
VULVO VAGINAL CANDIDIASIS: IMPORTANCE OF SPECIES IDENTIFICATIONM. Swarajya Lakshmi¹, G. Jyothi Lakshmi²**HOW TO CITE THIS ARTICLE:**

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ABSTRACT: OBJECTIVES: Vulvo Vaginal Candidiasis is a common nagging problem faced by 75% of women in reproductive age group. Present study was undertaken to determine the prevalence of Candida in patients suffering from vaginitis, to assess predisposing factors and correlate the symptoms with gram stain for presumptive diagnosis of Candidiasis. **METHODS:** A prospective study of the laboratory diagnosis of vulvovaginal candidiasis (VVC) was carried out in 100 women presenting with symptoms suggestive of vaginosis in the reproductive age group. Investigations included microscopy and culture for yeast. Candida is identified, based on growth on SDA, corn meal agar and Sabaraud's Triphenyl tetrazolium agar, and assimilation and fermentation of sugars. **RESULTS:** Candida was isolated in 33% of women. Clue cells on gram stain suggestive of bacterial vaginosis was seen in equal number of women, whereas mixed infection was found in 9%. Candida albicans accounted for 15% and nonalbicans species for 85%. Of the non albicans species, Candida glabrata was the commonest (42%). Pruritus with or without vaginal discharge and vaginal erythema were the most common symptoms and signs in women with positive Candida culture. **CONCLUSION:** On comparing the significance of gram stain and culture for presumptive diagnosis of candidiasis, culture was more significant than gram stain alone. In present study, the rate of culture positivity was 33% and C. glabrata was the predominant species. VVC cannot be diagnosed by clinical criteria alone and requires confirmation by culture including identification of species.

KEYWORDS: Vulvovaginal candidiasis, Candida albicans, Candida glabrata.

INTRODUCTION: Vulvovaginal candidiasis is a common fungal infection that affects healthy women of all ages. At least 75% of women develop one or more infections once during their lifetime with 5% of those developing recurrent type ¹ The etiology may be infectious, non-infectious or a combination of both. Yeast like fungus Candida is the commonest cause of infectious vulvovaginitis. Although Candida albicans is the most commonly found species, there is a significant increase in prevalence of non albicans Candida ² producing vulvovaginal pruritus, burning, irritation, soreness, dysuria, dyspareunia, and a cheesy white discharge.

Unfortunately, none of these symptoms either individually or collectively is pathognomonic of Candida infection. Lack of specific signs and symptoms precludes a diagnosis that is based on history and physical examination without corroborative evidence of laboratory tests. A study at Detroit Medical Center's Candida Vaginitis Clinic showed over 80% of clinically suspected cases of VVC were found to have some other cause of vaginitis ³.

In a study of patients with self-diagnosed VVC, more than half did not have yeast confirmed as a causative organism. In fact, the most common reason the patient failed to respond to antifungal therapy is incorrect diagnosis³. The present study was therefore undertaken to determine the rate of positive cultures of Candida species in relation to clinical manifestations in women presenting with vulvovaginal symptoms.

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MATERIALS AND METHODS: A prospective study over a period of one year was conducted at Gandhi Medical College / Hospital, Musheerabad, Secunderabad.

Patient population: Among women attending Gynecology OPD a total of 100 patients were included. Inclusion criteria for the study were, married and sexually active women between 18-49 years of age who presented with self-reported symptoms of vaginal discharge, pruritus, irritation, soreness, dysuria, and dyspareunia, not using any vaginal medication in previous 48 hrs. Pregnant women, women with medical disorders, women currently menstruating, never been sexually active, who had hysterectomy, and taken a course of antibiotics within preceding two weeks, and who had been previously enrolled in this study were excluded. Written informed consent was obtained from each participant.

Two sterile cotton tipped swabs were used to collect specimen from the lateral wall of vagina of each women. One swab was used for microscopy; to do a 10% KOH wet mount and Gram stain. The second swab was used for culture on two plates of Sabourauds Dextrose Agar (Hi media Mumbai India) supplemented with 0.6micro gms / ml Gentamicin, with and without cycloheximide (0.5%).

Identification: Species identification of yeast isolates was done by standard procedures including morphology, germ tube test, (figure 1), corn meal agar – production of chlamydospores (figure 2), (Hi media, Mumbai, India) Triphenyl Tetrazolium reduction (Hi media Mumbai, India), assimilation of various sugars and growth in presence of actidione.

RESULTS: Of 100 patients included in the study, majority of patients presenting with symptoms of vaginitis were in the age group of 21- 30yrs. 86% of them presented with pruritus and vaginal discharge whereas 14% of patients presented with pruritus alone. On examination, vaginal erythema was found to be more common in patients with positive culture than with negative culture. In this study it was observed that majority of patients lack recognizable predisposing factors.

Microscopy of yeast was positive in 24% of cases and an equal number showed presence of clue cells suggestive of bacterial vaginosis. Mixed infection was seen in 9% of cases. Culture is the gold standard for diagnosis and in our study, 82.5% of culture positive showed presence of yeast on microscopy.

Nonalbicans Candida showed much higher prevalence (85%) as compared to Candida albicans (15%). Among the Nonalbicans Candida species, *C. glabrata* was the commonest (42%) followed by *C.krusei* (12%), *C.parapsilosis* and *C.stellatoidea*(9% each) *C.guellarmondi*(6%) and *C.kefyr* (3%).

DISCUSSION: History of itching was given only by 13 women. H/O recurrence was given by 14 of which *C.glabrata* was commonest followed by *C.albicans*. Signs of erythema and excoriation were seen in only 2 patients. Only one patient was diabetic in whom *C.glabrata* was isolated. 2 patients presented with ectopic gestation and positive culture. In this study it was observed that majority of patients lack recognizable predisposing factors like they were not under oral contraceptives, exogenous hormones or diabetic or on antibiotic therapy.

Our study did not find any association between colony count and symptoms evaluated, as published by Lopez et al in 2004.¹

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On comparing gram stain and culture in diagnosing candidial vaginitis, significance of gram stain was 82.5 % only, while culture on SDA was 100%.

Despite availability of several antifungal agents, and new insights into host-fungus interplay, Candida vaginitis remains a common and frequently distressing infection affecting millions of women worldwide. On the basis of the clinical symptoms and the predicted response to antifungal therapy, in 1997 a new classification of Candida vaginitis, into uncomplicated (VVC) and complicated (severe or recurrent VVC, Nonalbicans Candida species or abnormal host) disease was recommended.⁴

Methods used for diagnosis of Vulvovaginal Candidiasis are variable. Currently culture is the most sensitive method for accurate diagnosis. Two other simple tests are estimation of vaginal pH and microscopic examination of vaginal secretions³. In the present study culture was positive for 33% of cases.

Microscopy for yeast and pseudohyphae was found to be positive in 82.5% of culture positive women. Sobel reported 65-85% sensitivity of direct microscopy which could be because of the difference in yeast concentration of vaginal secretions. Direct microscopy is reliable only if infection is heavy. Therefore in cases where microscopy fails, vulvovaginal swab culture becomes essential to arrive at a diagnosis of VVC. The importance of vaginal swab culture before starting treatment cannot be overemphasized³. Culture is also essential for identification of various candida species. A 1996 epidemiological study including a high prevalence of candidemia in patients with neoplasms reported an increase of nonalbicans Candida species. Candida glabrata is found in candiduria cases with an increasing frequency especially in diabetic patients, patients who receive multi antibiotic treatment or patients who have a urinary catheter.² In present study, Candida albicans is 15% and nonalbicans Candida is 85% of infections of which Candida glabrata was 42%.

Recently several authors have reported an increase in incidence of nonalbicans species of candida.^{5,8,10,11}, The highest among them is C.glabrata which correlates with other authors^{2,5,6,7}. These nonalbicans yeasts are relatively non-pathogenic but ultimately get selected and start appearing more frequently because of the wide spread abuse of over the counter antifungals, use of single dose oral and topical azole regimens, and long term maintenance regimens of oral azoles.³ Some host alterations that contribute to C.glabrata infections development are a decrease of vaginal secretory IgA, low inflammatory response and a quantitative/qualitative decrease of T-cells, which explains its higher prevalence in patients with AIDS, transplants or neoplasms.² Therefore vaginal culture is valuable not only for identifying the species of Candida but also for monitoring the changing trends in microbiology of VVC which is essential for the complete and prolonged treatment of symptomatic women.³

Study of various putative factors responsible for VVC showed that culture positivity for Candida was significantly related to pregnancy and increase in parity as reported by other authors. High level of reproductive hormones present during these conditions might be providing an excellent carbon source for the growth of Candida organisms by providing higher glycogen content in the vaginal tissue.

Of the various symptoms, pruritus was the commonest among those who were culture positive. Sobel also reported vulval pruritus as the most frequent clinical manifestation of VVC. The burning sensation of vulval epithelium reported frequently by women suffering from VVC is caused by yeast's metabolites and seldom by infection of vulvar skin. Therefore it is suggested that treatment in such patients be directed to vaginal source of infection and not to the vulvar area. Applying

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antifungal preparations to the vulva is not only ineffective but also worsens the contact dermatitis which is a feature of the complaint. At times vulvar area may present with mixed pathology and the commonest combination is vulvar dermatitis exacerbated by bouts of candidiasis. Vaginal swab culture is the only means of selecting the appropriate treatment in such patients.³

CONCLUSION: Majority of patients presenting with symptoms of vaginitis were in the age group of 21- 30yrs. Gram stain was more effective than wet mount to identify *Candida* from vaginal swab. Culture is the gold standard for diagnosis and in our study microscopy for yeast was positive in 82.5% of culture positive. *Candida albicans* was isolated in 15% of women and non *albicans* *Candida* in 85%. *Candida glabrata* was the most common nonalbicans species isolated. It is important to establish the relationship between recurrence of VVC and nonalbicans *Candida* species. Mycological tests should be carried out to determine the species. If results report *C.glabrata*, long term treatment will be justified to eradicate the mycosis or to perform microbiological sensitivity tests to provide a specific treatment.

VVC cannot be definitely identified by clinical criteria alone. Culture is valuable not only for the accurate diagnosis of VVC but also to avoid indiscriminate use of antifungal agents. In conclusion, our study shows a significant increase of vulvovaginitis from *C.glabrata* compared with reports from previous years. Therefore, we consider this an emergent change for this species. Fortunately, available techniques to identify this species are accessible.

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| Age – yrs. | Number of patients. |
|------------|---------------------|
| 15 – 20 | 03 |
| 21 – 30 | 29 |
| 31 – 40 | 08 |
| 41 – 50 | 01 |

Table 1: Distribution of Candida isolates in different age groups.

| Method | Infection | % |
|---------------------------------|---------------------|-----|
| Wet mount | Trichomoniasis | Nil |
| Gram stain(budding yeast cells) | Candidiasis | 24 |
| Gram stain – clue cells s/o | Bacterial vaginosis | 24 |
| Clue cells + yeast cells | Mixed infection | 09 |

Table 2: Etiology of vaginitis by grams stain and wet mount

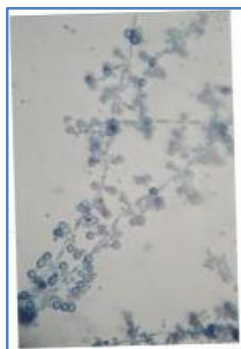
| Symptom/sign | No.of patients. | % |
|-------------------------------------|-----------------|------|
| Pruritus with or without discharge. | 29 | 88 |
| Vaginal erythema | 20 | 60.6 |

Table 3: Symptoms and signs among women with positive Candida culture.

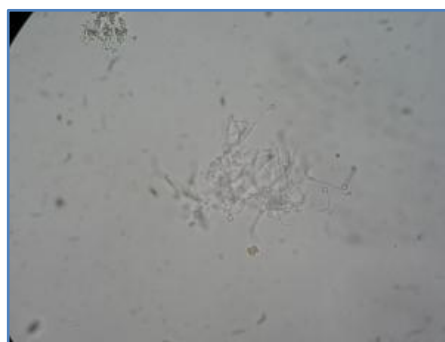
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| Candida species. | Number | % |
|-------------------|--------|-----|
| C. albicans | 5 | 15 |
| C. glabrata | 14 | 42 |
| C. krusei | 4 | 12 |
| C. parapsilosis | 3 | 9 |
| C. stellatoidea | 3 | 9 |
| C. guilliermondii | 2 | 6 |
| C. kefyr | 1 | 3 |
| Unidentified | 1 | 3 |
| Total | 33 | 100 |

Table 4: Prevalence of Candida isolates in study group.



chlamydospores on corn meal agar



germ tube

AUTHORS:

1. M. Swarajya Lakshmi
2. G. Jyothi Lakshmi

PARTICULARS OF CONTRIBUTORS:

1. Associate Professor, Department of Microbiology, MNR Medical College.
2. Associate Professor, Department of Microbiology, Gandhi Medical College, Musheerabad, Secundrabad.

NAME ADDRESS EMAIL ID OF THE CORRESPONDING AUTHOR:

Dr. M. Swarajya Lakshmi,
Associate Professor,
Department of Microbiology,
MNR Medical College Sangareddy Medak-502294,
Andhra Pradesh.
E-mail: drswarajya@yahoo.com

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