

Microbiological Study of Organisms Causing Abnormal Vaginal Discharge

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ABSTRACT

Vaginal discharge is one of the most common complaints seen in gynecology outpatient departments (GOPD). Alteration in balance of normal vaginal flora can lead to the overgrowth of other organisms that cause abnormal vaginal discharge. One hundred one patients attending GOPD of Nepal Medical College and Teaching Hospital (NMCTH) in between August 2011 to July 2013 with vaginal discharge were evaluated to determine microbiological etiology. Three high vaginal swabs were taken from each patient who visited the outpatient department (OPD) with vaginal discharge. These specimens were subjected to different microbiological evaluations using standard microbiological techniques. Of the total patients studied, the mean (SD) age was found to be 30.15 (6.93) years. Around fifty-seven percent of women had vaginal discharge of infectious etiology. The most prevalent disease was Bacterial Vaginosis (29.7%) followed by Candidiasis (24.75%). Fifty-six percent of the total Candidal infections were caused by *Candida albicans* and the remaining by *Candida glabrata*, *Candida tropicalis*, *Candida parapsilosis* and *Candida krusei*. Trichomoniasis was among the least common (0.99%) cause of vaginal discharge. Two cases were identified as having intermediate vaginal flora. Among the total, 42.57% of females, who gave a history of abnormal vaginal discharge, were found to have normal vaginal flora. Our study concluded that the most common cause of vaginal discharge is bacterial vaginosis followed by candidiasis. Trichomoniasis is less prevalent in our part of the world as compared to the developed countries. Although the syndromic approach for treatment of vaginal discharge is being followed, this study clearly depicted around 43% of those presenting with vaginal discharge had normal vaginal flora. Since simple microbiological tests not only can help to distinguish between normal and pathological vaginal discharge, they can also be used for identifying etiological agents. This will avoid the unnecessary exposure to drugs thus minimizing the economical, social and psychological burden to the lady in question.

Keywords: Bacterial Vaginosis, Candidiasis, Trichomoniasis, Vaginal discharge.

INTRODUCTION

Reproductive tract infections (RTIs) contribute to a large portion of the global burden of ill health.¹ Vaginal discharge is one of the most common presenting complaints seen in gynecology out-patient departments (GOPD). It occurs in 1-14% of all women in the reproductive age group and is responsible for 5-10 million out-patient department (OPD) visits per year throughout the world.^{2,3} It does not only constitute a considerable problem for many women but also causes discomfort and anxiety. Alteration in balance of normal vaginal organisms can cause the overgrowth of the pathogenic organisms that creates abnormal vaginal discharge.

Vaginal discharge is one of most common complaints amongst Asian women.⁴ In India and Nepal, sexually transmitted infections (STIs) remains one of the

commonest diseases next to malaria and pulmonary tuberculosis; and its incidence is increasing in Nepal.⁵ The prevalence of vaginal discharge in India is estimated to be 30%.⁶ However, our part of the world carries a culture of silence; therefore, in most cases, there is delay in seeking help. Some women may ignore pathological discharge whereas some sensitive women may consider normal physiological discharge as abnormal. Abnormal vaginal discharge is predominantly caused by replacement of normal vaginal flora by pathogenic bacteria, *Candida* spp. and parasites like *Trichomonas vaginalis*. Normally, resident vaginal microbial flora is thought to provide protection against infection by a number of different mechanisms. The consequences of abnormal vaginal discharge can be severe and life threatening.¹ In non-pregnant women, the presence of bacterial vaginosis (BV), candidiasis and vaginal trichomoniasis are associated with an increased risk of

upper genital tract infections, STIs and acquisition of human immunodeficiency virus (HIV) during unsafe sex. In pregnancy, abnormal vaginal discharge can increase the risk of postabortal sepsis, early miscarriage, recurrent abortion, late miscarriage, preterm prelabor rupture of membranes (PPROM), spontaneous preterm labor (SPTL) and preterm birth (PTB), histological chorioamnionitis, and postpartum endometritis. As a result, abnormal vaginal flora may predispose women to ascending colonisation of the genital tract, infiltration of the fetal membranes, microbial invasion of the amniotic cavity and fetal damage. Preterm birth due to infectious etiology is associated with high perinatal mortality and morbidity, and a high cost to the healthcare system.⁷ Due to the above mentioned complications, early detection of vaginal discharge and identifying normal vaginal secretion from an abnormal one plays an important role in patient management and limits the cost of diagnostic shopping, side-effects of inadequately treated disease, and unnecessary anxiety.⁸ The challenge ahead is to create a system that can accommodate new needs of the community and adapt modern concepts of health care in regards to genital infections. With this consideration, this study aimed to evaluate the clinical and microbiological profiles of women presenting at the GOPD of Nepal Medical College and Teaching Hospital with abnormal vaginal discharge. Genital infection in women is usually caused by vulvovaginitis and its main etiologies are considered to be bacterial vaginosis, vaginal candidiasis and trichomoniasis.^{9,10} Hence, these three most common causes of abnormal vaginal discharge of infectious origin are considered for this study.

MATERIALS AND METHOD

Operational definitions

1. Bacterial vaginosis: A gram stain score of seven or more based on the Nugent scoring system,¹¹ or fitting 3 of 4 diagnostic criteria of Amsel's criteria.¹²
2. Candidiasis: If budding yeast or pseudohyphae were seen on wet mount or gram stain, or if growth of organism in SDA is proven to be *Candida* species.
3. Trichomoniasis: If wet vaginal smear microscopy shows motile *Trichomonas vaginalis*

This study was carried out at Nepal Medical College Teaching Hospital (NMCTH), Kathmandu, Nepal from August 2011 to July 2013. After clinical, bimanual and per speculum examination, three vaginal swabs were collected from the posterior fornix. The pH of the vaginal secretion was checked from the fluids that coated the speculum. The first swab was used to prepare the wet mount to look for the presence of motile *Trichomonas* species, yeast cells and clue cells. Observation of pus

cells were also noted, if any. The second swab was used to prepare Gram's stain and to perform Whiff's test to look for different morphotypes of bacteria (Nugent scoring), clue cells, yeast cells, pus cells and amine odour. The third swab was used to inoculate two tubes containing Sabouraud Dextrose Agar (SDA) slant and a plate of chocolate agar. The diagnosis of BV was made by evaluation of patients clinically, scoring of gram stained slide of vaginal secretions using Nugent score, fitting 3 of 4 diagnostic criteria of Amsel's criteria and comparing the growth of lactobacilli and *Gardenerella vaginalis* by semi-quantitative culture of vaginal secretions in chocolate agar.

Fungal culture tubes for the isolation of *Candida* spp. were examined on days 1, 3, 5 and 7. Identification and speciation of different *Candida* spp. was done by using germ tube test, formation of chlamydo spores on cornmeal agar, sugar assimilation test, growth at 42°C and urease test.

RESULT

A total of 101 samples from patients who attended GOPD with complaints of vaginal discharge were included in the study. All patients were married and sexually active. The mean (SD) age of the patients was found to be 30.16 (6.93) years. Abnormal vaginal discharge was found in 58 patients (57.43%). The most common cause of abnormal vaginal discharge was found to be BV (29.7%) and Candidiasis (24.75%). Intermediate vaginal flora was found in 1.98% cases and Trichomoniasis was identified in 0.99%. However, 42.57% of women who presented in the OPD with complaints of discharge were found to have normal vaginal flora. (Figure 1).

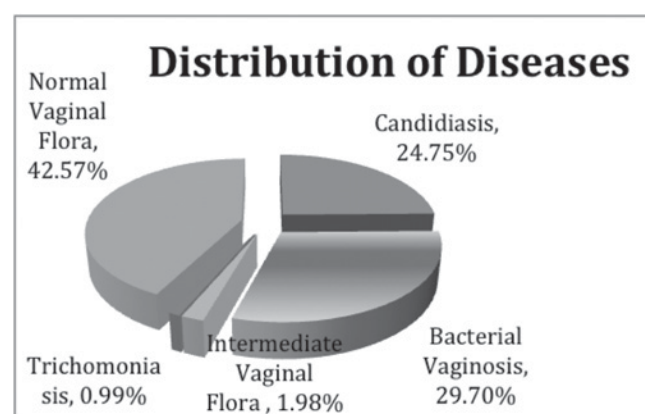


Fig 1: Distribution of Disease according to etiology

Similarly, prevalence of abnormal vaginal discharge was found to be almost equally distributed among age groups 18-24 and 25-29 whereas lowest prevalence was observed among 40 years and above. BV and Candidiasis was common among the age group of 18-24 and 25-29 respectively. (Table 1)

Table 1: Distribution of etiology in relation to age.

Age	Candidiasis	Bacterial Vaginosis	Intermediate Vaginal flora	Trichomoniasis	Total
18-24	6	10	0	1	17
25-29	9	7	1	0	17
30-34	4	5	1	0	10
35-39	4	4	0	0	8
40 and above	2	4	0	0	6

Among the 101 vaginal swabs taken, 28(27.72%) gave a positive Nugent’s score; in contrast, Amsel’s criteria diagnosed 26(25.74%) as suffering from BV (Table 2).

Table 2: Contingency table (2 x 2) comparing the results of Amsel’s criteria with the Nugent’s score

		Amsel criteria		
		BV positive	BV negative	
Nugent Score	Bacterial vaginosis	24	4	28
	Non-Bacterial vaginosis	2	71	73
	Total	26	75	101

Two cases were positive with Amsel’s criteria, which were not consistent with BV by Nugent score. At the same time, four cases were diagnosed as BV using Nugent score which didn’t fit the Amsel’s criteria. Similarly, a total of 25 cases (24.75%) were diagnosed as vulvo-vaginal candidiasis. Figure 2. shows the different species of candida isolated. *Candida albicans* was found to be the most common, isolated from 14 (56%) cases. The remaining 11 were candida species other than *Candida albicans* and were identified as *C. glabrata* 3 (12%), *C. tropicalis* 3 (12%), *C. parapsilosis* 2 (8%) and *C. krusei* 1 (4%).

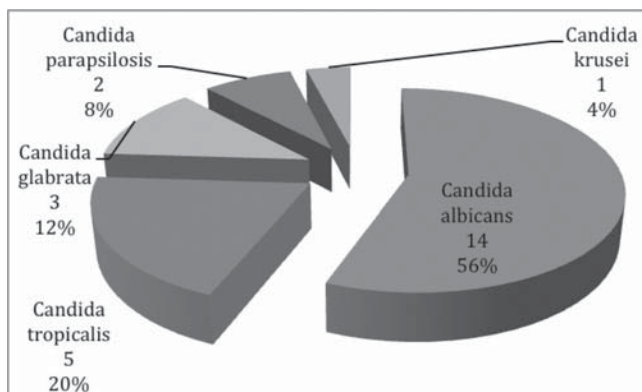


Fig 2: Distribution of Candida species responsible for causing Vaginal Candidiasis.

A single case of trichomoniasis, observed as containing motile parasites during wet-mount examination, was found among the total patients. Two of the cases which showed intermediate vaginal flora were excluded from the diagnosis of BV as they neither fitted Amsel’s criteria nor had a Nugent score of more than seven.

DISCUSSION

This study was done with an objective to determine the prevalence of vaginitis among women of reproductive age group and has revealed a total of 58 (57.42%) positive cases. However, 43 (42.57%) patients complained of abnormal vaginal discharge but were not diagnosed as vaginosis according to clinical and microbiological criteria. In a study done by Shrestha S *et al* (2006) among patients visiting Paropakar Maternity and Women’s Hospital in Kathmandu, prevalence of vaginitis was found to be 39% among pregnant women.¹³ This prevalence rate is lower than that found in our study. These noticeable differences might be due to different selection criteria of the patients. High prevalence in the current study might also be due to use of more than one method for the diagnosis of BV and candidiasis. Even though the patient gives a history of abnormal vaginal discharge, true infective cause of vaginal discharge may not always be present in them. Our study has shown that there is presence of single etiology causing abnormal vaginal discharge for each case. Hence, the WHO approach of syndromic management may be over treating the patient.

Vaginal infections were found to be more common in women of age group 25-30 years. Similar findings were reported in different studies done by S. Rekha *et al* and F. Sarwat in India.^{14,15} White colored vaginal discharge was the most common complaint in this study, which was narrated by 79 patients. Majority of patients gave the history of thick vaginal discharge. BV was the most common cause of vaginal discharge which accounted for 29.7 % (n=30) of total cases followed by vulvo-

vaginal candidiasis, 24.75% (n=25) and trichomoniasis 0.99% (n=1). Bhalla P *et al*, from India (2002) had also reported similar findings where prevalence of BV was highest accounting for 32.8% of patients, followed by candidiasis (16.9%) and (2.8 %) trichomoniasis.¹⁶ This similar finding may be due to cultural similarity among women of India and Nepal. Similar results were found in a study conducted (in 1998) in a hospital in Saudi Arabia. In a similar study conducted by Joharah M among 2719 females attending the PCC over a 6-month period, 175 (6.4%) complained of vaginal discharge and only 99(56.6%) of them had vaginal discharge of infectious origin. The type of organisms isolated in the study were *G. vaginalis* 49 (28%), *C. albicans* 21 (12%) and *T. vaginalis* 7 (4%).¹⁷ Moreover, BV was diagnosed using clinical and Amsel's criteria and also with the Nugent scoring system. Hence, prevalence of BV may have been high due to more than one diagnostic tool used. In our study, out of 30 cases diagnosed as BV, 28 cases gave Nugent's score consistent with BV, in contrast, Amsel criteria diagnosed 26 (25.74%) as suffering from BV (Table 2). From the total, 2 cases were positive with Amsel's criteria which were not consistent with BV by Nugent score. At the same time, 4 cases were diagnosed as BV using Nugent score which did not fit the Amsel's criteria. Similar result was observed by Modak *et al* (2010) in a study conducted at Kolkata.¹⁸ In their study, using Nugent score and Amsel criteria, the prevalence of BV was 24% and 20% respectively. Similarly, Demba E *et al* (2005), found the prevalence rate of BV as 47.6% by Nugent's score and 30.8% by Amsel's clinical criteria.¹⁹ Whereas, in another study at Siriraj Hospital (2003) conducted by Chaijareenont K *et al*, BV was diagnosed by Amsel's criteria in 32 (14.7%) and by Nugent's score in 26 (12%) of 217 women.²⁰

Our study shows vulvo-vaginal candidiasis as the second most common cause of vaginal discharge accounting for 25 (24.75%). Similar prevalence rate of vaginal candidiasis (29.5%) was observed in a study conducted by Shrestha S *et al* (2006) at Paropakar Maternity and Women's Hospital, Kathmandu.¹³ Of the 25 patients who had candidiasis, *C. albicans* was the most common pathological organism (56% of the total cases) followed by *C. tropicalis* (12%), *C. glabrata* (12%), *C. parapsilosis* (8%) and *C. krusei* (4%). This was not unexpected because over the past three decades, an increasing trend in the number of vaginal infections attributable to yeasts other than *Candida albicans* has emerged.²¹ High incidence of *C. albicans* as a causative agent of vulvo-vaginal candidiasis was also seen in a study done by Nwadioha S.I *et al*.²² Similarly, in another study done by Jindal N *et al*, among 350 women of age group 16-45 years, positive culture for *Candida* species

was obtained in 82 (23.4%) women. Of these, 61 (74.4%) were *C. albicans* and 21 (25.6%) were non *C. albicans* (*C. glabrata*- 11%, *C. tropicalis*- 6%, *C. krusei*- 3.6% and *C. parapsilosis*- 2.43%).²³ Candidiasis was more common among the age group of 25-29 years and most of the patients gave the history of curdy white heavy vaginal discharge. Twenty patients (80%) who were diagnosed to have vulvo-vaginal candidiasis gave the association of itching with the condition. Rylander E *et al*²⁴ also found that 85% of patients with vulvo-vaginal candidiasis complained concurrent history of itching in the vulva. Our finding is more or less similar to their findings. Intermediate vaginal flora was identified in 2 (1.98%) patients which signifies the need of further evaluation for the possibility of developing BV or other STIs. Sewankambo *et al* reported BV in 50.2 % and intermediate flora in 31.7 % women.²⁵ Likewise, only one case of *T. vaginalis* was diagnosed in this study. Similar prevalence rates were seen in other studies.^{26,27} In Islamic countries, the prevalence of trichomoniasis ranges from 1.2% in Libya and Jordan to 3.2% in Turkey to 28.1% in Saudi Arabia.²⁸⁻³¹ The difference in the results may be due to the selection of different population groups. In conclusion, along with clinical examination, microbiological diagnostic approach for the etiological diagnosis of symptomatic vaginal discharge plays an important role in the treatment of patient. The treatment plan can be formulated if simple diagnostic tool that can be implemented in the rapid and accurate diagnosis of symptomatic vaginal discharge is taken under consideration.

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