diplococcus and other indicators) in the ultrahigh power microscopic system. C. Take out the joint inspection card from the package, gently remove the plate stick, and use a straw to absorb the sample liquid. Add 1 drop of sample liquid to each of the first five holes of the joint inspection card, each drop is about 30 ul; 3) Put the five couplet detection card into Antu autowomo automatic reproductive tract secretion workstation for detection, judge with software, and score in strict accordance with Nugent and BV scoring standards. The results are transmitted to the information system and the inspection report is printed.

2.2.4. Result Judgment

1) The first hole (hydrogen peroxide): no color or light yellow (marked as "+") is positive, indicating imbalance of vaginal flora, pathological or sub-healthy vaginal environment; Light red (marked "±"), indicating the presence of moderate lactobacillus; 2) (Leukocyte esterase): no color or light color (marked with "−") indicates negative, indicating The second hole that the leukocytes are less than 5/HP; light blue (marked with "±") indicates negative, indicating that leukocytes 5 ~15/HP; blue (marked as "+", "++", "+++") indicates positive, indicating that the white blood cells are greater than 15/HP; 3) The third hole (neuraminidase):

no color or orange (marked with "−") indicates negative; light red (marked with "±") indicates weak positive; red, purple, blue, brown or black (marked with "+") indicates a positive hole that is positive for bacterial vaginosis; 4) The fourth hole (proline aminopeptidase): no color or light color (marked with "−") indicates negative; light yellow (marked with "±") indicates weak positive; yellow (marked with "+") means positive. If the hole is positive at the same time as the 5th hole is negative, it indicates BV infection. If the 5th hole is positive at the same time, the result will be judged according to the 5th hole; 5) The fifth hole (acetylglucosaminidase): no color or light color (marked with "−") indicates negative; light yellow (marked with "±") indicates weak positive; yellow (marked with "+") indicates positive. If the hole is positive at the same time Ph ≥ 4.8, it indicates trichomoniasis vaginitis, and if the hole is positive at the same time pH ≤ 4.6, it indicates vulvovaginal candida infection; 6) The sixth hole (PH): the color changes from yellow → cyan → green → blue, indicating that the pH changes from 3.8 to 5.4. Compared with the five joint test color card of vaginitis, the green pH is ≤4.6 and the blue pH is ≥4.8.

2.2.5. Interpretation of Inspection Results

1) For suspicious specimens with weakly positive results detected by this kit, re-determination and dynamic observation are recommended; 2) Due to methodological or specific reasons, different results may be obtained when the same sample is tested with reagents from different manufacturers. Therefore, the results obtained from different reagents should not be directly compared with each other to avoid false medical interpretation.

2.3. Explanation of Relevant Scores

Nugent score was an index to examine the microecology of vagina. Bacterial morphology was observed by gram staining and oil microscope, and lactobacillus, Gardnerella, Prevobacterium and Mobiluncus were scored by semi-quantitative evaluation method. The sum of the morphological values of the above 4 kinds of bacteria is the total value. If there are no miscellaneous bacteria, the score is zero, which means that there is no bacterial vaginosis. The more miscellaneous bacteria, the higher the score, and the greater the possibility of bacterial vaginosis. The normal range is 0 - 3 points, BV: Nugent score ≥ 7 points, BV intermediate: Nugent score 4 - 6 points, if the score is higher than 3 points, bacterial vaginosis is considered, and timely treatment is recommended. The AV score includes five detection indicators: LBG grade, white blood cell count, percentage of white blood cells containing toxic particles, background colonies, and percentage of parabasal epithelial cells. The score of each indicator can be divided into normal or mild (0 points), moderate (1 point) and severe (2 points), the scores of each indicator are summed up to obtain the total AV score. Diagnostic criteria: AV score < 3 points, no AV signs; AV score 3 - 4 points, mild AV. The routine inspection report of vaginal discharge generally includes the inspection of cleanliness, mold and trichomoniasis.

2.4. Statistical Analysis

Statistical analysis was performed using statistical software SSPS 24.0. Data statistics were expressed as ($x \pm s$), the comparison of the number of cases of vaginosis was performed by the chi-square test, and the comparison of the mean data between groups with normal distribution was performed by the t test. $P < 0.05$ was considered to be statistically significant.

3. Results

Among the 1498 women with vaginal microecological imbalances, 534 cases were classified as grade II, accounting for 35.65%; 868 cases were grade III, accounting for 57.94%, and 96 cases were grade IV, accounting for 6.41%; Nugent score of 9 - 10 points in 9 cases, 8 points in 122 cases, 7 points in 30 cases, 6 points in 18 cases, 5 points in 8 cases, 4 points in 717 cases, 3 points in 248 cases, 2 points in 209 cases, 1 point in 118 cases, 0 point in 19 cases; 8 points of AV score in 1 case, 7 points in 1 case, 6 points in 2 cases, 5 points in 9 cases, 4 points in 84 cases, 3 points in 207 cases, 2 points in 850 cases, 1 point in 284 cases, and 0 points in 60 cases; Among the pathogen infections, there were 99 cases of Mycoplasma infection, accounting for 6.61%; 361 cases of Candida infection, accounting for 24.10%; 199 cases of bacterial infection, accounting for 13.28%, and 8 cases of trichomonas infection, accounting for 0.53%; In chemical analysis, 138 cases with PH value of 5.1, accounting for 9.21%, 326 cases with PH value of 4.8, accounting for 21.76%, 775 cases with PH value of 4.6, accounting for 51.74%, 217 cases with PH value of 4.4, accounting for 14.49%, and 42 cases of PH4.1, accounting for 2.80%; Hydrogen peroxide was positive in 1390 cases, accounting for 92.79%, negative in 108 cases, accounting for 7.21%; glucosidase positive in 505 cases, accounting for 33.71%, negative in 993 cases, accounting for 66.29%; acetylglucosidase positive in 487 cases, accounting for 32.51%, 1011 cases were negative, accounting for 67.49%; 184 cases were sialidase positive, accounting for 12.28%, 1314 cases were negative, accounting for 87.72%; 290 cases were proline aminopeptidase positive, accounting for 19.36%, and 1208 cases were negative, accounting for 80.64%; Coagulase positive in 501 cases, accounting for 33.44%, negative in 997 cases, accounting for 66.56%; leukocyte esterase positive in 1064 cases, accounting for 71.03%, negative in 434 cases, accounting for 28.97%. The specific results are shown in **Tables 1-5**.

4. Discussion

Female Genital Tract (FGT) is an open cavity and is an important microecological settlement area of human body. Under normal circumstances, the vaginal microecological environment is composed of four parts of various vaginal flora, periodic endocrine changes, normal vaginal anatomy and vaginal local immune system [13]-[16]. When the vaginal flushing, menstruation, contraceptives, sexual intercourse frequency and long-term use of immunosuppressors, broad-spectrum antibiotics,

Table 1. Comparison of cleanliness in 1498 female patients with vaginal microecological imbalance.

Cleanliness	Number of cases	Percentage (%)
II degree	534	35.65
IIIdegree	868	57.94
IVdegree	96	6.41

Notes: Comparison of cleanliness class II and class III, χ^2 = 149.5541, P < 0.0000; the comparison between degree II and degree IV, χ^2 = 385.5980, P < 0.0000; the comparison between degree III and degree IV, χ^2 = 911.5399, P < 0.0000.

Table 2. Correlation score comparison of 1498 female patients with vaginal microecological imbalance.

Score	Nugent score (cases)	Percentage (%)	AV score (Cases)	Percentage (%)
9 - 10 Points	9	0.60	0	0.00
8 Points	122	8.14	1	0.07
7 Points	30	2.00	1	0.07
6 Points	18	1.20	2	0.13
5 Points	8	0.53	9	0.60
4 Points	717	47.86	84	5.61
3 Points	248	16.56	207	13.82
2 Points	209	13.95	850	56.74
1 Points	118	7.88	284	18.96
0 Points	19	1.27	60	4.01

Notes: Nugent A score of 0 - 3 is normal; If the score is greater than 3, bacterial vaginosis is considered; AV score < 3 points, no AV signs; AV score 3 - 4 points, mild AV.

Table 3. Comparison of pathogen infection in 1498 female patients with vaginal microecological imbalance.

Pathogen	Number of cases	Percentages (%)
Mycoplasma	99	6.61
Candida	361	24.10
Bacteria	199	13.28
Trichomonad	8	0.53

Note: comparison between mycoplasma infection and Candida infection, χ^2 = 176.2939, P < 0.0000; Compared with mycoplasma infection and bacterial infection, χ 2 = 37.2635, P < 0.0000; Comparison between mycoplasma infection and Trichomonas infection, χ^2 = 80.2589, P < 0.0000; Candida infection compared with bacterial infection, χ^2 = 57.6377, P < 0.0000; Comparison between Candida infection and Trichomonas infection, χ^2 = 385.1277, P < 0.0000; Comparison between Bacterial infection and Trichomonas infection, χ^2 = 189.3170, P < 0.0000.

Table 4. Comparison of pH in 1498 female patients with vaginal microecological imbalance.

PH value	Number of cases	Percentage (%)
5.1	138	9.21
4.8	326	21.76
4.6	775	51.74
4.4	217	14.49
4.1	42	2.80

Note: pH ≥ 4.8 indicates trichomonal vaginitis, and pH ≤ 4.6 indicates vulvovaginal Pseudofilament yeast infection.

Table 5. Comparison of chemical analysis results of 1498 female patients with vaginal microecological imbalance.

Project	Positive (cases)	Negative (cases)	χ^2 Value	P value
Hydrogen peroxide	1390	108	2194.2911	0.0000
Glucosidase	505	993	317.9493	0.0000
Acetyl glucosaminidase	487	1011	366.5901	0.0000
Neuraminidase	184	1314	1704.8064	0.0000
Proline aminopeptidase	290	1208	1125.1322	0.0000
Clotting enzyme	501	997	328.4593	0.0000
White cell esterase	1064	434	529.9065	0.0000

corticoids and other endogenous and exogenous factors, vaginal microecological environment will change which cause vaginal flora disorders and a large number of conditional pathogenic bacteria reproductive infection, leading to the occurrence of vaginitis.

Female genital tract infection is a common and frequently occurring disease in gynecological diseases [17] [18] [19] [20]. It is mainly manifested in peculiar smell, increase of vaginal secretions, pruritus of vulva, swelling of lower abdomen, burning sensation and other symptoms. It can be caused by bacteria, fungi, gonococcus, Trichomonas vaginalis, mycoplasma, chlamydia and other conditional pathogens. If it is not diagnosed and treated in time, it can cause cervicitis, cervical erosion Vaginitis, pelvic inflammatory disease, premature delivery, infertility and other complications; severe cases can also increase the risk of cervical cancer and AIDS infection. The physical and mental health of women of childbearing age has been seriously affected, which is a major global social problem and public health problem.

According to statistics, there are about 1 billion outpatient visits in obstetrics and gynecology in China every year, of which about 500 million have vaginal infections, and recurrent infections account for about 50%, resulting in infertility of about 30 million women of childbearing age every year, premature rupture of

membranes or even premature delivery of 3 million pregnant women, and the annual cost of medical treatment is as high as 40 billion Yuan. It is the number one killer of women's health in China [21] [22]. Female genital tract infection needs to be clear pathogen infection in order to symptomatic treatment and restore vaginal microecological balance.

With the continuous development and progress of inspection technology, the application of vaginal secretion zymography analysis technology is more and more widely, and its functional enzyme index combined with routine vaginal secretion microscopy can quickly and comprehensively reflect the existence of various inflammations and microecology in the vagina which can provide a basis for the diagnosis of vaginal inflammation types, so as to better guide clinical diagnosis and treatment, and improve the current status of diagnosis and treatment of female reproductive tract diseases [23] [24] [25].

The results of this study showed that in 1498 women with vaginal microecological imbalances, the cleanliness accounted for 64.35% between grades III and IV, and the differences were statistically significant in the pairwise comparison of cleanliness, $P < 0.001$. It suggests that the incidence of vaginitis is relatively high in women with vaginal microecological imbalance; There were 904 cases with Nugent score greater than 3, accounting for 60.34%, It was suggested that bacterial vaginitis occurred in female patients; in the AV score, 304 cases had more than mild AV signs, accounting for 20.29%, suggesting that about one fifth of the cases in this group had AV signs. Among the pathogen infections, 667 cases were found to have related pathogen infections, accounting for 44.52%, indicating that there was a clear pathogen infection in this group of patients; in terms of the four infectious pathogens, the pairwise comparisons showed that bacterial infections and fungal infections were more frequent; the differences were statistically significant, $P < 0.001$. In chemical analysis, glucosidase, Acetylglucosaminidase, sialidase, proline aminopeptidase, coagulase and leukocyte esterase were all positive in varying degrees, and the positive rates were 33.71%, 32.51%, 12.28%, 19.36%, 33.44% and 71.03% respectively; the difference between the negative and positive results of functional enzyme test was statistically significant, $P < 0.001$. All the positive results suggested that there was some vaginosis in the female patients with microecological imbalance.

5. Conclusion

Female reproductive tract microecology is more complex, and the infection pathogens are also diverse. The systematic examination can clarify the occurrence and development of diseases, thus providing reliable laboratory data for clinical treatment and guiding clinical medication. Vaginal microecological analysis can assist in the diagnosis of vaginal health conditions in women, including microecological environment, vaginitis, bacterial vaginosis (BV), trichomonas vaginitis, and vulvovaginal candidiasis; it can be used for the evaluation of vaginal microecology in women, auxiliary diagnosis of suspected vaginal infection, finding asymptomatic infection, investigating cases with drug resistance caused by conventional treat-

ment, helping clinical selection of treatment plan, judging recovery and investigating the prevalence of vaginal infection; it is worthy of popularization and application.

6. Limitations of the Study

Since the detection principle of the kit is based on biochemical reactions, if the sample is improperly collected, erroneous results can be produced. In addition, due to the complex changes in the vaginal flora of women and many interfering factors, the positive results should be diagnosed and treated in combination with clinical symptoms. In addition, the cases selected in this study are the population of Chongzuo City in the border area, and there may be differences in other areas.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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