

Prevalence of Vulvovaginal Candidiasis and Trichomoniasis among Women of Reproductive Age in a Rural Tertiary Care Hospital in West Bengal, India

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Abstract

Introduction: Vaginal infections are common among women of reproductive age. Among the more frequent microorganisms are the *Trichomonas vaginalis* (TV) and *Candida* spp. Both of these sexually transmissible infections are associated with enhanced susceptibility to HIV infections and adverse health outcomes. Adequate investigation and proper treatment will reduce the risk of adverse outcomes. **Aim:** The aim of our study is to focus on the prevalence of *T. vaginalis* and *Candida* infection in non-pregnant women and also to verify the co-existence of these two agents and other social and demographic variables. **Materials and Methods:** The study was planned as a cross sectional observational study. 201 women with symptomatic vaginal discharge attending the Obstetric and Gynaecological clinic of Malda Medical College and willing to provide written informed consent, were included in the study. Three high vaginal swabs were collected following standard procedures and wet mount preparations, Pap smear, Grams staining, culture (on SDCA, cornmeal agar, candida CHROMagar), germ tube test, fermentation and assimilation of various sugars were carried out to determine the presence of TV and candida spp. **Results:** Candidiasis was present in 37.81% patients and 2.49% had *Trichomonas vaginalis* (TV). *C. albicans* (57.89%) was the most common species isolated, followed by Non albicans candida spp. (NAC spp., 42.11%). Among the NAC spp. *C. tropicalis* was the predominant isolate followed by *C. glabrata*. Co-infection with the two etiologic agents, TV and candida was not found. **Conclusion:** The high prevalence of genital tract infections warrants the implementation of constant health education, sensitization, adequate investigation and treatment, which will prevent adverse outcome of genital infections.

Key Words: vulvovaginal candidiasis, *Trichomonas vaginalis*, non pregnant women, socioeconomic

Date of Submission: 25-08-2018

Date Of Acceptance: 06-09-2018

I. Introduction

Vaginal infections are common among women of reproductive age. They can be caused by sexually transmitted microorganisms or by some disorder resulting in the proliferation of endogenous microbiota. Among the more frequent microorganisms are the protozoan *Trichomonas vaginalis* (*T. vaginalis*) and *Candida albicans* (*C. albicans*).¹

Trichomonas vaginalis, is an anaerobic flagellated protozoan parasite, that infects the human urogenital tract.² Infection in women can cause vaginitis, urethritis, and cervicitis.³ The disease encompasses a broad range of symptoms ranging from a state of severe inflammation and irritation with a frothy malodorous discharge to a relatively asymptomatic carrier state.⁴ Trichomoniasis is associated with adverse reproductive sequelae including preterm birth, pelvic inflammatory disease, and infertility in women.⁵ This parasite has also been implicated as a cofactor in the transmission of the human immunodeficiency virus (HIV) and other nonulcerative STD agents. In addition, a relationship between *T. vaginalis* infection and cervical cancer has recently been suggested.⁶ According to the World Health Organization, *T. vaginalis* is the most common curable sexually transmissible infection (STI) worldwide, with 170 million to 190 million new cases each year.⁷

Vulvovaginal candidiasis (VVC) is an opportunistic fungal infection of the female lower genital tract caused by *Candida* species.⁸ Numerous studies showed that *Candida albicans* is the common pathogen in 80-90% of cases but *Candida non-albicans* species are gaining importance as pathogens over the past few decades.⁹ It is clinically characterized by curd like discharge, itching, dyspareunia, dysuria, oedema and vulvovaginal erythema. The manifestations of VVC may range from asymptomatic colonization to severe acute symptomatic infection.¹⁰ Vaginal candidiasis if untreated can lead to chorioamnionitis with subsequent abortion and prematurity in pregnant women, congenital infection of the neonate and pelvic inflammatory disease resulting in infertility

in non-pregnant women.¹¹ Vulvovaginal candidiasis has been associated with considerable direct economic cost, enhanced susceptibility to HIV infection.¹²

As reduction of HIV transmission and of adverse birth outcomes remain public policy priorities in India – the aim of our study is to focus on the prevalence of *T. vaginalis* and *Candida* infection in non-pregnant women and also to verify the co-existence of these two agents and other social and demographic variables.

II. Aims And Objectives

1. To determine the prevalence of *T. Vaginalis* and *Candida* infection in women of reproductive age group attending the Obstetric and Gynaecological clinic in Malda Medical College, Malda, WB.
2. To describe socio-demographic factors related to these types of reproductive tract infections in women of reproductive age group.

III. Materials And Methods

A cross sectional observational study was carried out in the Department of Microbiology, Malda Medical College, Malda. A total of two-hundred one women with complain of symptomatic vaginal discharge, attending the Obstetric and Gynaecological clinic in Malda Medical College, Malda , were included in the study. The study proposal received ethical approval from the ethical committee of the Malda Medical college, Malda (West Bengal, India).

Study participants, Inclusion and exclusion criteria:

Married women of childbearing period (18-45yrs) with complain of vaginal discharge of any type, with itching, burning sensation or both, dysuria, inflammation of the genital tract, vaginal malodor and with any other gynaecological manifestations suggestive of trichomoniasis or vulvovaginal candidiasis (VVC) and willing to provide written informed consent, were included in the study. Those excluded from the study were pregnant, were menstruating, had received antibiotics in the past 3wks or refused to give consent.

Data collection procedure:

Socio-demographic data were collected using a semi-structured questionnaire. Three high vaginal swab specimens were collected by the attending physician following standard procedures. The samples were immediately transported to the microbiology laboratory for processing.

Laboratory identification of Microorganisms:

One of the swabs taken was immediately washed in a drop of physiological saline on a clean grease-free glass slide and covered with cover slip and examined under 10X and 40X objectives of the light microscope. *Trichomonas vaginalis* was identified by observing characteristic morphology and jerky motility. The second swab was smeared on a clean grease free slide and fixed in ether-alcohol for 30 minutes. The specimen was then stained by the Papanicolaou (Pap) stain as follows: Harris's haematoxylin without acetic acid for 5 minutes, rinsed in tap water and differentiated in 1% acid alcohol for 30 seconds and blued in Scott's water for 2 minutes, rinsed in 95% alcohol and stained in EA 35 for 2 minutes. Smears were then taken to two changes of absolute alcohol, xylene and mounted in DPX. The stained smears were examined under the light microscope at low and high power objectives for the presence of *Trichomonas vaginalis* and perinuclear halo. The third high vaginal swab was used for *Candida* culture on Chloramphenicol-impregnated Sabouraud's dextrose agar (SDCA) followed by incubation at 37°C for 48 hrs. *Candida* isolates were identified by the standard mycological techniques like cultural characteristics and Gram stain. Species identification of *Candida* isolates were done by following standard mycological protocol including germ tube test, morphology on cornmeal agar, fermentation and assimilation of various sugars and colony color on *Candida* CHROMagar (HiMedia).

IV. Results

A total of 201 symptomatic women of reproductive age (18-45 yrs) were included in the study. Out of 201 selected symptomatic patients 81 (40.29%) participants had confirmed infections with *Candida* spp. and *Trichomonas vaginalis*. Out of 201 selected symptomatic patient, 5 participants (5/201, 2.49%) had *Trichomonas vaginalis* (TV) and 76 participants (76/201, 37.81%) had candidiasis. The prevalence of *Candida* infections exceeded the prevalence of *T. vaginalis* infection. The result is shown in **Table 1**.

Table 1: No of positive and negative cases, and prevalence of candidiasis and *T. vaginalis* infection

Total symptomatic participants	No of confirmed infections n (%)	No of negative cases n (%)
201	81 (40.29%) - <i>Candida</i> 76 (37.81%) - <i>Trichomonas vaginalis</i> 5 (2.49%)	120 (59.70%)

In our study two methods were used to detect Trichomonas infection- wet mount and Pap smear examination. Trichomonas vaginalis was detected in 5 out of 201 participants by using wet mount technique and Pap smear was positive in same 5 cases. The result is shown in the **Table 2**.

Table 2: Detection of Trichomonas vaginalis in vaginal secretion by wet mount and Pap smear technique

Method	Positive n (%)	Negative n (%)	Total
Wet mount	5 (2.49)	196 (97.51)	201
Pap smear	5 (2.49)	196 (97.51)	201

Out of 81 confirmed cases, most of the participants were in the age group of 26-35 yrs (43/81, 53.09%), followed by 36-45 yrs (25/81, 30.86%) and 18-25yrs (13/81, 16.05%). All of them were married and cohabiting with their husbands. Majority of the participants were Muslims (52, 64.20%), followed by Hindu (28, 34.57%) and most of them reside in rural areas (58, 71.60%) and were housewives (48, 53.09%). Majority of the participants had lower or no education. The Socio-demographic characteristics of the study population are shown in **Table 3**.

Table 3: Socio-demographic characteristics of the participants (n=81)

Serial no	Variable	No of participants (%)
1	Age	
	18-25yrs	13 (16.05%)
	26-35yrs	43(53.09%)
2	Religion	
	Hindu	28 (34.57%)
	Muslim	52 (64.20%)
3	Locality	
	Rural	58 (71.60%)
4	Occupation	
	Housewife	43 (53.09%)
	Labour service	17 (20.99%)
	Business others	06 (7.40%)
5	Education	
	Illiterate	13 (16.05%)
	Literate	56 (69.14%)
	(primary- 32/56,57.14%	
	Lower primary-17/56,30.36 %	
6	Monthly household income	
	Low income group (income < Rs 5000/month)	52 (64.20%)
7	Have toilet at home	
	Yes	29 (35.80%)
	No	52 (64.20%)

*marital status: all are married

Associated co-morbid condition and the risk factors were recorded from the requisition proforma on the first visit of the patient and out of 76 candidiasis patients, 12 (15.79%) patients were diabetic, 8 (10.53%) patients were with IUCDs, 5 (6.58%) patients were on oral contraceptive pills. The presenting symptoms of the confirmed cases were vaginal discharge, vaginal malodor, vaginal or vulvar itch, burning sensation in the genitalia, dyspareunia, and lower abdominal pain / discomfort. The predominant clinical signs and symptoms of patients infected with Trichomonas vaginalis and Candida spp. are shown in **Table 4**.

Table 4: Clinical symptoms associated with TV and Candidiasis

Complaints	Trichomonas vaginalis n=5	Candidiasis n=76
Vaginal discharge	2 (40%)	7 (9.21%)
Vaginal/vulvar itch	1 (20%)	22 (28.95%)
Vaginal malodor	2 (40%)	17 (22.37%)
Burning sensation in the genitalia	0	16 (21.05%)
dyspareunia	0	12 (15.79%)

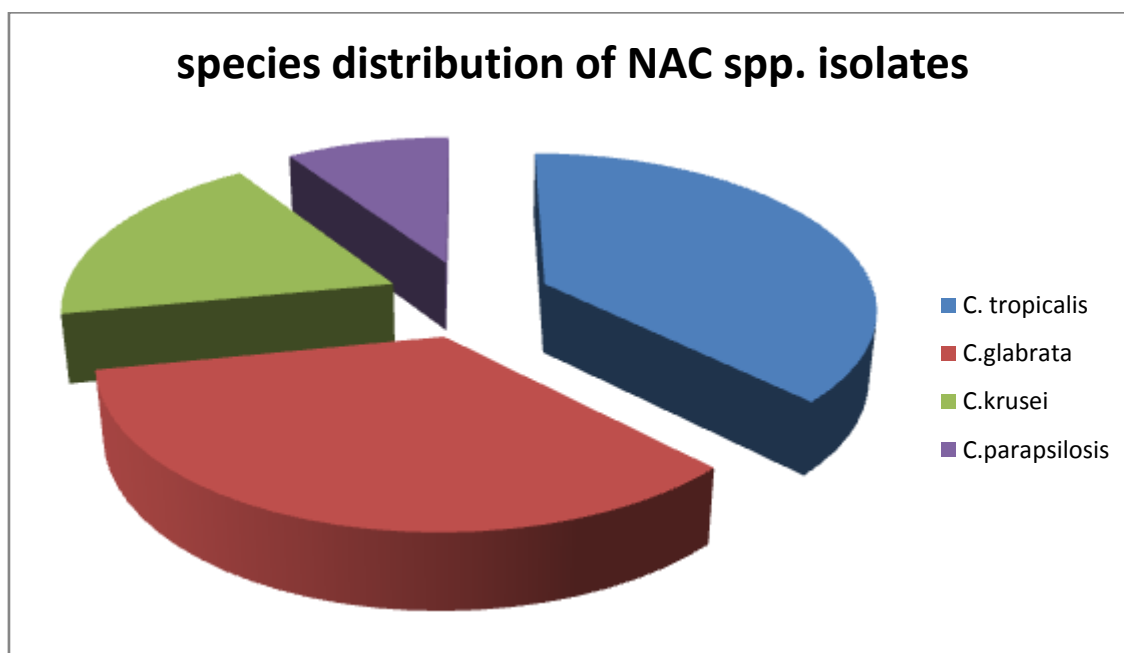
Lower abdominal pain/ discomfort	0	2 (2.63%)
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Out of 81 confirmed infections with *Candida* spp. and *Trichomonas vaginalis*, 5 participants had *Trichomonas vaginalis* and 76 participants had candidiasis. Distribution of *Trichomonas vaginalis* and *Candidiasis* among the participants with respect to their age is shown in **Table 5**.

Table 5: Age wise distribution of vaginal infections

Age group	No of women (n=81)	<i>Trichomonas vaginalis</i> TV (n=5)	vulvovaginal candidiasis VVC (n= 76)
18-25yrs	13	0	13
26-35yrs	43	3	40
36-45yrs	25	2	23

It was found that out of 76 isolated *Candida* species, most (44/76, 57.89%) were *Candida albicans*, whereas others (32/76, 42.11%) belong to NAC spp. *Candida tropicalis* (*C. tropicalis*, 12/32, 37.5%) followed by *C. glabrata* (11/32, 34.38%) were the major NAC spp. isolates, others were *C. krusei* (6/32, 18.75%) and *C. parapsilosis* (3/32, 9.37%). Different NAC spp. isolated from the infected patients are shown in the **Figure** below.



V. Discussion

Vaginitis is common reproductive tract infection in sexually active women and associated with a significant risk of morbidity and adverse reproductive sequelae.

In the present study, out of 201 selected symptomatic patients, candidiasis was detected in 37.81% (76/201) cases. Its prevalence was comparable with earlier reports.^{13, 14, 15} Associated co-morbid condition and the risk factors were recorded from the requisition proforma on the first visit of the patient. In our study, major risk factors identified for vulvovaginal candidiasis was diabetes (10.53%), followed by IUCDs and oral contraceptive pills. Kandati Jithendra et al also reported diabetes as major predisposing factor as in another study by N. Lakshmi et al.^{16, 9}

In our study *C. albicans* (44, 57.89%) was the most common species isolated, followed by Non *albicans candida* spp. (NAC spp., 32, 42.11%) as in other studies.^{17, 18} Though *C. albicans* remained the predominant species isolated in the present study, NAC were isolated in considerable proportions. Among the NAC spp. *C. tropicalis* was the predominant isolate followed by *C. glabrata*. Our observation is in contrast to that of N Twinkle et al where *C. glabrata* was reported as the most common isolate and it was also reported in another study.^{19, 20} But our study result corresponds with a study by Sachin et al at Rural Medical College, Loni Maharashtra.²¹

In our study, a low prevalence (2.49 %, 5/201)) was observed for *Trichomonas vaginalis* which is lower than the prevalence reported by B, Anuradha et al and by others.^{22, 23, 15} But similar results by wet mount technique were observed in studies by Dharma et al, Dickson et al and Olowe OA et al.^{24, 25, 26}

In our study, wet mount and Pap smear techniques were used to isolate TV from clinical samples. Wet mount is cheap and easily available method for detection but it has its own limitations. Though it can be correlated with Pap smear examination to some extent, culture is the gold standard method. Introduction of molecular methods such as PCR is also helpful. In our study negative samples were not confirmed by other specific techniques like culture or molecular methods. So, these variations in prevalence may be related to the difference in the type of techniques used in the isolation of the pathogens, difference in patient's characteristics, socio-demographic profile, and the presence or absence of symptoms in the study population.

All the *Trichomonas vaginalis* cases were found in women aged between 26-45yrs which corresponds with other studies.^{22, 24}

In this recent study not a single case of co-infection with *Trichomonas vaginalis* and candida was found. The occurrence of co-infection with the two etiological agents has been reported by other authors^{27, 28, 29} and varies from 0%²⁶ to 21.73%.³⁰

VI. Conclusion

We can conclude that the rates of prevalence of vulvovaginal candidiasis in the women examined were high and it was higher than the prevalence of *T. vaginalis*. Co-infection with the two etiologic agents was not found in our study. That may be due to the fact that TV is mostly an asymptomatic sexually transmitted infection and conditions for the establishment of TV in the genital tract differ from those required by *Candida* spp. The association between genital infections and certain socioeconomic variables might be useful for the development of risk scoring. Constant health education, sensitization, adequate investigation and treatment will prevent adverse outcome of genital infections.

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Tapati Mondal "Prevalence of Vulvovaginal Candidiasis and Trichomoniasis among Women of Reproductive Age in a Rural Tertiary Care Hospital in West Bengal, India." *IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)*, vol. 17, no. 9, 2018, pp 45-50.