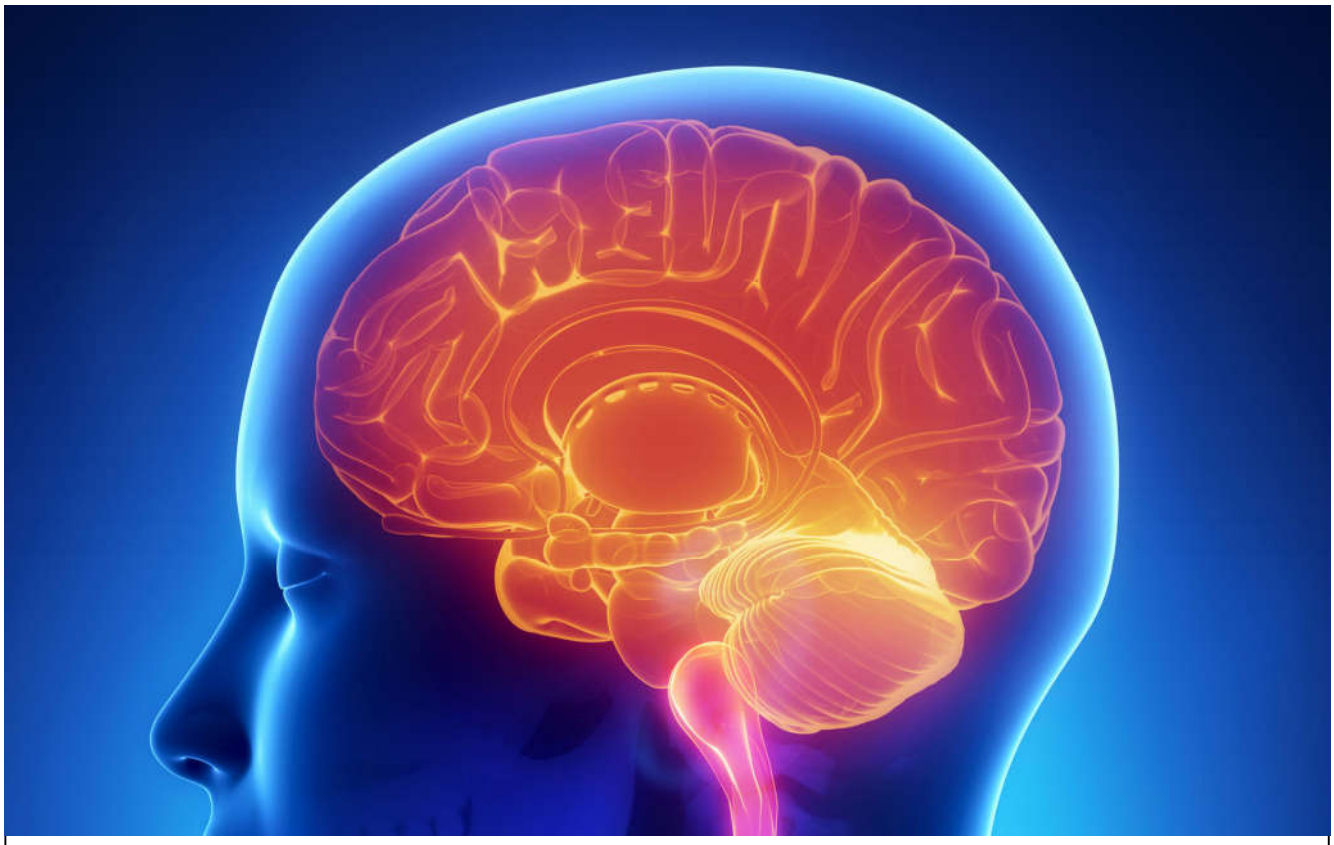

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Sharma Bhawna, Dey Sangeeta and Ganguly Udayan

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RESEARCH ARTICLE

VULVOVAGINAL CANDIDIASIS IN WOMEN OF REPRODUCTIVE AGE GROUP: IMPORTANCE OF PROPER DIAGNOSIS AND ALARM FOR EMERGING NON-ALBICANS CANDIDA AMONG CANDIDAL VULVOVAGINITIS CASES

Sharma Bhawna[1*], Dey Sangeeta[2] and Ganguly Udayan[3]

[1]Department of Microbiology, Vardhman Mahavir Medical College and associated Safdarjang Hospital, New Delhi
[2,3]Department of Microbiology, KMCH, Katihar, Bihar

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ABSTRACT

Vulvovaginal candidiasis is caused by overgrowth of *Candida* yeast species in the vagina. The most common clinical manifestations of VVC are pruritus, hyperemia, vaginal discomfort etc. Our study was aimed to ascertain the prevalence of vulvovaginal candidiasis and speciation of the *Candida* species with the help of culture and biochemical tests in the women attending Obstetrics and Gynaecology outpatient department (OPD). Isolation, Gram's stain, culture and bjo chemical tests were done for identification. Out of 93 samples 62 showed growth of organism. Out of 62 grown organisms 56.45% were *candida* isolates and rest were *Trichomonas vaginalis*, *Gardnerella vaginalis* and enterobactericeae family organism (which were usually considered as faecal contaminants). The most common candida species was *Candida albicans* (80%) followed by *C. intermedia* (8.58%), C. *k*rusei (5.71%) and C. *guillermondii* (5.71%). On the basis of gram's stain findings, out of 93 examined smears only 37.63% showed presence of budding yeast cells with pseudohuphae. So this study depicts the importance of culture for right diagnosis and also alarms the emergence of non-albicans species in the vaginal discharge.

INTRODUCTION

Vulvovaginal candidiasis (VVC) is defined as signs and symptoms of inflammation of the vulva and vagina in the presence of *Candida* spp. And in the absence of other infectious etiology VVC is caused by overgrowth of *Candida* yeast species in the vagina and is characterized by curd-like vaginal discharge, itching, and erythema [1]. VVC often referred to as a yeast infection and it is a common gynecologic aliment, affecting 3 out of 4 women in their lifetimes [2]. Candida is now the second most common cause of vaginal infections.75% of women experience at least one episode of vulvovaginal candidiasis during their childbearing years and approximately 40 to 50% of them experience a second attack. Candida species may be isolated from genital tract of approximately 20% of asymptomatic healthy women of childbearing age [3]. The most common clinical manifestations of VVC are pruritus, hyperemia, vaginal discomfort and leucorrhea, burning, soreness, dyspareunia and vaginal or vulvar erythema, which may cause a problem in marital and sexual relations [4]. The vaginal vault is colonized within 24 hours of a female child s birth and persists until death, comprising an ever-changing and fine-tuned ecosystem with numerous factors that have the potential to destruct the vaginal ecosystem [5]. Pregnancy, use of high estrogen oral contraceptive pills, steroids, antibiotics, chemotherapy drugs and aging favour its growth. Increased secretion of estrogen during pregnancy results in increased amount of glycogen in the vagina that acts as a good source of carbon needed for *Candida* growth and germination. Fortunately the infection is rarely life threatening, whereas it is usually associated with such morbidities like discomfort, pain, sexual dysfunctions, vulvar dryness, cracks, itching, burning, soreness and finally health care costs [6-8]. There is a balance between *Candida*, normal bacterial flora, and immune defence mechanisms. When this balance is disturbed, colonization is replaced by infection. It is possible that there are multiple mechanisms by which *Candida* can cause cell damage and lead to direct invasion of hyphae in epithelial tissues. During vaginal candidiasis, vagina is in the normal pH range (pH 4- 4.5) [9]. Women are usually ignorant about their gynaecological problems which lead to them into chronic cases, so our study was aimed to ascertain the prevalence of vulvovaginal candidiasis and speciation of the *Candida* species with the help

Corresponding author:* **Sharma Bhawna
Department of Microbiology, Vardhman Mahavir Medical College and associated Safdarjang Hospital, New Delhi

of culture and biochemical tests in the women attending Obstetrics and Gynaecology outpatient department (OPD).

MATERIAL AND METHODS

The present study was conducted between January 2010 to April 2011.A total number of 186 samples of vaginal swabs were collected from 93 patients attending Gynaecology & Obstetrics OPD of KMCH, Katihar, Bihar, India. After selection of cases, a brief clinical history regarding occupation, personal hygiene, menstrual cycles was noted down. Obstetric history and history of any antifungal usage either systemically or locally was also taken from these patients. Vaginal swabs were collected under speculum examination. For collection of vaginal swabs, sterile cotton swabs were inserted into the upper part of the vagina and rotated to collect any discharge before withdrawing it taking adequate precautions not to touch the lower vaginal walls. Two swabs were collected from each patient; one swab was used for microscopy and the other for culture. The swabs were sent to the laboratory for processing immediately.

Gram's stained examination of the slide

Thin smears were made from the swabs then air dried, heat fixed and Gram stained. After drying the smears, they were examined under the oil emersion lens to look for the presence of pus cells, yeast like organisms with pseudohyphae and for number of lactobacilli present.

Culture

Routine culture was performed on Sabouraud's Dextrose Agar (SDA). One tube is incubated at 25° C and another tube at 37° C in adequate environment of incubator. The tubes were examined after 48 hours of incubation for any growth. All positive tubes showed growth of candida species after 48 hours of incubation. Then they were again gram stained to confirm the candida species growth. After that their speciation was done by performing battery of tests like – germ tube formation, chlamydospore production, sugar fermentation and sugar assimilation test (by using 10% of sugar disc).

RESULT

Out of a total of 93 samples, 31 samples showed no growth while 62 samples showed growth of various *Candida* species and bacterial isolates. Out of 62 positive findings, 35 isolates were *Candida* species that is 56.45% of the total isolates. Others organisms are as follows- *Trichomonas vaginalis, Gardnerella vaginalis* and enterobactericeae family organism (which are usually considered as the faecal contamination of vagina.

Table 1 Isolation pattern of *Candida* species from clinical samples

Organisms	No of organisms	% of organisms
Pure *Candida* species	62/28	45.16
Candida sp. with others	62/7	11.29
Total	62/35	56.45

Table I shows the distribution of candida species as a pure sole isolate and a mixture with other organisms.

Table 2 Gram's stain findings of vaginal discharge

Findings	Total no gram stain findings	% of gram stain findings
Clue cells	10	10.75
Yeast like cells	35	37.63
No relevent findings	48	51.62
Total	93	100

Table 2 shows the Gram's stain finding in which 51.62% of samples showed no specific finding but 37.63% budding yeast like cells with pseudohyphae and 10.75% showed clue cells from the vaginal discharge samples.

Table 3 Distribution of *Candida* species according to the Germ Tube formation

Germ tube formation	No of *candida* sp.	% of *candida* sp.
Positive result	30	85.71
Negative result	5	14.29
Total	35	100

Table 3 shows the distribution of candida species according to the germ tube formation. Out of 35 different candida isolate, 30 (85.71%) samples showed germ tube formation while only 5 (14.29%) samples had not shown germ tube formation.

Table 4 Distribution of candida species according to chlamydospore formation

Chlamydospore formation	No of candida sp.	% of candida sp.
Positive result	25	71.42
Negative result	10	28.58
Total	35	100

Table 4 shows the distribution of candida species according to the chlamydospore formation. Out of 35 different species of candida, 25 (71.42%) isolates showed chlamydospore formation test positive while 10 (28.58%) isolates had shown negative chlamydospore test .

Table V shows the distribution of different candida species according to their ability to ferment different sugars, to show germ tube formation and chlamydospore formation. On the basis of these tests 35 different strains of candida were identified to species level. 28 of them were *C.albicans,* 3 were *Candida intermedia* and 2 each of *Candida krusei* and *Candida guilliermondii.*

Table VI shows the distribution of different candida species isolated from clinical samples. Out of 35 different isolates of candida species, maximum number of candida species were *C. albicans* (80%) followed by *C. intermedia* (8.58%), *C. krusei* (5.71) and *C. guilliermondii* (5.71 %)

DISCUSSION

Vaginal discharge, itching, and erythema, while quite common, were insufficient to diagnose vulvovaginal candidiasis in the absence of laboratory confirmation. We found a prevalence of 37.63% candidal vulvovaginitis in our setup, which was higher than the previous reports of 20.47% and 26.6% [10, 11]. In this study, Gram's staining from vaginal swab material showed budding yeast cells with pseudohyphae in all 35 cases, thus showing positivity in only 37.63% in direct sample of the

Table 5 Correlation of sugar fermentation test with germ tube test and chlamydospore formation test

Isolate no	Glucose	Sucrose	Maltose	Lactose	Germ tube test	Chlamydospore test	Candida species
1	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
2	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
3	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
4	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
5	A+G	A+G	A+G	neg	neg	neg	*C.intermedia*
6	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
7	A+G	A+G	A+G	neg	neg	neg	*C.intermedia*
8	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
9	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
10	A+G	A+G	neg	neg	neg	neg	*C.guilliermondii*
11	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
12	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
13	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
14	A+G	neg	neg	neg	neg	neg	*C. krusei*
15	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
16	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
17	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
18	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
19	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
20	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
21	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
22	A+G	neg	neg	neg	neg	neg	*C. krusei*
23	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
24	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
25	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
26	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
27	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
28	A+G	A+G	neg	neg	neg	neg	*C.guilliermondii*
29	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
30	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
31	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
32	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
33	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
34	A+G	A+G	neg	neg	pos.	pos.	*C.albicans*
					neg	neg	*C.intermedia*

Table 6 Distribution of different candida species isolated from clinical samples

Different candida sp.	No of diff. Candida sp.	% of diff. Candida sp.
C. albicans	28	80
C. intermedia	3	8.58
C. krusei	2	5.71
C. guilliermondii	2	5.71
Total	35	100

cases. Similarly, there are reports of 27.84% positivity of yeast from microscopic examination of direct specimen also [10] which is quite close to our result showing only gram's stain cannot diagnose a case of VVC. Another study from India showed 167 positive microscopic finding out of 1050 patients (15.90%) [11]. The difference in sensitivity of direct microscopy may be due to the difference in the concentration of yeast in different vaginal secretions [12, 13]. Direct microscopy is reliable only if the infection is fairly heavy [14]. With the issue like insufficient clinical diagnosis and failure of simple microscopy in screening cases other than severe cases, vaginal swab cultures are important for diagnosing VVC. Different laboratory tests revealed the presence of four *Candida* species in the present study, the full sequence of isolated candida species is as follows; *C.albicans* (80%), *C. intermedia* (8.58%), *C. krusei* (5.71%) and *C. guillermondii* is also (5.71%). According to a comparable study on 1050 women it was indicated that 215 women were associated with candidal vulvovaginitis and the highest frequency was related to *C.albicans* (46.9%) out of six *Candida* species [13]. Therefore in cases where microscopy fails to provide an answer, vulvovaginal swab culture becomes essential for elucidation of diagnosis of VVC. The importance of taking a vaginal swab culture before starting the treatment must be emphasized to the patients [14]. Antifungal therapy may then be prescribed before the availability of culture results. Elimination of yeast after therapy, together with simultaneous resolution of signs and symptoms verifies the diagnostic significance of positive candida culture.

The percentage of non-albicans species in this study were 20%, composing of *C.intermedia* (8.58%), *Candida gullermondii* (5.17%) and *Candida krusei* (5.17%). The highest proportion of nonalbicans candida reported is this study was *C. intermedia* (8.58%). Another study from India revealed the overall percentage of non-albicans vaginitis as 64.8% [15]. These studies showed the increasing trends of non-albicans *Candida*. Though the number of isolation of *Candida* species in the present study were less (three) than the previous studies who isolated six and seven species respectively [11, 16]. However, in all these studies predominance of non-albicans *Candida* was notably seen. The difference might be due to fewer patients enrolled in the study or the level of social activities, drug abuse, sexual promiscuity and environmental variation. These non albicans yeasts are relatively non pathogenic but ultimately get selected and start appearing more frequently because of the widespread abuse of over the counter antifungal, use of single dose oral and topical azole regimens, and long term maintenance regimens of oral azoles [17,18]. Therefore vaginal culture is valuable not only for identifying the species of vaginal candida but also for monitoring the changing trends in the microbiology of vulvovaginal candidiasis which is essential

for the complete and prolonged treatment of the patients of VVC.

CONCLUSION

Vulvovaginal candidiasis (VVC) cannot be definitely identified by clinical criteria alone. It requires culture for candida species and its correlation to vulvovaginal symptoms. Culture is valuable not only for the accurate diagnosis of VVC but also to avoid indiscriminate use of antifungal agents, which may ultimately decrease the incidence of VVC caused by resistant and nonalbicans candida species. The emergence of nonalbicans *Candida* spp. from VVC cases and their association with virulence factors cannot be overlooked. We suggest increased isolation and complete identification of *Candida* spp. in all microbiology laboratories so that the epidemiology, emergence and spread of non-albicans *Candida* could be revealed. Right identification prior to therapy is very much required because treatment of these cases is quite different from the *C.albicans* VVC cases and also it prevent their emergence drug resistant VVC [19].

References

1. Achkar JM, Fries BC. *Candida* infections of the genitourinary tract. ClinMicrobiol Rev. 2010; 23:253-73.
2. Das N J, Pinto E, Teixeira B, Dias G, Rocha P, Cunha T, *et al*. Local treatment of vulvovaginal candidosis: general and practical considerations. Drugs. 2008; 68(13):1787-802.
3. Ferrer J. Vaginal candidosis. Epidemiological and etiological factors. Intl J Gynecol Obstet. 2000; 71: S21-7.
4. Moreira D, Paula CR. Vulvovaginal candidiasis. Int J GynaecolObstet. 2006; 92:266-7.
5. Marshall W, Tanner J. Puberty. In: Davis J, Dobbing J., 1981. editors Scientific Foundations of Paediatrics. London: Heinemann
6. Ehrstrom S, Yu A, Rylander E. Glucose in vaginal secretions before and after oral glucose tolerance testing in women with and without recurrent vulvovaginal candidiasis. Obstet Gynecol. 2006; 108 (6):1432-7.
7. Nyirjesy P, Peyton C, Weitz MV, Mathew L, Culhane JF. Causes of chronic vaginitis: analysis of a prospective database of affected women. Obstet Gynecol. 2006; 108 (5):1185-91.
8. Paulitsch A, Weger W, Ginter-Hanselmayer G, Marth E, Buzina W. A 5-year (2000-2004) epidemiological survey of Candida and non-Candida yeast species causing vulvovaginal candidiasis in Graz, Austria. Mycoses. 2006; 49 (6):471-5.
9. J.L. Anderson, B. Cundiff, Schnars, Hypha formation in the white-opaque transition of *Candida albicans*. Infect. Immun. 1989; 57: 458–467.
10. Esmaeilzadeh S, Omran SM, Rahmani Z. Frequency and etiology of vulvovaginal candidiasis in women reffered to gynaecological center in Babol, Iran. Int J Fertil Steril. 2009; 3:74-7.
11. Ahmad A, Khan AU. Prevalence of *Candida* species and potential risk factors for vulvovaginal candidiasis in Aligarh, India. Eur J Obstet Gynecol Reprod Biol. 2009; 144:68-71.
12. Jindal N, Gill P, Aggarwal A. Significance of *Candida* culture in women with vulvovaginal symptoms. J ObstetGynecol India. 2006; 56:139-41.
13. Lopes Consolaro ME, AlineAlbertoni T, Shizue Yoshida C, Mazucheli J Peralta RM, EstivaletSvidzinski TI. Correlation of *Candida* species and symptoms among patients with vulvovaginal candidiasis in Maringá, Paraná, Brazil. Rev Iberoam Micol. 2004; 21:202-5.
14. Sobel JD. Vulvovaginal candidiasis. In: New York. Sexually Transmitted Diseases. 1999; 629-37.
15. Mohanty S, Xess I, Hasan F, Kapil A, Mittal S, Tolosa JE. Prevalence & susceptibility to fluconazole of *Candida* species causing vulvovaginitis. Indian J Med Res. 2007; 126:216-9.
16. Okungbowa FI, Isikhuemhen OS, Dede AP. The distribution frequency of *Candida* species in the genitourinary tract among symptomatic individuals in Nigerian cities. Rev Iberoam Micol 2003; 20:60-3.
17. Kent HL. Epidemiology of vaginitis. Am J Obstet Gynecol. 1991; 165:1168-76.
18. Graeme Dennerstein Review. The treatment of candida vaginitis and vulvitis. JAMA India. 2001;4: 50-2.
19. Sobel JD, Chaim W, Nagappan V, Leaman D. Treatment of vaginitis caused by *Candida glabrata*: Use of topical boric acid and flucytosine. Am J ObstetGynecol. 2003;189: 1297-300.

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