



## Investigation of the Microbial Content and Antimicrobial Activity of Isolates from the Vagina of Non-Pregnant Women in AL-Najaf Governorate

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### ABSTRACT

**Background:** Vaginal discharge is a common problem among many women and causes unpleasant odor and discomfort. Therefore, the study was conducted on a group of these women to find out the microbial content causing this problem and to conduct a drug sensitivity test.

**Objectives:** The purpose of this study is to find out the microbial content causing this problem and to conduct a drug sensitivity test.

**Methodology:** Specimens were collected from 92 non-pregnant women, aged (20-60) years, from the city of AL-Najaf in Iraq. 78 (84.78%).

**Results:** Samples were positive, and the largest percentage of microbial content was for fungi represented by yeasts of *Candida spp.* in the rate of 54%. *C. albicans* was the most prevalent kind by 15%. The remainder is made up of non-*Candida albicans* kinds. Which is *C. glabrata*, *C. Ciferrii*, *C. Guilliermondii*, *C. Tropicals*, *C. krusi*. This was followed by gram-positive bacteria at a rate of 28%, This percentage was distributed between *Streptococcus pyogenes* by 15% and the rest for other types of bacteria *Staphylococcus spp.* While Gram-negative bacteria accounted for the least percentage of the total positive specimens, at 18%, distributed between *Klebsiella spp.* 13% and *E. coli*, 5%. As for the drug sensitivity test, *C. glabrata*, *C. ciferrii* and *C. Krusi* showed resistance to fluconazole (MIC= 8) and were sensitive to Voriconazole. Both Gram-positive and Gram-negative bacteria showed various drug sensitivity.

**Conclusion:** This study showed that there are two main causes of vaginal infections: first: an overgrowth of *Candida* yeasts, and second: some types of bacteria, which are similar to those that cause urinary tract infections, as these bacteria are transmitted between the vagina and the urinary canal due to the closeness of the two areas to each other in women.

**Keywords:** Investigation, Microbial, Drug, Non-Pregnant Women, the Vagina.

### INTRODUCTION

For women, vaginitis is a frequent condition that causes pain. Millions of clinical visits are attributed to symptomatic vaginitis each year. Patients who visit obstetrics and gynecology clinics frequently complain about abnormal vaginal discharge, itching, burning sensations, irritation, and

pain. Nonetheless, some vaginal infections are asymptomatic (Adeyba et al;2003).

Many lactobacilli also produce hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and small amounts of H<sub>2</sub>O<sub>2</sub>-producing lactobacilli in the vagina are thought to contribute to the development of BV (Vaginal Lactobacilli) with physiological functions can also

produce bacteriocins, which proceeds with lactic acid and (H<sub>2</sub>O<sub>2</sub>) to inhibit the growth of facultative anaerobic pathogens. Bacteriocins, bio surfactants, and congregating molecules inhibit the adhesion of such bacteria to the vaginal wall (Mendling et al;2016).

Vaginal infections are among the most frequent causes of consultations with gynecologists. The overabundance of residing microorganisms is the cause of many of infections. Due to the broad clinical signs of vulvovaginal candidiasis, it can be difficult to diagnose the condition correctly, which frequently results in ineffective or delayed therapy. Lack of a reliable and practical diagnostic method usually results in misdiagnosis ( Achkar and Fries:2010).

The three Infections are trichomoniasis, vulvovaginal candidiasis, and bacterial vaginosis. In a study of women who had symptoms, 22–50% had been diagnosed with bacterial vaginosis, 17–39% with vulvovaginal candidiasis, and 4–35% with trichomoniasis (Anderson et al ;2004 and Koumans et al;2007).

The most frequent cause of abnormal vaginal discharge in women who are of reproductive age is bacterial vaginosis (BV), about 30% of women who are of reproductive age have BV, The typical vaginal flora (Lactobacilli) is replaced by a mixed flora of aerobic, anaerobic, and microaerophilic species in BV, which is characterized by elevated vaginal pH and milky discharge. The vagina is mostly colonized by anaerobic organisms such as Gardnerella vaginalis, Prevotella spp., Mycoplasma hominis, and Mobiluncus spp. in BV (Lamichhane et al;2014).

A lower proportion of women have been shown to have aerobic vaginitis, with facultative anaerobic or aerobic bacteria predominating in their microbiota (lactobacilli), including S. aureus, group B streptococci, E. coli, and Klebsiella spp. (Prospero ;2014)

Lactobacilli produce lactic acid, which is why they play a major part in the commensal vaginal flora. Common lactobacilli found in the vaginas of healthy

women include Lactobacillus crispatus and Lactobacillus jensenii (Taylor et al;2013).

If there is no immunopathology, immunosuppression, or mucosal injury, the presence of Candida spp. in the vagina is typically not clinically noticeable. It is appropriate to classify these cases as vaginal fungal colonization. Objective clinical symptoms with evidence of inflammation, and the presence of Candida spp. in the vaginal discharge in the absence of another infection cause are all combined to form vulvovaginal candidiasis. (Pappas et al;2016).

### AIMS OF THE STUDY

The purpose of this study is to find out the microbial content causing this problem and to conduct a drug sensitivity test.

### METHODOLOGY

#### Specimens collection:

92 Swabs were collected from the vaginal area of non-pregnant women aged between 23 and 60 years, for the period from May 2023 to February 2024 from AL-Najaf hospitals. The total number of positive specimens was 78, a rate of 84.78%. Immediately after collection, the Specimens were sent to the laboratory for the purpose of diagnosis and drug sensitivity testing.

#### Identification of Gram-positive bacteria

Using culture medium and biochemical tests, staphylococci were identified based on their morphological characteristics (colony size, shape, color, hemolysis, translucency, edge, elevation, and texture). Every sample was investigated under a microscope to ascertain the existence of bacteria-causing staphylococci (MacFaddin, 2000 ; Mohsin et al;2024).

The laboratory will inoculate on blood agar in one hour, MacConkey agar, Mannitol salt agar or MSA agar which will then be incubated at 37 °C for a duration of 24 to 48 hours (Forbes et al;2007).

### Identification of Gram -negative bacteria

The principal isolation of bacterial pathogens in the vagina, lactose fermentation based on colonial morphology on MacConkey agar, oxidase and indole assays, and a small number of standard biochemical tests were used to make the identification. The characteristics of Enterobacteriaceae, which are bacilli, non-sporulated, have variable motility, grow in the presence and absence of oxygen, ferment organisms of glucose, are cytochrome oxidase negative, and can reduce nitrate to nitrite, must be exposed because laboratory characterization is a crucial component when it comes to microorganisms (Ng et al;2010).

### Identification of *Candida spp*

The diagnosis of isolation is based on microscopic, molecular, biochemical, and cultural traits. In order to study the form, size, and budding of the cells, a glass slide containing the colonies was produced with a drop of normal saline, covered with the cover slide, and subjected to light microscopy staining (De Hoog et al., 2000).

Following a 24- to 48-hour incubation period, the yeast colonies' form, size, edge, color, and appearance were recorded on Sabouraud dextrose agar (SDA) medium. One cell was taken from the yeast colony on SDA, and the isolation was recorded

and accepted for 24-48 hours at 37 ° C. The CHROM agar *Candida* assay was utilized to help identify the species of *Candida* based on the color (Horvath et al;2003; Alyassree and Alrufae;2021).

### Antimicrobial Resistance test

Tests for broth dilution. The macrobroth, also known as the tube-dilution method, was one of the first techniques for testing for antibiotic resistance. In this process, liquid growth media was distributed in test tubes containing two-fold dilutions of antibiotics (e.g., 1, 2, 4, 8, and 16 µg/mL) (Jorgensen et al; 2007).

The disk diffusion susceptibility method is a straightforward, useful, and well-standardized technique [2, 11, 12]. A bacterial inoculum of around 1-2×10<sup>8</sup>CFU/mL is applied to the surface of a large Mueller-Hinton agar plate (diameter of 150 mm) to conduct the test. On the infected agar surface, up to 12 commercially manufactured paper antibiotic disks with a predetermined concentration are inserted (Wayne;2009).

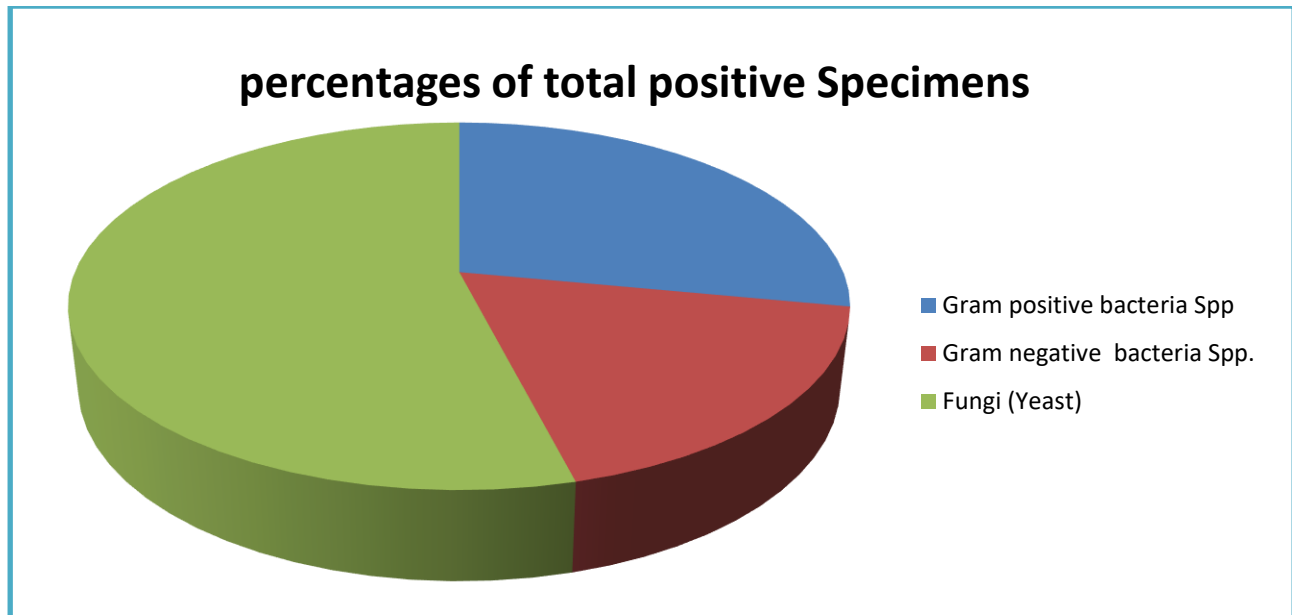
A standard medium, two-fold serial dilutions of an antifungal medication, and a predefined quantity of test organism inoculum are utilized in a test tube or microtiter plate. The medication concentration that stops the test organism's growth to a certain extent is the assay's endpoint (Wayne;2017).

## RESULTS

**Table (1):** Numbers and percentages of total positive Specimens divided by Type of microbe.

| Type of microbe.                   | No.       | percentage  |
|------------------------------------|-----------|-------------|
| Fungi (Yeast)                      | 42        | 54%         |
| Gram positive bacteria <i>spp.</i> | 22        | 28%         |
| Gram negative bacteria <i>spp.</i> | 14        | 18%         |
| <b>Total</b>                       | <b>78</b> | <b>100%</b> |

Table No. (1) shows the numbers and percentage of bacteria and fungi represented by yeasts in a total of 78 positive Specimens out of 92 Specimens collected by taking swabs from the vaginal area.



**Figure (1):** percentages of total positive Specimens divided by Type of microbe.

Figure No. (1) represents the percentage of microbial content after identifying isolates taken from the vagina in the laboratory, where the largest percentage was 54% for fungi (yeasts), followed by gram-positive bacteria at 28% and gram-negative bacteria at 18%. This result was consistent with (Mulu et al;2015).

**Table (2):** Numbers and percentage of *Candida* spp. isolated from vagina.

| <i>Candida</i> spp.      | No.       | percentage  |
|--------------------------|-----------|-------------|
| <i>C. albicans</i>       | 12        | 15 %        |
| <i>C.glabrata</i>        | 10        | 13 %        |
| <i>C. ciferrii</i>       | 10        | 13 %        |
| <i>C. guilliermondii</i> | 4         | 5 %         |
| <i>C. tropicalis</i>     | 2         | 3 %         |
| <i>C. krusi</i>          | 2         | 3 %         |
| <i>C. parapsilosis</i>   | 2         | 3 %         |
| <b>Total</b>             | <b>42</b> | <b>54 %</b> |

Table No. (2) shows that *C. albicans* was present at a greater rate of 15%, followed by *C. glabrata* and *C. ciferrii* by 13%, followed by *C. guilliermondii* by 4%, and the lowest percentage was 3% for *C. tropicalis*, *C. parapsilosis* and *C. Krusi*.



**Figure (2): A:** Micrography reveals colonies of *Candida* spp. developing on SDA at 37C for two days. **B:**

Micrographs showing *C.tropicalis* (blue). **C:** *C.krusei* (pink), *C.parapsilosis* (whiet), *C.albicans* (green), *C.ciferrii*(mauve) and *C.glabrata* (mauve) colonies growing on CHROMagar 2 days 37 C°.

**Table (3):** Antifungal Susceptibility of a Few Yeast Isolated in This Investigation

| Yeast                    | Antifungal   | MIC    | Interpretation |
|--------------------------|--------------|--------|----------------|
| <b>C. albicans</b>       | Fluconazole  | ≤ 1    | S              |
|                          | Voriconazole | ≤ 1    | S              |
| <b>C. glabrata</b>       | Fluconazole  | 8      | R              |
|                          | Voriconazole | ≤ 0.12 | S              |
| <b>C. ciferrii</b>       | Fluconazole  | 8      | R              |
|                          | Voriconazole | ≤ 0.12 | S              |
| <b>C. guilliermondii</b> | Fluconazole  | ≤ 1    | S              |
|                          | Voriconazole | ≤ 0.12 | S              |
| <b>C. tropicals</b>      | Fluconazole  | <8     | S              |
|                          | Voriconazole | ≤ 1    | S              |
| <b>C. krusi</b>          | Fluconazole  | ≥64    | R              |
|                          | Voriconazole | ≥4     | S              |
| <b>C. parapsilosis</b>   | Fluconazole  | <8     | S              |
|                          | Voriconazole | ≤ 1    | S              |

\*\*\*R: Resistance, \*S: Sensitive, \*\*I: Intermediate.

Table No. (3) displays that *C. albicans* , *C. guilliermondii* , *C. Tropicals* and *C. Parapsilosis* susceptibility to Fluconazole and Voriconazole. While *C.glabrate* , *C.ciferrii* and *C. Krusi* shown resistance to fluconazole (MIC 8), *C.ciferrii* demonstrated susceptibility to voriconazole (MIC < 0.12). The outcomes aligned with the findings of (Gunsilius et al;2001).

**Table (4):** Numbers and percentage of gram positive bacteria *spp.* isolated from vagina.

| Gram positive bacteria <i>spp.</i> | No.       | percentage |
|------------------------------------|-----------|------------|
| <b>Strept. pyogenes.</b>           | 12        | 15%        |
| <b>Staph. saprophytic</b>          | 6         | 8%         |
| <b>Staph. arues</b>                | 2         | 3%         |
| <b>Staph. haemolytics</b>          | 2         | 3%         |
| <b>Total</b>                       | <b>22</b> | <b>28%</b> |

Table No. (4) shows the positive bacterial content of Gram stain, where Streptococcus pyogenes bacteria had the highest percentage, at 15%, followed by Staphylococcus *spp.* By 13%, distributed between Staph. saprophytic by 8% and 3% for each. The outcome was the same (Li et al., 2012).

**Table (5):** Antibiotic Susceptibility of gram positive bacteria *spp* Isolated

| Gram positive bacteria <i>Spp.</i> | Antibiotic Susceptibility |                      |
|------------------------------------|---------------------------|----------------------|
|                                    | S                         | R                    |
| <b>Streptococcus pyogenes.</b>     | TOB/ Lev/ Mem/Cip         | AMC / CTX /CTR /IMI. |
| <b>Staph. saprophytic</b>          | Lev/CN/AK/AMC/Cip/ IMI    | E/CTX/AZM/MeM        |
| <b>Staph. arues</b>                | IMI /AK                   | AMC/Lev/ Cip / CTX   |
| <b>Staph. haemolytics</b>          | C/Lev/CN/ OXA             | pi                   |

\* Ciprofloxacin (Cip), Meropenem (Mem), Levofloxacin (Lev), Tobramycin (TOB), Cefotaxime (CTX), Ceftriaxone (CTR), Imipenem (IMI), Amoxicillin /clavulanic acid (Amc), Azithromycin (AZM) ,cefotetan (CN), Oxacillin (OXA), Chloramphenicol (C), pi-pipemidic acid (pi).

Table No. (5), displays pharmacological sensitivity and resistance. Ciprofloxacin (Cip), Meropenem (Mem), Levofloxacin (Lev), and Tobramycin (TOB) all proved responsive to Streptococcus pyogenes. Despite being resistant to Imipenem (IMI), Cefotaxime (CTX), Ceftriaxone (CTR), and Amoxicillin /clavulanic acid (Amc). Staph. saprophytic was resistant to Azithromycin (AZM) and erythromycin( E), however it was susceptible to cefotetan (CN) and Amikacin (AK) and imipenem (IMI) were both effective against Staph. aureus ,as shown in Figure No. (3).

**Figure (3):** A: Staph. arues on blood agar. B: displays pharmacological sensitivity and resistance to Staph. arues on Mueller Hinton agar.

**Table (6):** Numbers and percentage of gram negative bacteria *spp.* isolated from vagina.

| Gram negative bacteria <i>spp.</i> | No.       | percentage |
|------------------------------------|-----------|------------|
| <i>Klebsiella spp.</i>             | 10        | 13%        |
| <i>E.coli</i>                      | 4         | 5%         |
| <b>Total</b>                       | <b>14</b> | <b>18%</b> |

**Table (7):** Antibiotic Susceptibility of gram negative bacteria *spp* Isolated

| Gram negative bacteria <i>spp.</i> | Antibiotic Susceptibility  |                 |
|------------------------------------|--|-----------------|
|                                    | S  | R               |
| <i>Klebsiella spp</i>              | AMK / IMI / NA/ Lev / MeM / Cip / CN                                 | ATM / CAZ / CTX |
| <i>E. coli</i>                     | CN / Cip / NOR / IPM / OF / ATM / CRO / Lev / CTX / CPM / MeM / TOB/ | NA / TMP        |

Norfloxacin (NOR), Ceftazidime (CAZ), Amikacin (AMK), Ofloxacin (OF), Ceftriaxone (CRO), Cefepime (CBM), Imipenem (IPM).

Table No. (7) shows the sensitivity of negative bacteria to Gram stain that were isolated from the vagina after they were diagnosed and a drug sensitivity test was performed. It shows that both *Klebsella spp.* and *E.coli* Sensitive to Ciprofloxacin (Cip), Meropenem (Mem), Levofloxacin (Lev) and cefotetan (CN). *Klebsella spp* Sensitive to Nalidixic acid (NA) but *E.coli* was resistant to it. *E.coli* Sensitive to Aztreonam (ATM) While *Klebsiella spp.* was resistant to it, as shown in Figure No. (4). This result was consistent with (Najjuka et al;2016).



**Figure (4):** A: *E.coli* on MacConkey agar (MAC). B: displays pharmacological sensitivity and resistance to *E.coli* on Mueller Hinton agar.

#### DISCUSSION:

Every balance between good and bad bacteria in the vagina is very fragile, and an imbalance can occur when the vaginal pH is not acidic enough. To maintain healthy vaginal acidity, the vaginal pH should be between 3.8 and 4.5. If the vagina is not acidic enough due to a lack of lactic acid bacteria, fungus and "bad" bacteria can multiply more than

normal. Two examples of infections caused in this way are thrush (caused by an overgrowth of the fungus *Candida*) and bacterial vaginosis (caused by an overgrowth of "bad" bacteria). (Sally Robertson; 2024).

Makes it abundantly evident that the microbiological composition of vaginitis is identical to that of urinary tract infections caused by both Gram-positive and Gram-negative bacteria as well as some

fungi. Uropathogenic *Escherichia coli* is the most prevalent causal agent of both simple and complex UTIs (UPEC). *Klebsiella pneumoniae*, *Staphylococcus saprophyticus*, *Enterococcus faecalis*, Group B *Streptococcus* (GBS), *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Candida spp.* are the agents that transmit UPEC in simple UTIs. (Kline et al;2011 and Foxman;2014).

Table No. (2) shows the results mentioned above were similar, showing that around 75% of women had experienced at least one Vulvovaginal Candidiasis (VVC) episode in their lifetimes, in addition to changes in hormones, pregnancy, immunity, hygiene practices, and other risk factors. The risk of VVC is increased by antibiotic medication. (Ferreira et al., 2016).

Non-albicans species have become more prevalent in recent decades. The investigation revealed that *C. glabrata*, *C. tropicalis*, *C. famata*, *C. kefyr*, and *C. krusei* were the most often isolated non-albicans *Candida* isolates (Haddadi et al;2014).

Non- *Candida albicans* ((NAC) species cause a rise in the incidence of Vulvovaginal Candidiasis (VVC), but they also tend to be more resistant to antifungal drugs. *Candida albicans* is often responsible for 70–90% of VVC cases; (NAC) species, such as *Candida glabrata*, *Candida parapsilosis*, *Candida tropicalis*, and *Candida krusei*, are also responsible for certain infections, as in Figure No. (2) (Ilkit and Guzel, 2011).

*Candida* species vary in their susceptibility, depending on the specific species that cause the illness, the geographic location, the patient population, and the way health care is managed in each area. While *Candida albicans* is sensitive to most antifungal medications and is eliminated from the patient's body during prophylaxis, non-albicans *Candida* species, such as *C. glabrata* and *C. krusei*, are resistant and progressively appearing in the infected individuals (Badiie et al;2017).

Group B streptococci (GBS) and Enterobacterales colonization in mothers is thought to be a major risk factor for the development of newborn sepsis (Frank et al;2021).

When lactobacilli are destroyed, these bacteria take control. The bacterial vaginosis BV-associated bacteria produce a variety of volatile amines. It is these compounds that give BV its characteristic smell. Because the amines smell stronger at the higher pH linked to semen, this stench is usually more noticeable after intercourse, especially unprotected sex (Lobo et al;2016).

Table No. (5) these result was consistent with the emergence of bacterial resistance has posed a threat to the effectiveness of antibiotics in recent times. As we've previously shown, baicalin (Bac) and azithromycin (AZM) worked together to combat *Staphylococcus saprophyticus* that was resistant to Azm (ARSS) (Wang et al 2022).

*Staph. hemolytics* was susceptible to Levofloxacin (Lev), cefotetan (CN), Oxacillin (OXA) and Chloramphenicol (C). However, it exhibited resistance to pi-pipemidic acid (pi). This result was not consistent with (Lee ;2003).

Antimicrobial resistance (AMR) is a global issue that arises from resistant diseases, the number of which is predicted to surpass 10 million annually by 2050 (Klinker et al., 2021).

Table No. (6) shows the percentage of Gram-negative bacteria isolated from vaginal samples after their diagnosis in the laboratory. *Klebsiella spp.* was 13% and *E. coli* was 5%. It is noted that in most previous studies, these bacteria were diagnosed as one of the pathogens of urinary tract infection. Which confirms the transfer of this bacteria from the urethra to the vagina, causing inflammation. This result was consistent with (Golsha et al;2021).

The Enterobacteriaceae family of bacteria is one of the key groups that cause these diseases. *Escherichia coli* (*E. coli*) and *Klebsiella* are two important genera in this family; they are both

nosocomial opportunistic pathogens (Eskandari-Nasab et al;2018).

These two genera *E. coli* and *Klebsiella pneumoniae* in particular—are opportunistic pathogens that can cause urinary tract infections of soft tissues, meningitis, septicemia, bacteremia, and infantile enteritis. Hospitalized individuals with immunodeficiency and underlying diseases like diabetes mellitus and chronic lung ailments are susceptible to infections and illnesses caused by these human pathogens (Kaye and Pogue;2015).

Physicians and legislators can better address the issue of antibiotic resistance by making informed judgments based on changes in sensitivity and resistance patterns (Gopalakrishnan and Sureshkumar, 2010).

Enterobacterales are becoming resistant to beta-lactam antibiotics, which presents a clinical problem. With high-quality data supporting their safety, beta-lactam antibiotics are the most widely used class of antibiotics for treating infections during pregnancy (Bookstaver et al;2015).

### CONCLUSIONS:

Bacterial vaginosis (BV) is an infection of the vaginal tract. It results from a change in the normal bacterial balance of the vagina.

A yeast infection and BV are not identically. Bacteria cause bacterial vaginosis (BV), whereas yeast (commonly known as *Candida*) fungus causes yeast infections. These infections result from an imbalance in the normal amounts of yeast or bacteria in the vagina. Treatment is necessary for both yeast infections and bacterial infections that increase vaginal discharge.

For women who are of reproductive age, bacterial vaginosis, candidiasis are prevalent conditions. Consequently, it is important to start screening women who are of reproductive age for vaginal infections. Additionally, ciprofloxacin, norfloxacin, and gentamicin are the prescribed

medications for prophylactic use and empirical therapy, respectively.

### REFERENCES:

- Anderson M.R., Klink ,K.,Cohrssen, A.(2004). Evaluation of vaginal complaints. *JAMA*. 2004;291:1368–79.
- Adeyba, O.A., Adeoye M.O., Adesiji ,Y.O.(2003). Bacteriological and parasitological vaginitis in pregnant women in iseyin, oyo state, Nigeria. *Clin Exp Microbiol*. 2003;4:11–6.
- Achkar, J,M., Fries, B,C. (2010). *Candida* infections of the genitourinary tract. *Clinical microbiology reviews*, 23(2), 253-273.
- ALyassree, H., AlrufaeMohamed, M. (2021). Increased Ear Infections of *Candida Ciferrii* and *Candida Guilliermondii* compared to *C. Albicans* and other Non-Albicans *Candida* Species in AL-Najaf Governorate. *Annals of the Romanian Society for Cell Biology*, 25(6), 8350-8361.
- Badiee, P., Choopanizadeh, M., Moghadam, A.G., Nasab, A.H., Jafarian, H., Shamsizadeh, A., & Soltani, J. (2017). Antifungal susceptibility patterns of colonized *Candida* species isolates from immunocompromised pediatric patients in five university hospitals. *Iranian journal of microbiology*, 9(6), 363–371.
- Bookstaver, P.B., Bland, C.M., Griffin, B., Stover, K.R., Eiland, L.S., McLaughlin, M.(2015). A Review of Antibiotic Use in Pregnancy. *Pharmacotherapy* 2015, 35, 1052–1062.
- Clinical and Laboratory Standards Institute.(2009). Performance standards for antimicrobial susceptibility testing. Nineteenth informational supplement M100-S19, 2009Wayne, *PA Clinical and Laboratory Standards Institute*.
- Clinical and Laboratory Standards Institute. (2017). Reference method for broth dilution antifungal susceptibility testing of yeasts, 4th ed Approved standard M27 Clinical and Laboratory Standards Institute, *Wayne, PA*.
- De Hoog ,G.S.J.,Guarro, j.,Gene,M.J. and Figueras, F. (2000) .Atlas of clinical fungi ,2nd.,centraabureeau

- voorschimmelculturees Utrecht, the Netherlands. Vol.53-No.1,P: 45-49.
- Eskandari-Nasab ,E., Moghadampour, M., Tahmasebi ,A.(2018). Prevalence of blaCTX-M gene among extended-spectrum  $\beta$ -lactamases producing *Klebsiella pneumoniae* clinical isolates in Iran: A meta-analysis. *Iranian Journal of Medical Sciences*. 2018;43(4):347.
- Ferreira, C.,Alves, C.T., Henriques, M.,Azeredo, J and Silva, S. (2016). Vulvovaginal candidiasis: Epidemiology, microbiology and risk factors. *Crit. Rev. Microbiol.*, 42, 905–927.
- Forbes, B.A., Sahm, D.F., Weissfeld, A.S., et al. (2007) *Baily and Scott Diagnostic Microbiology*. 12th Edition, Mosby Elsevier, Philadelphia, 93-107, 187-197, 842-854.
- Foxman, B. (2014) Urinary tract infection syndromes: occurrence, recurrence, bacteriology, risk factors, and disease burden. *Infect. Dis. Clin. North Am.* 28, 1–13.
- Frank Wolf, M., Abu Shqara, R.,Naskovica, K.,Zilberfarb, I.A.,Sgayer, I.,Glikman, D.,Rechnitzer, H., Fleisher Sheffer, V., Bornstein, J.(2021). Vertical Transmission of Extended-Spectrum, Beta-Lactamase-Producing Enterobacteriaceae during Preterm Delivery: A Prospective Study. *Microorganisms* 2021, 9, 506.
- Haddadi, P., Zareifar, S.,Badiie ,P.,Alborzi ,A.,Mokhtari, M.,Zomorodian, K, et al.(2014). Yeast colonization and drug susceptibility pattern in the pediatric patients with neutropenia. *Jundishapur J Microbiol* 2014;7(9): e11858.
- Horvath,L.L.,Murray, D.R.,C.K. and Dooley, D.P.(2003).Direct isolation of *Candida* spp. from blood cultures on the chromogenic medium chromagarCandida. *J.ClinMicrobiol* .41:2629-32.
- Golsha, R., Montazeri, M., Razaghi, N., Zade, M. E. (2021). Frequency of Beta-Lactamase Antibiotic Resistance Genes in *Escherichia Coli* and *Klebsiella pneumoniae*. *Ethiopian journal of health sciences*, 31(3), 663–672
- Gopalakrishnan ,R., Sureshkumar ,D. (2010). Changing trends in antimicrobial susceptibility and hospital acquired infections over an 8 year period in a tertiary care hospital in relation to introduction of an infection control programme. *J. Assoc. Physicians India* 58 Suppl, 25–31.
- Gunsilius, E.,Lass-Flörl , C.,Kähler, C.M.,Gastl, G and Petzer, A.L.( 2001). *Candida ciferrii*, a new fluconazole-resistant yeast causing systemic mycosis in immunocompromised patients. *Ann Hematol*. Mar; 80 (3):178-9.
- Ilkit, M and Guzel, A. B. (2011). The epidemiology, pathogenesis, and diagnosis of vulvovaginal candidosis: a mycological perspective. *Crit Rev Microbiol* 37, 250–261.
- Jorgensen ,J.H.,Turnidge, J.D.,Murray, P.R.,Baron, E.J.,Jorgensen, J.H.,Landry, M.L.,Pfaller, M.A.(2007). Antibacterial susceptibility tests: dilution and disk diffusion methods, Manual of clinical microbiology, 20079th ed. Washington, DC American Society for Microbiology pg. 1152-72.
- Kaye, K.S.,Pogue, J.M.(2015). Infections caused by resistant gram-negative bacteria: epidemiology and management. *Pharmacotherapy: The Journal of Human Pharmacology and Drug Therapy*. 2015;35 (10):949–962.
- Kline, K. A., Schwartz, D. J., Lewis, W. G., Hultgren, S. J., Lewis, A. L. (2011) .Immune activation and suppression by group B *Streptococcus* in a murine model of urinary tract infection. *Infect. Immun.* 79, 3588–3595.
- Klinker K. P., Hidayat L. K., DeRyke C. A., DePestel D. D., Motyl M., Bauer K. A. (2021). Antimicrobial stewardship and antibiograms: importance of moving beyond traditional antibiograms. *Ther. Adv. Infect. Dis.* 8, 20499361211011373. doi: 10.1177/20499361211011373.
- Koumans, E.H.,Sternberg, M.,Bruce, C.,McQuillan, G., Kendrick ,J.,Sutton, M, et al.(2007). The prevalence of bacterial vaginosis in the United States, 2001–2004; associations with symptoms, sexual behaviors, and reproductive health. *Sex Transm Dis.* 2007; 34:864–9.
- Lamichhane, P.,Joshi, D.R.,Subedi, Y.P.,Thapa ,R.,Acharya, G.P.,Lamsal, A, et al.( 2014). Study on

- types of vaginitis and association between bacterial vaginosis and urinary tract infection in pregnant women. *IJBAR*. 2014;05(06):305–7.
- Lee, J. H. (2003). Methicillin (Oxacillin)-resistant *Staphylococcus aureus* strains isolated from major food animals and their potential transmission to humans. *Applied and environmental microbiology*, 69(11), 6489–6494.
- Li, J., McCormick, J., Bocking, A., Reid, G. (2012). Importance of vaginal microbes in reproductive health. *Reprod Sci* 2012;19:235–42.
- Lobo, R.A., Gershenson, D.M., Lentz, G.M., Valea, F.A. (2016). *Comprehensive Gynecology*. New York, NY: Elsevier Health Sciences; 2016.
- MacFaddin, J. F. (2000). Biochemical tests for identification of medical bacteria - Ghent University Library. 1–928.
- Mending, W. (2016). Normal and abnormal vaginal microbiota. *Laboratoriums Medizin*, 40(4), 239-246.
- Mohsin, A. J., Sarhan, N. H., Idreess, H. G. (2024). Isolation and Identification of *Staphylococcus Aureus* and *Candida spp.* from Throat Infections.
- Mulu, W., Yimer, M., Zenebe, Y., Abera, B. (2015). Common causes of vaginal infections and antibiotic susceptibility of aerobic bacterial isolates in women of reproductive age attending at Felegehiwot Referral Hospital, Ethiopia: a cross sectional study. *BMC women's health*, 15, 42.
- Ng, L.S., Tan, T.Y., Yeow, S.C. (2010). A cost-effective method for the presumptive identification of Enterobacteriaceae for diagnostic microbiology laboratories. *Pathology*. 2010 Apr;42(3):280-3.
- Najjuka, C. F., Kateete, D. P., Kajumbula, H. M., Joloba, M. L., Essack, S. Y. (2016). Antimicrobial susceptibility profiles of *Escherichia coli* and *Klebsiella pneumoniae* isolated from outpatients in urban and rural districts of Uganda. *BMC research notes*, 9, 235.
- Pappas, P. G., Kauffman, C. A., Andes, D. R., Clancy, C. J., Marr, K. A., Ostrosky-Zeichner, L., ... & Sobel, J. D. (2016). Clinical practice guideline for the management of candidiasis: 2016 update by the Infectious Diseases Society of America. *Clinical Infectious Diseases*, 62(4), e1-e50.
- Prospero, F.D. (2014). Focus on candida, trichomonas, bacteria and atrophic vaginitis. Available at <http://womanhealthgate.com/focus-candida-trichomonasbacteria-atrophic-vaginitis/> on July 10, 2014.
- Robertson, Sally. (2019, February 27). Vaginal Flora. News-Medical. Retrieved on March 13, 2024 from <https://www.news-medical.net/health/Vaginal-Flora.aspx>.
- Taylor, B.D., Darville, T., Haggerty, C.L. (2013). Does bacterial vaginosis cause pelvic inflammatory disease? *Sex Transm Dis*. 2013;40:117–22.
- Wang, J., Meng, J., Zhu, J., Qiu, T., Wang, W., Ding, J., Liu, Z., Li, K., Wang, D., Liu, J., & Wu, Y. (2022). Reversal of Azithromycin Resistance in *Staphylococcus saprophyticus* by Baicalin. *Frontiers in veterinary science*, 9, 827674.