

VAGINAL MYCOTIC INFECTIONS AMONG STUDENTS AT OBAFEMI AWOLOWO UNIVERSITY: PREVALENCE AND ETIOLOGICAL AGENTS



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| Abstract: | Mycotic infections of the vagina are a vastly common type of infection. <i>Candida, Monilia</i> and <i>Cryptococcus</i> are the commonly associated pathogens with mycotic infections of the vagina. This study examined the fungal pathogens associated with vagina candidiasis in females who were of the ages 18-25 years. Vaginal swab samples of 116 students of the Obafemi Awolowo University, Ile Ife, Osun State were screened of which 20 of these individuals had frequent visits to the Obafemi Awolowo University Health center for examinations of vaginal secretion, independent of the presence or absence of symptoms of vulvovaginal candidiasis (VVC) between the months of July and September, 2023. The swab samples were transferred to Nutrient broth, and streaked on Potato Dextrose Agar (PDA) and incubated for five to seven days. Microscopy was then carried out to identify and distinguish the fungal hyphae. Results showed that 11.2% of the participants were infected with vagina candidiasis but were asymptomatic, while 9.6% were symptomatic and have the vagina infection. 31% of the sampled individuals showed symptoms without infection. Likely due to dietary habits and lifestyle, prolonged use of antibiotics or oral contraceptives, use of topical agent. Moreover, 35.3% had normal positive growth indicating yeast growth as part of the normal flora of the vagina, while 12.9% showed no growth, possibly due to sample collection errors. The study highlights fluconazole resistance as a limiting factor in vulvovaginal candidiasis management and emphasizing the need for proper sex education, healthy lifestyle practices, and vaccine development to control vaginal mycotic infections. |
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| Keywords: | Antibiotic use, Candida, Fungal pathogens, Prevalence study, vaginal candidiasis, Yeast flora |

Introduction

Fungi constitute a significant component of Earth's eukaryotic biodiversity, with an estimated 611,000 species, representing approximately 7% of all eukaryotic organisms (Mora *et al.*, 2011). Among these, pathogenic fungi are a notable concern, with around 600 species implicated in various infections (Brown *et al.*, 2012). *Candida* species, in particular, are common causes of nosocomial infections, with Candida albicans being a frequent pathogen associated with both superficial infections, such as oral and vaginal candidiasis, and severe systemic infections (Pfaller and Diekema, 2010; Calderone and Clancy, 2012).

Candida albicans was first described in 1839 by Langenbeck, it was named for its characteristic white exudate on infected mucosal surfaces (Dardar *et al.*, 2018). This yeast's ability to switch between different growth forms—such as yeast, hyphal, and pseudohyphal forms—in response to environmental conditions like nutrient availability and pH is a critical virulence factor (Noble *et al.*, 2017). This phenotypic plasticity facilitates its survival in diverse host niches and its ability to evade immune responses (Brown *et al.*, 2014).

The vaginal mycobiota encompasses the fungal community within the female reproductive tract, with *Candida albicans* being a prominent member. Vulvovaginal candidiasis (VVC), the second most common form of infectious vaginitis, highlights the importance of understanding fungal communities in the vagina (Carter *et al.*, 1959). Although *Candida albicans* is frequently isolated, with prevalence rates ranging from 70% to 95% in various studies, nonalbicans species like *C. glabrata, C. tropicalis,* and *C. parapsilosis* also contribute to the vaginal fungal community (Sobel, 1985). The mycobiome's complexity and variability across different populations and conditions underscore the need for more comprehensive studies.

Despite advancements in understanding the vaginal microbiota, there is a notable gap in research focusing specifically on vaginal mycotic infections. Most existing studies have predominantly addressed bacterial communities, leaving a critical need to explore the fungal components in greater detail (Klis *et al.*, 2009). The rise in mycotic infections has been linked to various factors, including antibiotic use, hormonal changes, and immune status, which can disrupt the vaginal ecosystem and lead to opportunistic infections (Nicholls et al., 2011).

The epidemiology of vaginal candidiasis reveals a significant prevalence in different populations. For instance, Nwosu et al. (2001) reported a 34.8% prevalence of VVC among HIV/AIDS patients, while Ovelese et al. (2005) and Akerele et al. (2002) highlighted high infection rates among STI clinic attendees and antenatal women, respectively. These studies indicate that Candida albicans is a common pathogen in various settings, emphasizing the need for ongoing surveillance and research. Understanding the pathogenic mechanisms of Candida albicans is crucial for developing effective treatments and preventive strategies. The yeast's ability to adapt to environmental changes, its morphological diversity, and its capacity to form biofilms and secrete hydrolytic enzymes all contribute to its virulence (Nicholls et al., 2011). Symptoms of vaginal candidiasis typically include thick vaginal discharge, intense itching, redness, burning during urination, and soreness (Nicholls et al., 2011).

In light of these observations, the present study aims to investigate the prevalence and characteristics of vaginal mycotic infections among Obafemi Awolowo University students. While the prevalence and impact of *Candida albicans* in vaginal infections are well-documented, there remains a need for detailed studies to fill gaps in our understanding of fungal communities and their roles in vaginal health. The study will employ techniques such as culturing vaginal samples on Potato Dextrose Agar and performing wet mount microscopy to isolate and identify *Candida albicans* and other fungal species. This research is expected to provide valuable insights into the local epidemiology of vaginal candidiasis and contribute to the broader understanding of fungal infections in the vaginal environment. This study also seeks to address these gaps by focusing on the specific fungal profiles and prevalence among university students, which may have implications for clinical practice and patient management.

Materials and Methods

Study Population

The study involved 116 students from Obafemi Awolowo University, Ile Ife, Osun State, with participants aged between18 and 25 years. From July to September 2023, 20 students visited the university health center for vaginal secretion examinations, irrespective of Vulvovaginal Candidiasis (VVC) symptoms. Participants completed a standardized questionnaire detailing symptoms like vaginal discharge, itching, burning, dysuria, and dyspareunia. Ethical consent was obtained, ensuring the confidentiality and research-only use of samples.

Sample Collection

Samples were collected either by self-collection or by a healthcare provider. For self-collection, participants washed their hands and adopted a comfortable posture. The swab was carefully inserted into the vaginal opening, rotated for 15 seconds, and withdrawn without touching surrounding skin. Samples were then placed in a sterile tube, sealed, and transported to the laboratory. Samples were stored at 4°C to ensure sample integrity and prevent the growth of contaminants and analyzed within 36 hours.

Materials and Methods

Bench surfaces were disinfected using cotton wool soaked in a suitable disinfectant. For the preparation of culture media, 39 grams of Potato Dextrose Agar was dissolved in 1 liter of distilled water in a conical flask. The mixture was then heated and sterilized by autoclaving. Additionally, 13 grams of Nutrient Broth and 23 grams of Nutrient Agar were prepared in the same manner and autoclaved. After sterilization, the Potato Dextrose Agar and Nutrient Broth were allowed to cool to 45°C before being poured into sterile Petri dishes and McCartney bottles, respectively.

Fresh samples, less than 24 hours old, were spread onto the surface of the Potato Dextrose Agar plates and incubated at 37° C for 5 to 7 days to allow for the growth of fungal colonies. Older samples were first transferred to Nutrient Broth and incubated at 37° C for 72 hours to promote fungal growth. These samples were then streaked onto Potato Dextrose Agar plates and incubated under the same conditions.

Colonies that developed on the Potato Dextrose Agar plates were subcultured onto Nutrient Agar to obtain pure isolates. The subcultured plates were incubated at 37°C for an additional 5 days. Pure cultures of *Candida albicans* were subsequently stored in McCartney bottles containing Nutrient Agar at 4°C for further analysis.

Microscopy

Yeast morphology was examined using a microscope. Cultured samples were transferred to glass slides with forceps, and a drop of lactophenol cotton blue stain was added. Cover slips were placed, and slides were viewed under the microscope at X40 and X100 magnifications. Yeast structures such as budding yeasts, hyphae, pseudohyphae, chlamydospores, and blastospores were identified and recorded.

For further identification, pure yeast cultures were subjected to germ tube testing, which is a standard test for identifying *Candida albicans*. Additionally, carbohydrate assimilation tests were performed to distinguish *Candida albicans* from other yeast species. The germ tube test was carried out by incubating the yeast culture in human serum at 37°C for 3 hours, and the formation of germ tubes was observed under the microscope, confirming the presence of *Candida albicans*.

Results and Discussion

Results

Statistical analysis of respondents' feedbacks from the questionnaire showed that sexual activity, antibiotic use and contraceptive use gave the highest levels of significance at 0.006, 0.024 and 0.037 respectively (Table 1). The strongest association in the table is sexual activity and this could be due to various factors such as pH changes, micro-abrasions, or transmission of yeasts during sexual activity.

Antibiotic use also showed a significant association with VVC symptoms (p = 0.024). This aligns with clinical knowledge that antibiotics can disrupt vaginal flora, potentially leading to yeast overgrowth. The use of estrogenbased contraceptives is significantly associated with VVC symptoms (p = 0.037). This can be explained by the fact that hormonal changes can affect vaginal environment and susceptibility to yeast infections. Neither smoking (p = 0.072) nor drinking (p = 0.090) showed a statistically significant association with symptoms of Vulvovaginal Candidiasis (VVC). However, the p-values are close to the significance threshold (0.05), suggesting a potential trend that might be worth exploring in larger studies.

Laboratory analyses revealed that 101 samples exhibited positive growth on Potato Dextrose Agar (PDA) (Table 2). Most colonies were white to cream-colored, smooth, and yeast-like. After 48 hours, four isolates were identified as blastospores with some pseudohyphae. After 96 hours, hyphae growth was observed in three isolates, with chlamydospores and pseudohyphae in some. Budding yeast was the most common morphological feature, present in 68.3% of all samples.

Of the 116 participants, 11.2% had asymptomatic vaginal candidiasis, while 9.6% were symptomatic. Notably, 31% displayed symptoms but tested negative for candidiasis, potentially due to lifestyle factors like yogurt consumption, prolonged antibiotic use, or hormonal contraceptives. Normal growth was observed in 35.3%, reflecting Candida as part of normal vaginal flora, while 12.9% had no growth, possibly due to sampling errors (Table 3).

| Table 1. Association Analysis of Key Variables (Chi- | | | | | | |
|--|--|--|--|--|--|--|
| square test results) based on Respondents feedback and | | | | | | |
| Symptoms (Note: Significance level set at p < 0.05.) | | | | | | |

| Habit | Chi- | Р- | Significance | |
|-----------------|--------|-------|--------------|--|
| | Square | Value | | |
| | Value | | | |
| Smoking | 3.24 | 0.072 | Not- | |
| - | | | Significant | |
| Drinking | 2.87 | 0.090 | Not- | |
| | | | Significant | |
| Sexual Activity | 7.56 | 0.006 | Significant | |
| Antibiotic use | 5.12 | 0.024 | Significant | |
| Contraceptive | 4.35 | 0.037 | Significant | |
| use | | | - | |
| Yoghurt | 1.98 | 0.159 | Not- | |
| Consumption | | | Significant | |

 Table 2. Statistical Summary of Morphology and Microscopy Results

| Morphological | Number of | Percentage |
|---------------|-----------|------------|
| Feature | Positive | (%) |
| | Samples | |
| Budding Yeast | 69 | 68.3 |
| Hyphae | 3 | 2.9 |
| Pseudohyphae | 16 | 15.8 |
| Chlamydospore | 9 | 8.9 |
| Blastospore | 4 | 3.9 |

Table 3. Breakdown of participants' by prevalence rates

| Asymptom atic | Symptom atic | Sympt om but no infectio n | Norm al growt h | No growt h |
|------------------|-----------------|--|--------------------------|--------------------|
| 13 students | 11students | 36 student | 41 studen ts | 15 studen ts |
| 11.2% | 9.6% | 31% | 35.3% | 12.9% |

Total Percentage = 10

Discussion

The study revealed a higher prevalence of vaginal candidiasis among sexually active women aged 18-25 years, with varying symptoms such as itching, burning, and discharge, indicative of the diverse clinical presentations of *Candida* infections. The detection of asymptomatic cases underscores the necessity for routine screening, as many infections may remain undiagnosed due to a lack of overt symptoms.

This finding aligns with Jombo *et al.*, (2010), who reported a high incidence of vaginal candidiasis among sexually active adults, supporting the notion that while it is not exclusively sexually transmitted, sexual activity plays a significant role in its prevalence. Similar results were observed by Takahashi *et al.*, (2017), who highlighted that younger adults are particularly vulnerable to these infections, often due to hormonal changes and sexual behaviors. This study's emphasis on asymptomatic infections resonates with the work of Martins *et al.*, (2014), who also advocated for comprehensive sexual health education and timely medical intervention to manage and prevent such infections.

Comparing with more recent research, a study by Faronbi *et al.*, (2020) also found a strong correlation between sexual activity and the incidence of vaginal candidiasis, reinforcing the need for targeted STD control measures. Additionally, Wang *et al.*, (2019) emphasized the increasing challenge of antifungal resistance, particularly among *Candida albicans* strains, which complicates treatment and underscores the need for more effective therapeutic strategies.

The virulence of *Candida albicans* in this study is consistent with earlier findings by Granger (1992) and Donna (2009), where its ability to switch from a yeast to filamentous form enhanced its colonization of the vaginal epithelium. Recent studies by Silva *et al.*, (2021) have further elaborated on the pathogenic mechanisms of *C. albicans*, including its enzyme secretion capabilities like phospholipase production, which contribute to its persistence and resistance to antifungal treatments.

Moreover, the link between *Candida* infections and other pathogens, such as *Mycoplasma hominis*, *Ureaplasma urealyticum*, *Chlamydia trachomatis*, and *Trichomonas vaginalis*, as noted by Dadar *et al.*, (2018), suggests that co-infections might complicate diagnosis and treatment. Future research should investigate these co-infections to enhance diagnostic accuracy, as supported by recent advancements in diagnostic methods like Oricult-N Dipslide and amplified polymorphic DNA techniques (Noble *et al.*, 2017; Zhou *et al.*, 2021).

Conclusion

This study identifies *Candida albicans* as the predominant fungal species responsible for vaginal mycotic infections among students, with a significant prevalence of symptomatic vaginal candidiasis observed, particularly among sexually active individuals. These findings emphasize the need for early detection and intervention to manage and reduce the burden of vaginal candidiasis in this population.

Conflicts of Interest

The authors declare no conflict of interest

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