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RESEARCH ARTICLE

VULVOVAGINAL CANDIDIASIS AS AN UNDERESTIMATED COMPLAINT IN WOMEN OF REPRODUCTIVE AGE GROUP - A REVIEW OF 15 YEARS (2000-2015) FROM INDIA

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ABSTRACT

Vulvovaginal Candidiasis (VVC) is caused by overgrowth of Candida yeast species in the vagina. It is characterized by curd-like vaginal discharge, itching, and erythema. It affects 3 out of 4 women in their lifetimes. Pregnancy, use of high estrogen oral contraceptive pills, steroids, antibiotics, chemotherapy drugs and aging favour the growth of Candida species. Womwn are generally reluctant about VVC which leads to emergence of pelvic inflammatory diseases in them. Diagnosis of VVC is done by KOH wet mount, culture, germ tube test, chlamydospore test, and a battery of sugar fermentation and assimilation tests. A review of 15 years (2000-2015) has been done in this article. In conclusion of this review is that patients coming with pre disposing factors (usage of antibiotics, oral contraceptive drugs, hormonal therapy, and pregnancy) and co morbidities like diabetes mellitus, tuberculosis should also be examined for VVC in them.

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INTRODUCTION

Vulvovaginal candidiasis (VVC) is caused by overgrowth of *Candida* yeast species in the vagina and is characterized by curd-like vaginal discharge, itching, and erythema (Achkar *et al.*, 2010). 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 (Moreira *et al.,* 2006). Unfortunately, none of these symptoms either individually or collectively is pathognomonic of candida infection. The lack of specificity of symptoms and signs therefore precludes a diagnosis that is based on history and physical examination without the corroborative evidence of laboratory tests. 75% of women experience at least one episode of VVC 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 (Mohanty *et al*., 2007). It affects 3 out of 4 women in their lifetimes (Das *et al*., 2008). Greater than 40% of affected women will have 2 or more VVC episodes (Ferrer *et al.,* 2000, Eschenbach *et al.,* 2004). VVC has been associated with considerable direct and indirect economic costs enhanced susceptibility to HIV infection and is being investigated for a potential relationship with preterm birth (Foxman *et al.,* 2000, Røttingen *et al.,* 2001, Roberts *et al.,* 2011).VVC occurs due to disruption of the normal balance between Candida, bacterial flora and immune defence mechanisms leading to colonization that may occur during usage of broad spectrum antibiotics and hormonal fluctuations. Pregnancy, use of high estrogen oral contraceptive pills, steroids, antibiotics, chemotherapy drugs and aging favour its growth (Sobel *et al.,* 1998, Sobel *et al.,* 2005). 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), as opposed to mixed infections (bacterial, Trichomonas), where pH levels rises (Anderson *et al.,* 1989). Numerous studies around the world showed that *Candida albicans* is responsible for the largest number of symptomatic episodes of vaginal candidiasis. Percentage of non-albicans was high in the recent decades and varied from 85 to 90%.

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Culture

For fungal culture the Sabouraud dextrose agar with chloramphenicol (antibiotic) are used. The inoculated slant in incubated at 37C for 3-4 days and incubated for 7 days or more if required. Cream coloured colonies are seen on the slant. Make the Lactose phenol cotton blue (LPCB) mount to examine the yeast cell and pseudohyphae under the microscopes (Chander J, 3rd edi., Nerurkar *et al.,* 2012). CHROM agar for Candida isolation is rapid, plate based test for the simultaneous isolation and identification of various Candida species (Chander J, 3rd edi, Borg *et al.,* 1990). Study reported that the CHROM agar Candida is useful medium to differentiate the species based on colour development on CHROM agar Candida. Candida was differentiated by the colour produced by the medium as *C. dubliniensis, C. albicans, C. krusei, C. tropicalis, and C. glabrata* (Raut *et al.,* 2009).

Germ Tube Test (GTT)

The germ tube test is used for the identification of Candida albicans and differentiates the C. albicans from Candida species. This test is used for presumptive identification of Candida species (Chander J, 3rd edi., Nerurkar *et al.,* 2012).

Chlamydospore formation

Cornmeal agar (CMA) or rice starch agar (RSA) is used for detection of chlamydospore by inoculate the suspected strain of the Candida isolates and incubated at 25C for 2-3 days. The chlamydospores are highly refractile, thick walled, large in shape. These are seen in the case of C. albicans and C. dubliniensis (Chander J, 3rd edi).

Polymerase chain reaction (PCR) finger printing

PCR fingerprinting has introduced to differentiating the C. albicans from C. dubliniensis. Multiplex PCR has been used to identify various Candida species, particularly C. albicans, C. glabrata, and C. tropicalis in a single sample (Liquori *et al.,* 2009). RT-PCR can used for the detection of amplification of the reactions, measured the kinetics and has an advantage over the traditional PCR where agarose gels are used for the detection of PCR amplification at the final endpoint phase of the reaction.

Sugar assimilation and fermentation test

Different sugarslike glucose, sucrose, mannitol, xylose, arabinose etc are used. Different species od candida can assimilate and ferment different sugars and accordingly they are speciated (Rad *et al.,* 2011). A review of 15 years from 2000-2015 has been done for VVC and depicted in Table 1.

Table 1. Review of 15 years (2000-2015) of VVC cases from India

Author, Year, Place	Total no of cases	Prevalence of VVC	Most common affected age group between (20-40)	Techniques used for identification	Commonest Candida species	Predisposing factor	Treatment
Goswami, *et al*, 2000, New Delhi	78	46%	Yes	Direct microscopy, Gram,s stain, Culture, Biochemicals	*C. glabrata*	Diabetes mellitus	-
Jindal *et al*, 2006, Punjab	400	23%	Yes	Direct microscopy, culture	*C. albicans*	Pregnancy, use of broad spectrum antibiotics, contraceptive	
Jindal *et al*, 2007, Punjab	350	23.4%	Yes	Direct microscopy, Gram,s stain, Culture, Biochemicals	*C. albicans*	Use of antibiotics, OCP	
Mohanty *et al*, 2007, New Delhi	611	18.5%	Yes	Direct microscopy, Gram,s stain, Culture, Biochemicals	*C. glabrata*		Fluconazole
Tamsikar *et al,* 2011, Jabalpur	81	53.1%	Yes	Culture, biochemical tests	*C. glabrata*	Hormonal therapy	
Sujit D. Rathod *et al*, 2012, Mysore	898	35%	Yes	Culture			
Bhagat *et al,* 2013, Gujrat	135	80%	Yes				
Kumari *et al*, 2013, Varanasi	232	30.60%	Yes	Gram'stain, 10% KOH, Culture, Biochemical tests, Chrome agar	*C. glabrata*		
Tiyyagura *et al*, 2013, Hyderabad	100	43%	Yes	Direct microscopy, culture, biochemical	*C. albicans*		
Sasikala *et al,* 2013, Tamilnadu	200	36%	Yes	Gram'stani, Culture, Biochemical tests, chrome agar	*C. albicans*	Antibiotic usage	fluconazole, itraconazole and 5 flucytosine
Babin 2013 *et al*, Kerala	250	48.4%	Yes	Gram'stain, Culture, Biochemical tests, chrome agar	*C. albicans*	Pregnancy	amphotericin B, fluconazole, itraconazole, voriconazole, flucytosine
Ragunathan *et al*, 2014, Puducherry	118	22.2%	Yes	Direct microscopy, Culture, biochemical tests	*C. albicans*	Pregnancy, antibiotic use, contraceptive pills, Diabetes mellitus, tuberculosis	Fluconazole
Swarajya *et al,* 2014, Secundrabad	100	33%	Yes	Direct microscopy, Gram,s stain, Culture, Biochemicals	*C. glabrata*	Diabetes mellitus(1case), Unknown	
Shrivastava *et al*, 2015, M.P.	119	47.5%	Yes	Culture, biochemical, Chrome agar	*Candida krusei* in asymptomatic, *Candida albicans* in symptomatic	Diabetes mellitus	
Shailaja S. Dabashetty *et al.,* 2015, Karnataka	100	22%	Yes	Direct microscopy, culture		Antibiotic use, Pregnancy	

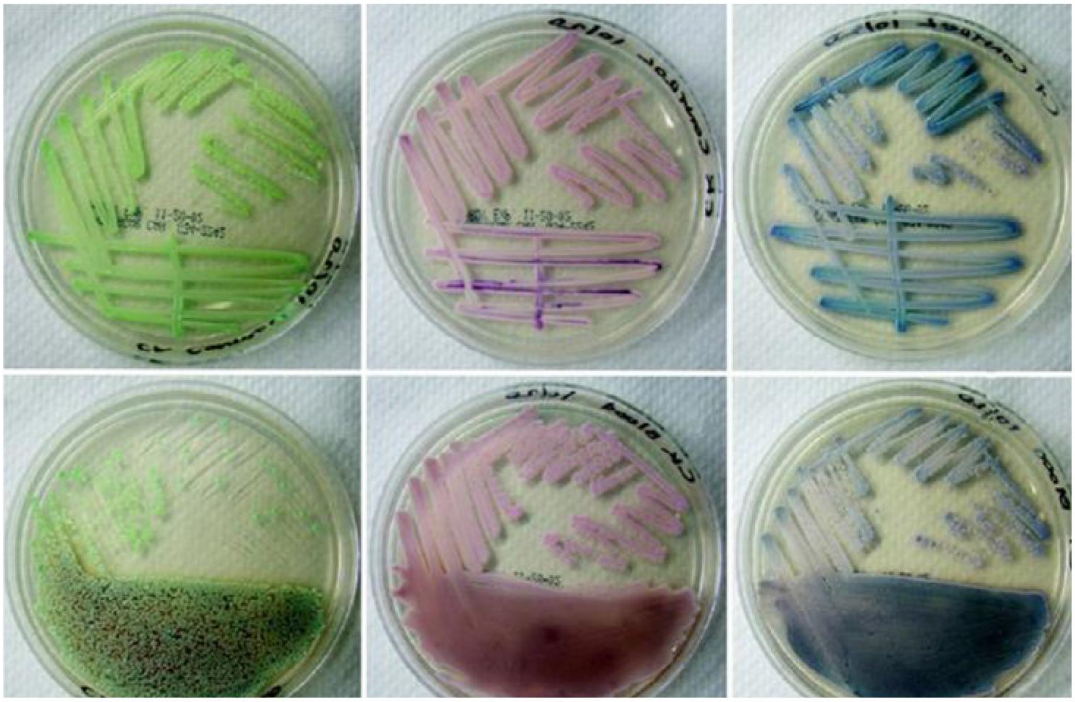

Figure 1.

Figure 2.
Figure 1, 2: shows CHROM agar Candida with different isolation of Candida species

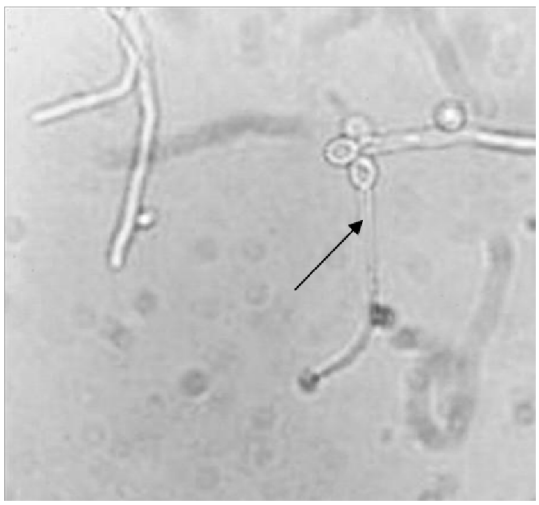

Figure 3. Germ tube for presumptive diagnosis of *C. albicans.*

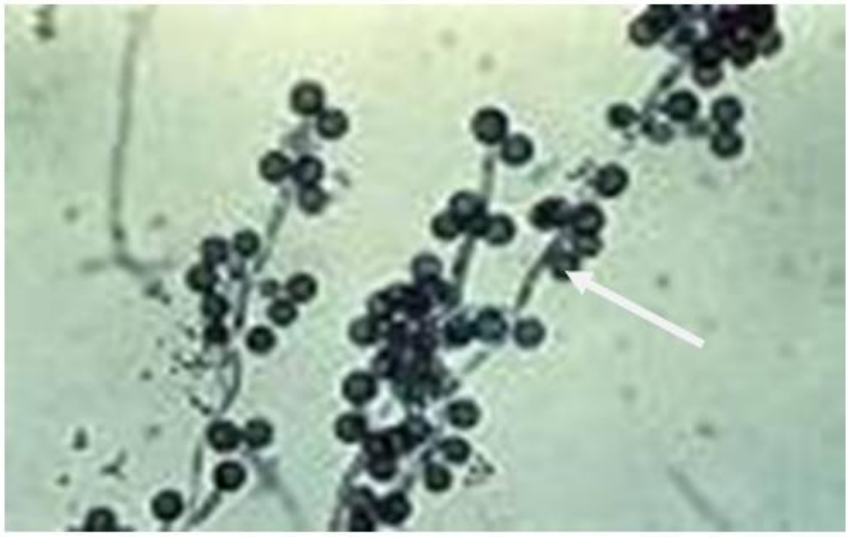

Figure 4. Chlamydospore formation after 2 to 3 days of incubation

DISCUSSION

We have done the review of VVC for 15 years (2000-1025) from India. Table 1 depicts the prevalence, most common age group, most common *Candida* species, potential predisposing factors and treatment taken by women of reproductive age group. The above table shows that the most common age group for VVC is the reproductive age group between 20-40 years and all the enrolled population complained of vaginal discharge, pruritus, dyspareunia, itching. They are those complaints which can be missed by any women until its leads to very uncomfortable condition to her health. So, any sort of these complaints should not be ignored. 20-40 years age group is the actual period for reproductive age group of life, so these age group women should be more careful regarding any above mentioned problems. The region behind this particular involved age group is that at this duration the sexual activity is more and the levels of estrogen hormone is also more as comparison to later or earlier age. The prevalence rate of VVC varies from 18-80% as shown in the above review Table 1. This wide range of prevalence shows the prompt and careful need for the diagnosis of VVC in the clinical microbiology set up. Those women who came with some other complaints to Gynaecology OPD or ward should also be questioned regarding any sort of vaginal discharge, pruritus, itching in the vaginal area, and the relevant sample should be send to the microbiology department for further investigations for VVC. In this way only we are able to look for the iceberg of VVC in the population. The above table also shows that the diagnosis of VVC cannot be only confirmed by the clinical complaints given by the patients, culture and biochemical tests has to be done not only for diagnosis but also for species identification. Use of chrome agar by different studies as depicted in table 1 also shows a good sensitivity for species identification (Roberts *et al.,* 2011; Røttingen *et al.,* 2001; Shrivastav *et al.,* 2015; Sobel, 2005). PCR has the highest susceptibility and sensitivity for VVC diagnosis, but is a sophisticated and expensive process. The female patients with co morbidities like diabetes mellitus, tuberculosis should also be put on regular investigation to rule out their VVC condition. These additional further investigations for VVC should be done for preventing these kinds of patients from further discomfort due to VVC of which they are not aware. The patients having predisposing factors like use of OCP, on hormonal therapy, use of excess antibiotics should also be regularly screened for their VVC conditions of which they are not aware.

It is also clear from the Table 1 that non albicans candida (NAC) is increasing gradually in this decade (Roberts *et al*., 2011; Røttingen *et al*., 2001; Sobet, 1997; Tamsikar, 2011; Tiyyagura, 2013). This also alarms us not to irrational use of antifungal drugs for VVC without species identification. NAC is more difficult to treat as comparison to Candida albicans and also there treatment regimen is different from C. albicans (Roberts *et al*., 2011). The focus should also be driven to the antifungal susceptibility tests for different candida species. We suggest proper isolation and identification of *Candida* spp. in female patients having co morbidities and predisposing factors should be done in all microbiology laboratories so that the epidemiology, emergence and spread of non-albicans *Candida* could be identified. Right identification prior to therapy is very much important because treatment of these cases is quite different from the *C.albicans* VVC cases and also it prevent their emergence antifungal drug resistant.

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