Original Article

Epidemiology and clinico-investigative study of organisms causing vaginal discharge

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Abstract

Background: Abnormal vaginal discharge is a common clinical problem in reproductive age group. It is the second most common problem after abnormal uterine bleeding. It is a neglected health problem, most commonly caused due to vulvovaginal candidiasis, trichomoniasis, and bacterial vaginosis (BV). **Objectives:** The present study was conducted to determine the prevalence of common organisms causing vaginal discharge and also to know the variety of clinical presentation. **Materials and Methods:** A cross-sectional descriptive study was conducted in the Skin and STD Outpatient Department of Vinayaka Mission Kirupananda Variyar Medical College Hospital, Salem, who presented with abnormal vaginal discharge between September 2012 and September 2014. A total of 100 women in the reproductive age group who had symptoms of vaginitis were examined. Data were coded and analyzed. **Results:** Out of the 100 patients examined, 77 (77%) cases were organism positive. Among the positive cases, BV (27%) was the most common microbiological cause of abnormal vaginal discharge, followed by trichomoniasis (25%), vaginal candidiasis (22%), combined infection (*Candida* and BV) (3%), and nonspecific cases (23%). **Conclusion:** Out of 100 cases, few cases showed discordance between clinical and laboratory diagnosis. This discordance can be due to pitfalls in identifying the causative agent clinically or obscuring of the findings due to improper treatment received for other ailments. Thus, clinico-investigative correlation is more important than other clinical findings alone.

Key words: Bacterial vaginosis, candidiasis, nonspecific vaginitis, trichomoniasis

INTRODUCTION

Abnormal vaginal discharge is a common clinical problem among women of reproductive age group with multiple etiologies. It is the second most common problem after menstrual disorders.[1] One in ten women will present with vaginal discharge in the course of a year.[2] Approximately, ten million office visits each year are attributed to vaginal discharge complaints.[3] Many women with vaginal complaints self-treat incorrectly with over-the-counter drugs.[4] Health-care providers themselves may miss the correct diagnosis if they fail to confirm the diagnosis with the proper laboratory test.[5]

The vagina, ectocervix, and endocervix are all susceptible to various pathogens, depending on type of epithelium and other factors in the microenvironment.[6] The stratified squamous epithelium of the vagina and ectocervix is susceptible to infection with *Candida* species and *Trichomonas vaginalis* (TV).[6] The columnar epithelium of endocervix is susceptible to *Neisseria*

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gonorrhoeae and Chlamydia trachomatis.[6] Herpes simplex virus may infect both types of epithelium.[4] Infection caused by any of these organisms can lead to vaginal discharge. Identifying its specific cause can be a challenging task because a large number of pathogens cause vaginal and cervical infections, and several infections may coexist.[6]

Abnormal vaginal discharge is usually related to one of the three conditions, such as bacterial vaginosis (BV), vulvovaginal candidiasis (VC), and trichomoniasis.[3] Apart from the above-mentioned reasons, we should also consider cytolytic vaginosis (lactobacillus overgrowth syndrome) also a cause for abnormal vaginal discharge which is characterized by abundant growth of *Lactobacilli* resulting in lysis of vaginal epithelial cells; and therefore, it is called as cytolytic vaginosis.[7] The signs and symptoms were similar to VC characterized by pruritus, dyspareunia, and vulvar dysuria.

BV, which is primarily characterized by a malodorous discharge, is common in women with multiple sex partners and is caused by the overgrowth of several facultative and anaerobic bacterial species.[6] VC is characterized by pruritus and a curd-like discharge. Vaginal trichomoniasis is associated with a copious yellow or green, sometimes frothy, discharge.[5] The present study is carried out to find the characteristics of vaginal discharge and the prevalence of various pathogenic agents causing vaginal discharge.

Objectives

This study of epidemiology and clinico-investigative study of patients presenting with vaginal discharge was undertaken:

1. To determine the prevalence of common organisms causing vaginal discharge with special reference to three organisms mainly Trichomonas, *Candida* species, and *Gardnerella*.
2. To know the various types presentation in those sexually active women presenting with vaginal discharge.

MATERIALS AND METHODS

The material for this study was from the patients attending the Skin and STD Outpatient Department of Vinayaka Missions Kirupananda Variyar Medical College, Salem, with abnormal vaginal discharge during the period of September 2012–September 2014. This was a cross-sectional descriptive study. All the patients clinically having the symptoms of vaginitis and with discharge were included in this study. Postmenopausal women and pregnant women were excluded from the study.

Method of collection

A total of 100 women of reproductive age group with the complaints of vaginal discharge were selected for this study at random after applying the criteria mentioned above. With the prior consent, a comprehensive history, general examination, and gynecological examination were initially carried out. After making a clinical diagnosis, a sterilized Cusco's speculum was inserted into the vagina to visualize the vagina and cervix. Any pathology of vagina and cervix such as vaginitis, discharge, cervicitis, and cervical erosions was looked for. The amount, color, character, and smell of the vaginal discharge were noted. The discharge was then collected by three sterile swabs from the upper part of posterior fornix.

The pH was measured using indication papers ranging from 2 to 10 by directly dipping pH strip in vagina. Color change was observed and matched against the indicator. One swab was used for making wet mount to look for the motility of TV. The second swab was used for making smears for Gram staining to find out clue cells, Gonococci. The third swab was used to do potassium hydroxide (KOH) mount to look for *Candida*.

Laboratory procedures

For *Candida*:

1. KOH preparation: a drop of 10% KOH was added to the vaginal secretions taken on a clean glass slide and mounted with a coverslip. *Candida* was identified as round or oval budding yeast cells
2. Gram-stained vaginal smears were examined which showed Gram-positive budding yeast cells with pseudohyphae
3. Cultures on Sabouraud's dextrose agar (SDA) medium showed a growth of creamy, grayish moist colonies

For TV: wet mount preparation was immediately made - a drop of discharge was mixed with a drop of normal saline on a clean slide and topped with a coverslip. This was then examined under microscope for flagellate organisms with characteristic motility.

For BV:

1. Whiff amine test was done as follows - one or two drops of vaginal discharge were taken on a clean glass slide, and one or two drops of freshly prepared

10% KOH solution were added to it. Both were mixed and smelt immediately

2. Wet film was examined for the presence of clue cells which are vaginal epithelial cells with a granular surface and blurred margins because of attached bacteria
3. Gram-stained smears were examined for the presence of altered vaginal flora in form of Gram-negative coccobacilli studding vaginal epithelial cells instead of normally predominant Gram-positive *Lactobacilli*.

For *Neisseria Gonorrhoeae*: Gram-stained smears were examined for the presence of intracellular Gram-negative diplococci. Data collection sheet included patient's demographic information, clinical features, examination findings, and laboratory investigation. Data were analyzed using appropriate statistical methods.

OBSERVATIONS AND RESULTS

In this study of 100 cases who presented with abnormal vaginal discharge, the following observations were made. Table 1 shows the demographic characteristics of patients with vaginal discharge. The patient's ages ranged from 18 to 50 years. Fifty percent of patients had completed at least elementary and intermediate education, and 60% of patients were employed.

Clinical diagnosis

Of the 100 patients who presented with abnormal vaginal discharge based on the signs and symptoms, a clinical diagnosis of nonspecific vaginitis was made in 60 (40%) patients, BV in 30 (20%) patients, VC in 25 (16.7%) patients, and trichomoniasis in 13 (8.7%) patients. Table 2 shows the clinical diagnosis.

Different microbiological etiologies of abnormal vaginal discharge among the cases

Table 3 shows the different microbiological etiologies. Of the 100 patients, 77 (77%) cases were organism positive. Among the positive cases, BV (27%) was the most common microbiological cause of abnormal vaginal discharge, followed by trichomoniasis (25%), vaginal candidiasis (22%), combined infection (*Candida* and BV) (3%), and nonspecific other urogenital cases (23%).

Demographic characteristics of patients by type of organism

Table 4 shows the association between demographic characteristics and the type of organism isolated. The peak age group for vaginal infections was 26–35 years (44%). Most of the specific vaginal infections occurred in this age group. Educational level does not influence the infection rate.

Prevalence of pathogens causing vaginal discharge in different age groups

BV was found in high frequency in the age group of 26–35 years (48%) followed

Table 1: Demographic Characteristics Of Patients Complaining Of Vaginal Discharge

Characteristics	Percent (%)
Age:	
18-25	16
26-35	34
36-40	26
40-50	21
Educational Level:	
Illiterate	50
Literate	50
Occupation:	
Employed	60
Unemployed	40

Table 2: Clinical Diagnosis of patients presenting with vaginal discharge

Clinical diagnosis	No. of Cases	Percent (%)
Non Specific Vaginitis	60	40
Bacterial vaginosis	30	20
Candidiasis	25	16.7
Trichomoniasis	13	8.7

Table 3: Aetiology of Abnormal vaginal discharge

Aetiology Of Abnormal Vaginal Discharge	Number (*n*=100)	Percent (%)
Bacterial vaginosis	27	27
Trichomoniasis	25	25
Vaginal candidiasis	22	22
Combined infection (Candida and Bacterial vaginosis)	3	3
Organism not found	23	23
Total	100	100

Table 4: Demographic Characteristics of Patients by type of Organism

Characteristics	Frequency (%)	BV	VC	TV	Combined (BV+VC)
Age:					
18-25	19	4	6	3	-
26-35	44	13	6	7	2
36-40	26	4	5	7	-
40-50	24	6	5	8	1
Educational Level:					
Illiterate	50	15	10	10	2
Literate	50	12	12	15	1
Occupation:					
Employed	60	17	12	14	1
Unemployed	40	10	10	11	2

by 40–50 years (22%), then by 36–40 and 18–25 years (15%). The most common age group affected by vaginal candidiasis was 26–35 and 18–25 years (27%) followed by 40–50 and 36–40 years (23%). Trichomoniasis was detected in the highest rate at the age group of 40–50 years (32%) followed by 36–40 and 26–35 years (28%), then by 18–25 years (12%).

Nonspecific vaginitis where no organism was found seen in high frequency in the age group of 36–40 years (44%) followed by 26–35 years (35%). Mixed infection of *Candida* and BV was seen in three patients mainly in the age group 26–35 years (67%) followed by 40–50 years (33%). Table 5 shows the prevalence of pathogens causing vaginal discharge.

Characteristics of vaginal discharge according to pathogenic agents

Characteristics of vaginal discharge of patients differed according to pathogenic agents. The pH of vaginal secretions was >5 in a large proportion of examined cases which included women with BV and TV, but women with *Candida* infection, the pH was slightly below the normal value. Whiff amine test was positive in 27 cases, most of them having BV.

Clinical signs and symptoms in relation to occurrence of vaginal infection

Clinical signs and symptoms were examined in relation to the infection. Out of the 100 patients, 30 patients had homogeneous white discharge suggestive of BV [Figure 1], 22 patients had curdy white discharge suggestive of candidiasis [Figure 2], 25 had frothy white discharge suggestive of TV [Figure 3], and 23 had mucopurulent discharge. Pruritus was present in 60% of cases, lower abdominal pain in 45% of cases, and menstrual disturbances in 40% of cases.

Nugent scoring

In all the 100 cases, Gram's staining was done for Nugent scoring. Nugent scoring is considered as the gold standard test for diagnosis of BV. It has a scoring system of 0–10 and allows for intermediates between normal and abnormal. The criteria for BV are a total score of 7–10, while a score of 0–3 is normal, and a score of 4–6 is intermediate. In our study, 30% cases had a score of 7–10 who were diagnosed to have BV, 24% of cases had a score of 4–6, and 46% of the cases had a score of 0–3.

Table 5: Prevalence of Pathogens causing vaginal discharge in different age groups

Pathogens	No	18-25		26-35		36-40		40-50	
		No	%	No	%	No	%	No	%
Bacterial Vaginosis	27	4	15	13	48	4	15	6	22
Vaginal Candidiasis	22	6	27	6	27	5	23	5	23
Trichomoniasis	25	3	12	7	28	7	28	8	32
Combined (Candida and Bacterial vaginosis)	3	-	-	2	67	-	-	1	33
No organism	23	3	13	8	35	10	44	2	9

DISCUSSION

A total of 100 cases who presented with abnormal vaginal discharge were examined during the period of study. Vaginal discharge is a common health problem among women in the reproductive age group. Whether asymptomatic or symptomatic, it is usually neglected by women making the diagnosis more difficult.

The incidence of pathogens in vaginal discharge varies in different regions of the world (Kumar, 1994). Vaginal discharge may be either physiological or pathological in origin. It is difficult to know what proportion of discharges belong to either category. Although many cases of abnormal vaginal discharge are not caused by sexually transmitted infections, common curable sexually transmitted infections can present with this symptom.

Demographic characteristics of patients complaining of vaginal discharge

Among the 100 cases with abnormal vaginal discharge, majority of the patients were in the age group 26–35 years (34%) because they belong to the sexually active age group. However, this was not statistically significant. There was also no significant association with both literacy level and employment status in contrast to Al Quaiz[8] study where the infection rates were highest among secondary school and university graduates. This may be attributed to the smaller sample size of ours when compared to Al Quaiz[8] study.

The majority of BV (56%) occurred in illiterates which in contrast to Al Quaiz[8] study where BV was common among educated people because educated patients are more likely to be informed about physiological and pathological vaginal discharge and thus more likely to seek health services. Most of the vaginal candidiasis (55%) cases occurred in literates which are similar to Al Quaiz[8] study, and about 60% of TV infections were isolated from the literate group.

Figure 1: Homogeneous white discharge suggestive of bacterial vaginosis

Figure 2: Curdy white discharge suggestive of candidiasis

Figure 3: Frothy white discharge suggestive of *Trichomonas vaginalis*

Different microbiological etiologies of abnormal vaginal discharge among the cases

In this study, organisms responsible for abnormal vaginal discharge were found in 77% of the cases. Among the positive cases, BV (27%) was the most common microbiological cause of abnormal vaginal discharge, followed by trichomoniasis (25%), vaginal candidiasis (22%), combined infection (*Candida* and BV) (3%), and nonspecific other urogenital causes (23%).

Bacterial vaginosis

BV (27%) was the most common microbiological cause of abnormal vaginal discharge in our study. Figure 4 shows the clue cells. This is comparable to the study of Koumans *et al*.[9] who had also found a 29.2% prevalence of BV. In Pawanarkar and Chopra study,[10] BV was the most common cause of genital tract infections as it was prevalent in 19% of women similar to our study. In a study by Gupta *et al*.,[11] 2005 of 139 women reported that BV was the most common cause of genital tract infections as it was found in 44.6% of women which is little bit higher than our study. Nessa *et al*.[12] in Bangladesh reported 48.1% cases of BV among the sex workers which is also on the higher side. This high prevalence may be the result of disturbance of vaginal microflora resulting from frequent sexual intercourse and the subsequent frequent washing with water and disinfectant. The variations in the prevalence could be related to geographical distribution or systematic differences in the various population samples; however, there is continuing controversy about its importance as a pathogen and its ability to cause vaginitis.

BV was found in high frequency in the age group of 26–35 years (48%) followed by 40–50 years (22%), then by 36–40 and 18–25 years (15%). The peak age of BV in this study was 26–35 years, similar to the findings of Chowdhury *et al*.,[13] but there was no statistically significant association found between age and infection such as Al Quaiz,[8] Madhivanan *et al*.,[14] and Bukusi *et al*.[15] also reported that BV is more common in younger age group. The reason is mainly due to the increased sexual activity which causes disruption of normal vaginal flora.

Trichomoniasis

Trichomoniasis (25%) was the second most common microbiological etiology of abnormal vaginal discharge in our study. It was detected in the highest rate at the age group of 40–50 years (32%) followed by 36–40 and 26–35 years (28%), then by 18–25 years (12%). The prevalence of trichomoniasis in our study was comparable to Bachmann *et al*.'s study,[16] whereas Lally *et al*.[17] showed a prevalence of 43% among women at a substance abuse center. This high prevalence may be due to the high-risk nature of the group that was studied.

Vaginal candidiasis

Vaginal candidiasis (22%) was the third most common microbiological etiology of abnormal vaginal discharge in our study. Figures 5 and 6 illustrate the KOH mount which shows budding yeast cells, pseudohyphae, and culture on SDA medium showing the growth of *Candida* as creamy white colonies respectively. Although the vaginal candidiasis is the highest diagnosed disease, the reason for low incidence in this study may be due to geographical variation. The most common age group affected by vaginal candidiasis was the 26–35 and 18–25 years (27%) followed by 40–50 and 36–40 years (23%). Nwadioha *et al*.[18] and Verbalis *et al*.[19] also reported a similar result of increased prevalence in younger age group because of increased sexual activity in this age group. Candidiasis is not usually a sexually transmitted disease; however, male contacts could be possibly involved.

Combined infection

The combination of two pathogens has been investigated in the present study. In our study, 3% of cases presented with combined infection (BV + vaginal candidiasis).

Nonspecific vaginitis

In 23 specimens out of the 100 cases, there were no pathogenic organisms isolated in spite of the infection in the speculum examination. These cases might have been caused by Chlamydia, *Mycoplasma*, or viral agents for which methods of isolation were not available in the present study. In short, abnormal vaginal discharge was most prevalent in the age groups 23–33 years. In the present work, the highest prevalence of sexually transmitted infection (44%) has been found in the age group of 26–35 years. This is in agreement with Saxena and Yadaf[20] (2001) that attributed to the higher sexual activities in this age group.

Characteristics of vaginal discharge according to pathogenic agents

Characteristics of vaginal discharge of patients differed according to pathogenic agents and sometimes its properties give impression about the causative agents that would be isolated and identified. The pH of vaginal secretions was >5 in a large proportion of examined cases which included women with BV (27%) and TV (25%), but women with *Candida* infection (22%), the pH was slightly below the normal value. These results are in agreement with Caillouette *et al*.,[21] who demonstrated that pH value in aerobic bacterial infection is higher than that obtained from patients with either normal flora or yeast infection. In the present study, positive Whiff test with KOH (amine test) was positive in 27 cases, most of them having BV. We chose Nugent scoring as the gold standard test for diagnosis of BV because the Nugent scoring system is an excellent method for

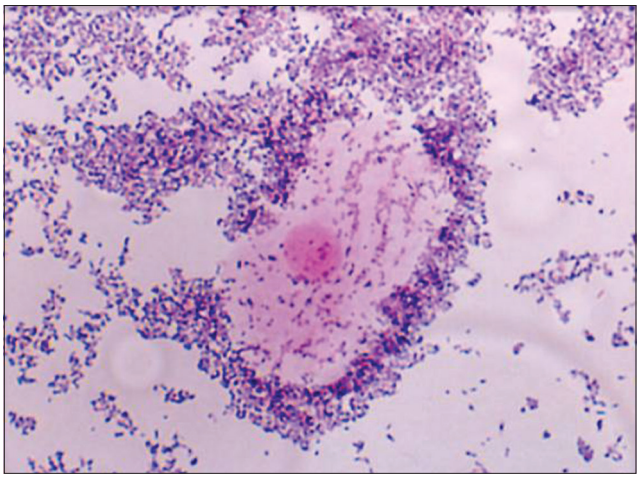

Figure 4: Photomicrograph showing clue cells

Figure 5: Photomicrograph of potassium hydroxide mount showing pseudohyphae and budding yeast cells

Figure 6: Sabouraud's dextrose agar medium showing creamy white colonies suggestive of *Candida*

laboratory evaluation of cases of BV, and it is more reliable than Amsel criteria.

CONCLUSION

The present study was done on 100 patients with signs and symptoms of vaginitis. The causative agents of BV, trichomoniasis, and vaginal candidiasis are the most commonly found infectious agents in abnormal vaginal discharge among sexually active women. BV is by far the most common cause of abnormal vaginal discharge in our study followed by trichomoniasis with vaginal candidiasis was the third most common cause for vaginal discharge. The highest prevalence of sexually transmitted infection (44%) has been found in the age group of 26–35 years which is attributed to the higher sexual activities in this age group. Educational level and employment status do not have significance influence on infection rate in our study. This may be attributed to the smaller sample size of our study.

Out of 100 cases, few cases showed discordance between clinical and laboratory diagnosis. This discordance can be due to pitfalls in identifying the causative agent clinically or obscuration of the findings due to improper treatment received for other ailments. Thus, the clinico-investigative correlation is more important than considering only the clinical findings. Clinicians need to be aware of emerging epidemiological data, the different presentations of vaginal discharge, and the approach of their management so that the symptom can be treated according to its etiology.

It is recommended that prevention, early diagnosis, and prompt treatment of abnormal vaginal discharge especially among the sexually active women should be done to avoid the complications and reduce HIV transmission. There is a need for creating community awareness about health-care facilities and self-concern in women for their own health needs. Hence, this study was done to emphasize the role of laboratory investigations in patients of vaginitis as clinical diagnosis alone can lead to false interpretation.

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Conflicts of interest

There are no conflicts of interest.

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