A STUDY OF PREVALENCE OF BACTERIAL VAGINOSIS IN SEXUALLY ACTIVE FEMALES- A CROSS-SECTIONAL STUDY IN TERTIARY CARE HOSPITAL, GAYA
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ABSTRACT

BACKGROUND
Vaginal discharge is very common problem among females. Alteration in balance of normal vaginal organisms can cause the overgrowth of the bacteria that creates vaginal discharge. It is common among sexually active women yet there still remain gaps in our knowledge of this infectious disorder. Bacterial Vaginosis (BV) also called no-specific vaginitis develops when the normally predominant peroxide producing lactobacillus species in the vagina are replaced by mixed predominantly anaerobic flora consisting of Gardnerella vaginalis, Mycoplasma hominis, Mobiluncus species, Bacteroides species, Prevotella species, Peptostreptococcus species, Fusobacterium species and Porphyromonas species. The present study was conducted to know the prevalence of bacterial vaginosis in sexually active females coming with the chief complaint of vaginal discharge to Obstetrics and Gynaecological OPD Bihar.

MATERIALS AND METHODS
A total of 200 otherwise healthy women of reproductive age group with the complaint of excessive vaginal discharge were included in the study. We exclude the patients who are married, women with known skin disease and post-menopausal. Normal saline wet-mount slide preparations were made for detection of motile trichomonads. Gram stained smear were prepared and scored as per classification developed by Nugent. The presence of pseudohyphae and/or budding yeast cells was considered diagnostic of candidal infection.

RESULTS
The median age of the study population was 28 years. Most common cause was bacterial vaginosis (positive= 18.5 %; intermediate score= 19.5 %), followed by candidiasis (13.5%) and trichomoniasis (0.5%). No etiological diagnosis for vaginal discharge could be established in approximately half of the women. Only one woman was HIV positive; one was reactive by VDRL and TPHA tests.

CONCLUSION
In addition, the laboratory services network needs to be strengthened to ensure accurate and standardized availability of diagnostic services. The study showed higher prevalence of Bacterial Vaginosis. There was significant correlation between vaginal pH, IUCD user, history of STD, RTI, VDRL and HIV positive patients.

KEYWORDS
Bacterial Vaginosis, Candidiasis, Trichomoniasis, Vaginal Discharge, Sexually Active Females, Intrauterine contraceptive Device, STD.


BACKGROUND
Bacterial Vaginosis (BV) also called no-specific vaginitis develops when the normally predominant peroxide producing lactobacillus species in the vagina are replaced by mixed predominantly anaerobic flora consisting of Gardnerella vaginalis, Mycoplasma hominis, Mobiluncus species, Bacteroides species, Prevotella species, Peptostreptococcus species, Fusobacterium species and Porphyromonas species.¹

The problem of vaginal discharge is probably the most frequently narrated complaint of woman of reproductive age group.¹,² Vaginal discharge constitute a considerable problem for many women causing discomfort, anxiety affecting women's quality of life and consuming considerable resources. Some vaginal discharges are normal and can vary...
with age, use of contraceptives, menstrual cycle and with the oestrogen level.\textsuperscript{4,5}

The vaginal flora is a dynamic ecosystem that can be easily altered. The most frequently encountered causes of vaginal discharge. Although there are four causes of vaginal discharges which cover almost 95% of cases. These are bacterial vaginosis, candidal vulvovaginitis, Trichomoniasis and normal physiological discharge.

The management of vaginal discharge is largely syndromic and empirical, it is usually based on naked eye examination of vaginal discharge and that is unsatisfactory because the diagnostic accuracy is lost without microscopic examination.\textsuperscript{6} The modern management of vaginal discharge demands a specific diagnosis which is a combination of naked eye examination plus laboratory work up. Most of the times laboratory assistance in patients of vaginal discharge is sought only after therapeutic failure of repeated courses of empirical therapy. It not only has a financial and social impact leading to noncompliance on the part of patients, but also contributes to overall emergence of resistance.\textsuperscript{7}

A common belief is that BV is the most common type of vaginal infection among women of reproductive age and accounts for at least one third of all vulvovaginal infections. BV is not caused by a single pathogen but rather it is a polymicrobial clinical syndrome. Common agents of BV include Gardnerella vaginalis, Mobiluncus, Bacteroides, saprophytes and Mycobacterium Hominus.\textsuperscript{8}

Candidiasis is mostly due to candida albicans\textsuperscript{9} and may be associated with diabetes, pregnancy and prolong use of antibiotics. Patient presents with vaginal discharge and pruritis. Discharge appears to be like curdled milk and deep erythema of vulva and vagina is often seen.

Trichomoniasis is a sexually transmitted disease (STD) that results from infection with flagellated protozoa named as Trichomonas Vaginalis. The prevalence of Trichomoniasis in American women is 3–5 million WHO estimates the worldwide prevalence of Trichomoniasis to be 170 million. The discharge is thin copious and pools in the vaginal vault. On examination vaginal and vulvar erythema is noted. The strawberry cervix in trichomoniasis resulting from punctuate hemorrhage is usually observed with colposcopy.

Risk factors associated with developing Bacterial vaginosis are include intrauterine contraceptive device, multiple sexual partners, recent antibiotics use and passive cigarette smoking. Although sexual intercourse is thought to play a role in its transmission, Bacterial vaginosis is not considered exclusively sexually transmitted disease.

Patients with bacterial vaginosis most commonly present with a foul (musty) fishy vaginal odor or a thin, white vaginal discharge. The diagnosis of bacterial vaginosis is determined if three of the following four sings (Ames’s Criteria) are Present-

1. Presence of clue cells.
2. Homogenous white, non-inflammatory discharge that adheres to the vaginal walls.
3. PH of vaginal fluid >4.5

4. Fishy odour from vaginal discharge before or after addition of 10 % potassium hydroxide.\textsuperscript{10}

Bacterial vaginosis is the syndrome thought to be the most prevalent cause of vaginitis. Several clinical diagnostic criteria, gram stain methods and biochemical markers have been developed to aid the diagnosis. Gram stain of vaginal secretion is relatively rapid, objective and in expensive method of diagnosing BV by identifying the characteristic change in the vaginal flora. It offers the advantage of allowing retrospective diagnosis.\textsuperscript{11} The interpretation of Gram staining is done by Nugent's scoring.\textsuperscript{12}

The present study was conducted to know the prevalence of bacterial vaginosis in sexually active females coming with the chief complaint of vaginal discharge to Obstetrics and Gynaecological OPD, Jay Prakash Narayan Hospital, Gaya, Bihar.

**MATERIALS AND METHODS**

This is a descriptive study and was conducted in Department of Obstetrics & Gynaecology, Anugrah Narayan Magadh Medical College, Gaya, and associated Hospital of Bihar (Jay Prakash Narayan Hospital, Gaya) between November 2016 and November 2017.

A total of 200 otherwise healthy women of reproductive age group with the complaint of excessive vaginal discharge were included in the study. We exclude the patients who are married, women with known skin disease and post-menopausal. In addition to a detailed history every patient underwent complete clinical examination and relevant investigations, then the data recorded in proforma. While examining the nature, colour and consistency of discharge the pH was also checked. The pH was measured with pH paper held with forceps and dipped into the vaginal discharge, care was taken to avoid contamination with cervical secretion as it falsely changes pH. Additionally, Whiff or Amine test was performed by mixing vaginal secretion with 10% KOH on the glass slide. Two plain cotton wool sterile vaginal swabs were used for High Vaginal Swab (HVS) for each patient. The swab was rubbed and rotated in post vaginal fornix.

During this period, married women in reproductive age groups (18-36 years of age), with the complain of vaginal discharge were included in the study. Consent was taken and the identity of the patients was kept confidential. Patients were managed on the basis of algorithms of the syndromic approach recommended by National AIDS control organization (NACO), India, after carrying out risk assessment.\textsuperscript{13,14} The laboratory tests were done at the department of Microbiology Anugrah Narayan Magadh Medical College, Gaya, and associated Hospital of Bihar (Jay Prakash Narayan Hospital, Gaya). Normal saline wet-mount slide preparations were made and observed under light microscope at 100x and 400x magnification for detection of motile trichomonads and budding yeast cells. For diagnosis of bacterial vaginosis, Gram stained smear were examined and graded as per standardized, quantitative, and morphological classification developed by Nugent. Briefly,
Lactobacillus morphotype were scored as 0, 1, 2, 3 and 4 when number of Lactobacilli morphotypic bacilli per oil immersion field were >30, 5-30, 1-4, <1 and 0, respectively. Gardnerella morphotype were scored as 0, 1, 2, 3 and 4 when number of Gardnerella morphotypic bacilli per oil immersion field were 0, <1, 1-4, 530 and >30. While, Mobiluncus morphotype were scored as 0, 1 and 2 when number of Mobiluncus morphotypic bacilli were 0, <1 and >5, respectively. Total score of ≥7 was considered positive for bacterial vaginosis, 4-6 was intermediate, <4 was negative.15 Gram stained smears of vaginal discharge were also examined for white blood cell (WBC) and clue cells. The presence of pseudohyphae and/or budding yeast cells was considered diagnostic of Candida infection. Three millilitres of blood were collected and transported to Serology laboratory for serological testing for syphilis; sera were separated by centrifuging at 2500 rpm for 10 minutes and stored at 2-8°C, if required. Venereal Diseases Research Laboratory (VDRL) test was carried out using antigen from Serologist to Govt. of India, Kolkata, India. Specimens reactive by qualitative tests were subsequently subjected to quantitative VDRL test with successive two-fold dilutions of the serum in 0.9% saline. All the sera reactive in qualitative VDRL test were confirmed for specific anti-treponemal antibodies by Treponema pallidum Haemaggulination Test (TPHA) test (Plasmatec Laboratory Products Ltd.), strictly according to the manufacturer's instructions.

**Statistical Analysis**

The X2 (Chi-square) test and analysis using the statistical software (SPSS version 18) was performed for quantitative variables to check for relationship in detecting the prevalence of bacterial vaginosis in sexually active females coming with the chief complaint of vaginal discharge to Obstetrics and Gynaecological OPD, Jay Prakash Narain Hospital, Gaya, Bihar and to know its effect on them. Percentages were calculated directly for the prevalence of bacterial vaginosis in sexually active females coming with the chief complaint of vaginal discharge to Obstetrics and Gynaecological OPD, Jay Prakash Narain Hospital, Gaya, Bihar and to know its effect on them. P = 0.05 was used as the accepted significance level. The minimum age of women was 18 years, while maximum age was 36 years.

**RESULTS**

A total of 200 women attended Obstetrics & Gynaecology department, Anugrah Narayan Magadh Medical College, Gaya, and associated Hospital of Bihar (Jay Prakash Narain Hospital, Gaya) between November 2016 and November 2017. Over a period of one year, 200 women fulfilling the criteria were included in the study. Of these 200 women, majority were between 26 to 30 years of age 79 (39.5%) (Table 1) (Graph 1). The median age of the study population was 28 years (range = 18-36 years). With regards to level of education, more than one third women were illiterate, and very few had completed graduation (Table 2) (Graph 2). Large majority were house-wives/ unemployed (78.5%) (Table 3) (Graph 3). Most common cause of vaginal discharge was found to be bacterial vaginosis, with almost one-fifth showing positive smear and 19.5% having intermediate score as per Nugent's scoring. Vulvovaginal candidiasis was also a common cause of vaginal discharge (Table 4) (Chart 1). Out of 200 Gram stained smears, 59 (29.5%) showed presence of ≥5 pus cells/ Oil immersion field (1000x magnification). No etiological diagnosis for vaginal discharge could be established in around half of the women. Only 1 women were HIV positive; one showed VDRL reactivity, though the VDRL titre was low (1:4), she was also reactive by TPHA test. The patients were managed according to their presenting syndromes as per NACO guidelines (vaginal discharge with/without lower abdominal pain).

<table>
<thead>
<tr>
<th>Age Group (in Years)</th>
<th>No. of Patients</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥20</td>
<td>7</td>
<td>3.5%</td>
</tr>
<tr>
<td>21-25</td>
<td>71</td>
<td>35.5%</td>
</tr>
<tr>
<td>26-30</td>
<td>79</td>
<td>39.5%</td>
</tr>
<tr>
<td>31-36</td>
<td>43</td>
<td>21.5%</td>
</tr>
</tbody>
</table>

**Table 1. Socio-Demographic Profile of Study in the Age Group (n=200)**

<table>
<thead>
<tr>
<th>Education</th>
<th>No. of Patients</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Illiterate</td>
<td>83</td>
<td>41.5%</td>
</tr>
<tr>
<td>High school</td>
<td>21</td>
<td>10.5%</td>
</tr>
<tr>
<td>Intermediate school</td>
<td>68</td>
<td>34%</td>
</tr>
<tr>
<td>Graduate/Undergraduate Student</td>
<td>13</td>
<td>6.5%</td>
</tr>
<tr>
<td>Post-graduate/Post-graduate student</td>
<td>15</td>
<td>7.5%</td>
</tr>
</tbody>
</table>

**Table 2. Socio-Demographic Profile of Study in the Education (n=200)**

<table>
<thead>
<tr>
<th>Occupation</th>
<th>No. of Patients</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Daily wages</td>
<td>14</td>
<td>7%</td>
</tr>
<tr>
<td>House-wife/ Unemployed</td>
<td>157</td>
<td>78.5%</td>
</tr>
<tr>
<td>Business</td>
<td>5</td>
<td>2.5%</td>
</tr>
<tr>
<td>Salaried</td>
<td>11</td>
<td>5.5%</td>
</tr>
<tr>
<td>Student</td>
<td>13</td>
<td>6.5%</td>
</tr>
</tbody>
</table>

**Table 3. Socio-Demographic Profile of Study in the Occupation (n=200)**

<table>
<thead>
<tr>
<th>Etiological Diagnosis of Vaginitis/ Vaginal Discharge Patients Managed According to Syndrome</th>
<th>Number (n=200)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacterial vaginosis: Positive Intermediate score</td>
<td>37</td>
<td>18.5%</td>
</tr>
<tr>
<td></td>
<td>39</td>
<td>19.5%</td>
</tr>
<tr>
<td>Vaginal candidiasis</td>
<td>27</td>
<td>13.5%</td>
</tr>
<tr>
<td>Trichomoniasis</td>
<td>1</td>
<td>0.5%</td>
</tr>
<tr>
<td>Total</td>
<td>104/200</td>
<td>52%</td>
</tr>
</tbody>
</table>

**Table 4. Etiological Diagnosis of Vaginitis/ Vaginal Discharge Patients Managed According to Syndrome**
DISCUSSION

Vaginal discharge is the second most common gynaecological problem after menstrual disorders. Some women regard almost any secretion from the vagina as abnormal discharge, and the first task for a primary care physician is to ascertain whether it is physiological or pathological. Although vaginitis is not a serious condition in strictly medical terms, it may have repercussion on woman’s life. The microbiology of vaginitis has been studied frequently and the most common types reported are Gardnerella, Candida and Trichomonas vaginitis.¹⁶

Then term 'Bacterial Vaginosis' (BV) is a variant of bacterial vaginitis and is the most prevalent vaginal infection.¹⁷ It is a clinical syndrome associated with Gardnerella and anaerobes and is characterized by foul smelling discharge. There are different diagnostic criteria like Amsel’s, Spiegella, and Nugent criteria. BV is the most common vaginal infection; however reported prevalence varies and based on the population studied. In our study, we included 200 married women, who were in the reproductive age group and presented with the general signs and symptoms suggestive of vaginal discharge. However, as seen in the present study only around one-third of the cases could be confirmed as having some infectious aetiology (bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis). Another 19.5% women had intermediate score for bacterial vaginosis. Such low rate of confirmation by laboratory methods of patients treated and managed syndromically has also been reported previously. Chauhan V et al detected bacterial vaginosis in 29.2%, C. albicans in 11.5% and T. vaginalis in 3.8% sexually active females with vaginal discharge.¹⁸ Similarly, Shah M et al found that of 183 cases diagnosed clinically as vaginal discharge syndrome, 38 (20.7%) were positive by laboratory investigations.¹⁹ Ray K et al reported high sensitivity of the syndromic approach for vaginal discharge syndrome, but the specificity of this method in diagnosing VD was low.²⁰ Another study done in sub-Saharan Africa found no significant associations between patient-reported STIs symptoms and laboratory confirmed STIs tests.²¹ This could be due to over-diagnosis of STIs by the syndromic approach, resulting in labelling of even the physiological discharge as pathological and unnecessary treatment. However, it might also be due to use of less than perfect techniques of specimen collection, transport and methods of laboratory diagnosis. Our study emphasizes this point and make us recommend laboratory testing in all cases of vaginal discharge. Efforts should be made to distinguish between the physiological and pathological discharge; infectious or non-infectious causes of vaginal discharge; and identify the causative agent of infectious vaginal discharges. Once the test results become available, switching treatment to specific therapy towards the particular agent seems to be the wisest recommendation. However, our study had few limitations. Firstly, the study was hospital based and not community based; therefore, findings may not entirely represent the local population as a whole. Also, conventional methods as
recommended by NACO were used for laboratory diagnosis of vaginal discharge, which might have missed a few cases.14

BV is a clinical syndrome characterized by disequilibrium in the vaginal microflora with decline in the number of lactobacilli.22–24 BV has been identified as an independent risk factor for the acquisition of sexually transmitted infections (STIs).25–29

Genital tuberculosis (GTB) is one form of extra pulmonary mycobacterium tuberculosis (MTB) and is responsible for a considerable proportion of female infertility, especially in developing countries.30–33

CONCLUSION
To conclude, the treatment maybe initiated on the basis of syndromic case management, however, it is essential that the treatment is modified as and when laboratory test results become available. In addition, the laboratory services network needs to be strengthened to ensure accurate and standardized availability of diagnostic services. The study showed higher prevalence of Bacterial Vaginosis. There was significant correlation between vaginal pH, IUCD user, history of STD, RTI, VDRL and HIV positive patients.

REFERENCES


