ORIGINAL RESEARCH

To Evaluate the Association between Vaginal PH Value and Type of Vaginal Microflora among Sexually Active Women in a Tertiary Hospital

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ABSTRACT

Background: The vagina acts as an external conduit that facilitates menstruation and childbirth. The vagina usually contains a distinctive flora that maintains its internal physical and chemical environment. The present study was conducted to assess association between vaginal pH value and type of vaginal microflora among sexually active women.

Materials & Methods: 150 sexually active women reporting to the Obstetric and Gynaecological department of Nalanda Medical college and hospital, Patna, Bihar were selected. Vaginal swabs were collected for vaginal *p*H measurement, Gram stain. Vaginal *p*H was evaluated immediately with the *p*H strips.

Results: Age group 20-30 had 80 (53.3%) and age group 30-40 years had 70 (46.7%) subjects. Out of 72 cases of bacterial vaginosis, 21 (29.1%) and 51 (69.9%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 46 cases of vulvovaginal candidiasis, 40 (86.9%) and 6 (12.1%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 8 cases of trichomonas vaginalis, 1 (12.5%) and 7 (87.5%) cases had pH range 4.0-5.0 and 5.0-6.0 respectively. All 5 (100%) gonorrhoea had pH range 4.0-5.0. Out of 6 non- specific urogenital cases, 2 (33.3%) and 4 (66.7%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 13 other causes, 3 (23.1%) and 10 (76.9%) had pH range 4.0-5.0 and 5.0-6.0 respectively. A significant difference was observed between causes of discharge and pH ($P< 0.05$).

Conclusion: Vaginal pH test can help monitor vaginal pH levels to identify vaginitis, particularly when used alongside other diagnostic methods. This tool can function as both a point-of-care test in clinics and a self-test at home. It can boost motivation for vaginal health checks and contribute to improved vaginal health.

Keywords: Bacterial vaginosis, pH, Vaginal microbiome

INTRODUCTION

The vagina acts as an external conduit that facilitates menstruation and childbirth. The vagina usually contains a distinctive flora that maintains its internal physical and chemical environment. Normal flora is dependent on the maintenance of various ecosystem components, which are in a state of dynamic equilibrium.[1] According to multiple published studies, the normal vaginal pH for women of childbearing age is moderately acidic, ranging from 3.8 to 5.0. A thin layer of transparent liquid, referred to as vaginal fluid, typically covers the normal vagina. The vaginal pH value can be altered or disrupted

by various factors, such as infections, the aging process, sexual intercourse, and the practice of vaginal douching.[2]

The typical vaginal microbiome, consisting of Lactobacilli species, can create an acidic pH and produce bacteriocins to eliminate other bacteria in the vagina. Lactobacilli generate an acidic milieu in the vagina, aimed at safeguarding women against sexually transmitted pathogens and opportunistic infections. The absence or significant reduction of these normal flora, such as Lactobacilli, can disrupt the balance of the vaginal ecosystem. This may result in the overgrowth of other microorganisms or bacteria within the vagina, potentially causing vaginitis.[3]

Vaginitis has different types, including bacterial vaginosis (BV), vaginal candidiasis, trichomoniasis, and aerobic vaginitis. Bacterial vaginosis is diagnosed by the presence of three out of four conditions, including homogenously milky vaginal discharge, vaginal pH over 4.5, positive KOH whiff test, and 20% at least of clue cells under wet-mount test by microscope.[4]

Vaginitis risk is heightened by abnormal pH levels, and vaginal pH measurement has been employed for preliminary screening. Prior studies indicate that a vaginal fluid pH of 4–4.5 or lower indicates no vaginitis, while a pH above 4.5 indicates the presence of vaginitis and bacterial vaginosis (BV). However, in the case of a trichomonas vaginalis infection, the pH level can rise to 6.5 or above.[5]

AIM AND OBJECTIVES

Aim

The primary aim of this study was to evaluate the association between vaginal pH values and the types of vaginal microflora among sexually active women attending a tertiary hospital. By understanding this relationship, the study sought to enhance diagnostic accuracy and inform treatment strategies for vaginal infections.

Objectives

1. To determine the distribution of vaginal pH levels among sexually active women presenting with vaginal discharge.
2. To identify the prevalence of different types of vaginal microflora associated with varying pH levels.
3. To assess the correlation between specific vaginal infections and corresponding pH ranges.
4. To analyze the age-wise distribution of subjects in relation to vaginal pH and microflora types.

MATERIALS AND METHODS

Study Design

This was a hospital-based cross-sectional observational study aimed at evaluating the association between vaginal pH and the type of vaginal microflora among sexually active women.

Study Population

The study included 150 sexually active women reporting to the Obstetrics and Gynaecology Department at Nalanda Medical College and Hospital, Patna, Bihar. All participants provided informed written consent before inclusion in the study.

Study Place

The research was conducted in the Department of Obstetrics and Gynaecology, Nalanda Medical College and Hospital, Patna, Bihar, India.

Study Duration

The exact study duration is not mentioned, but it is implied that data collection occurred over a defined period during which 150 subjects were recruited.

Study Place

The study was conducted in the Department of Obstetrics and Gynecology, Nalanda Medical College and Hospital, Patna, Bihar, India.

Study Duration

The study was carried out over a period of 24 months, from April 2017 to February 2019, allowing for recruitment, examination, and analysis.

Inclusion Criteria

Sexually active women.

Women attending the Obstetrics and Gynaecology outpatient department.

Women who provided informed written consent.

Exclusion Criteria

Pregnant women.

Women on antibiotics or antifungal treatment within the last 2 weeks.

Women with a history of recent vaginal douching or intercourse in the last 48 hours.

Women with known immunosuppressive conditions such as HIV/AIDS.

Menstruating women at the time of sample collection.

Ethical Considerations

- Institutional Ethical Clearance was obtained from the ethics committee of Nalanda Medical College and Hospital before the commencement of the study.
- Written informed consent was taken from all participants.

- Confidentiality of patient data was maintained throughout the study.

Study Procedure

1. Data Collection: A structured proforma was used to record demographic data (age, marital status, symptoms, etc.) and medical history.
2. Clinical Examination:

General and gynaecological examinations were performed.

Vaginal discharge was noted (colour, consistency, odour).

3. Sample Collection:

Vaginal swabs were collected using sterile speculum examination without lubrication.

One swab was used to immediately assess vaginal pH using commercially available pH indicator strips (range 3.8–6.5). The strip was applied to the lateral vaginal wall, and the colour change was matched with the scale.

Another swab was taken for Gram staining to assess the type of vaginal microflora and diagnose bacterial vaginosis, candidiasis, or normal flora using Nugent's scoring or equivalent.

4. Microbiological Evaluation:

Gram-stained smears were examined under a microscope to classify vaginal flora based on the presence of Lactobacilli, clue cells, and fungal elements.

Outcome Measures

Primary outcome: Association between vaginal pH value and type of vaginal microflora (normal, bacterial vaginosis, candidiasis, or mixed infection).

Secondary outcomes: Prevalence of abnormal vaginal microflora among symptomatic and asymptomatic women.

Statistical Analysis

- The data were compiled and analyzed using appropriate statistical software SPSS 21.0. version.
- Descriptive statistics such as mean, standard deviation, and frequency distributions were calculated.
- Chi-square test or Fisher's exact test was used to assess the association between categorical variables such as pH levels and types of microflora.
- P-value < 0.05 was considered statistically significant.

RESULTS

Table 1: Age Wise Distribution of Subjects

Age group (years)	Number	Percentage
20-30	80	53.3%
30-40	70	46.7%

Table 1 shows that age group 20-30 had 80 (53.3%) and age group 30-40 years had 70 (46.7%) subjects.

Table 2: Assessment of Cause of Discharge and pH

Cause	pH		P value
	4.0-5.0	5.0-6.0	
Bacterial vaginosis (72)	21 (29.1%)	51 (69.9%)	0.01
Vulvovaginal candidiasis (46)	40 (86.9%)	6 (12.1%)	0.01
Trichomonas vaginalis (8)	1 (12.5%)	7 (87.5%)	0.02
Gonorrhoea (5)	5 (100%)	0	0.01
Non- specific urogenital (6)	2 (33.3%)	4 (66.7%)	0.03
Others (13)	3 (23.1%)	10 (76.9%)	0.02
Total	72 (48%)	78 (52%)	

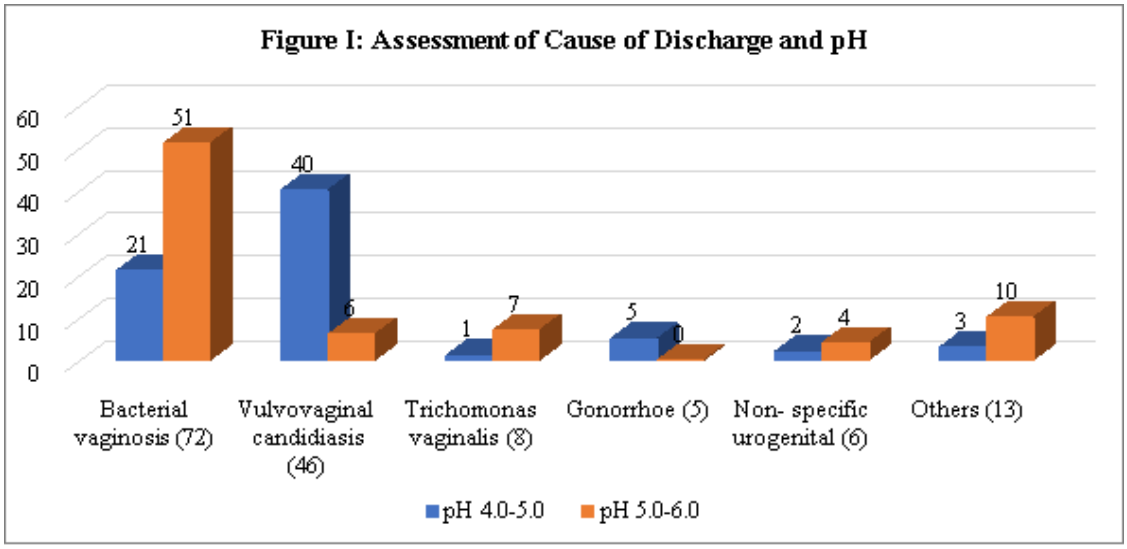

Table 2 and Figure I illustrate the distribution of vaginal pH levels across different causes of vaginal discharge among 150 cases. Among the 72 cases of bacterial vaginosis (BV), 21 (29.1%) exhibited a pH between 4.0 and 5.0, while 51 (69.9%) had a pH between 5.0 and 6.0, suggesting that BV is more commonly associated with a higher vaginal pH.

In the 46 cases of vulvovaginal candidiasis (VVC), 40 (86.9%) had a pH between 4.0 and 5.0, and 6 (12.1%) had a pH between 5.0 and 6.0, indicating that VVC is typically associated with a lower, more acidic vaginal pH.

Out of the 8 cases of *Trichomonas vaginalis* infection, 1 (12.5%) had a pH between 4.0 and 5.0, and 7 (87.5%) had a pH between 5.0 and 6.0, suggesting a strong association with higher vaginal pH levels.

All 5 cases (100%) of gonorrhoea had a pH between 4.0 and 5.0, indicating an association with a more acidic vaginal environment.

Among the 6 cases of non-specific urogenital infections, 2 (33.3%) had a pH between 4.0 and 5.0, and 4 (66.7%) had a pH between 5.0 and 6.0, showing a tendency towards higher pH levels.

In the 13 cases classified as 'others,' 3 (23.1%) had a pH between 4.0 and 5.0, and 10 (76.9%) had a pH between 5.0 and 6.0, again indicating a prevalence of higher pH levels.

Overall, out of the 150 cases, 72 (48%) had a vaginal pH between 4.0 and 5.0, and 78 (52%) had a pH between 5.0 and 6.0.

DISCUSSION

The vagina's pH level, known as the "vaginal pH value," is crucial for assessing vaginal health. The pH value is used to measure and determine whether the state is acidic and/or alkaline, based on the scale of hydrogen ion activity. Although the naturally neutral pH level is 7, the normal vaginal pH is moderately acidic, ranging from 3.8 to 5.0.[6] A more acidic vaginal environment (with a lower pH value than that of blood or interstitial fluids) can safeguard the vaginal mucosa against pathogenic organisms. Overall health conditions, such as age, vaginal hydration status, daily diet, and safe intercourse, can influence the vaginal pH. The pH value of the vagina is dependent on age. The normal vaginal pH value for a woman of reproductive age ranges from 4.0 to 4.5, but the value may be slightly higher than 4.5 among premenarchal and postmenopausal women.[7]

A woman's daily life includes various influences that can disrupt the normal balance of vaginal pH, including unprotected intercourse, antibiotic use, vaginal douching, and changes in the menstrual cycle.[8] Engaging in sexual activities without protection may result in a vaginal pH imbalance. The pH of semen is about 8.0, making it somewhat alkaline. During sex without protection, this can affect the vaginal pH. Semen can activate the proliferation of bacteria that serve as a physiological buffer.[9] Thus, unprotected sex can significantly alter the vaginal pH so that it remains elevated even after 10–14 h. As a result of this change, the vagina is less shielded from infection. Antibiotics can inhibit bacterial growth or kill bacteria to treat bacterial infections. In clinical routine, antibiotics are frequently used to treat vaginitis.[10]

The present study was conducted to assess association between vaginal pH value and type of vaginal microflora among sexually active women.

In our study, age group 20-30 had 80 (53.3%) and age group 30-40 years had 70 (46.7%)

subjects. Hemalatha R et al[11] studied 270 women. Priyadarshini[12] enrolled 100 sexually active women in reproductive age group with complain of abnormal vaginal discharge in their study

It was found that out of 72 cases of bacterial vaginosis, 21 (29.1%) and 51 (69.9%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 46 cases of vulvovaginal candidiasis, 40 (86.9%) and 6 (12.1%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 8 cases of trichomonas vaginalis, 1 (12.5%) and 7 (87.5%) cases had pH range 4.0-5.0 and 5.0-6.0 respectively. All 5 (100%) gonorrhoea had pH range 4.0-5.0. Out of 6 non- specific urogenital cases, 2 (33.3%) and 4 (66.7%) had pH range 4.0-5.0 and 5.0-6.0 respectively. Out of 13 other causes, 3 (23.1%) and 10 (76.9%) had pH range 4.0-5.0 and 5.0-6.0 respectively.

In the present study, 69.9% of Bacterial Vaginosis (BV) cases were associated with a vaginal pH between 5.0 and 6.0. This finding aligns with the study by Hemalatha et al. (2013), which reported that elevated vaginal pH levels are significantly associated with BV, due to the reduction of hydrogen peroxide-producing Lactobacillus species and an overgrowth of anaerobic bacteria.[11] The elevated pH creates an environment conducive to the proliferation of pathogenic bacteria, contributing to the development of BV. Hemalatha R et al[11] found that the mean vaginal *p*H in women with BV measured by *p*H strips and *p*H glove was 5 and 4.9, respectively. The vaginal *p*H was significantly higher in women with BV. Vaginal discharge was prevalent in 84.8 per cent women, however, only 56.8 per cent of these actually had BV by Nugent score (NS). Presence of clue cells and positive whiff test were significant for BV. Vaginal *p*H >4.5 by *p*H strips and *p*H Glove had a sensitivity of 72 and 79 per cent and specificity of 60 and 53 per cent, respectively to detect BV. Among the combination criteria, clue cells and glove *p*H >4.5 had highest sensitivity and specificity to detect BV.

A significant proportion (86.9%) of Vulvovaginal Candidiasis (VVC) cases exhibited a vaginal pH between 4.0 and 5.0, indicating that Candida infections typically occur in a more acidic environment. This observation is consistent with the findings of Bitew and Abebaw (2018), who noted that Candida species thrive in acidic conditions and that VVC does not significantly alter the vaginal pH.[12]

Infections with Trichomonas vaginalis were predominantly associated with higher vaginal pH levels, with 87.5% of cases exhibiting a pH between 5.0 and 6.0. This finding corroborates the study by McLaughlin et al. (2018), which indicated that Trichomonas infections elevate vaginal pH due to the organism's metabolic activities and the resultant inflammatory response.[13]

All cases of gonorrhoea in the study presented with a vaginal pH between 4.0 and 5.0. While Neisseria gonorrhoeae infections are not typically associated with significant changes in vaginal pH, this finding suggests that gonorrhoea can occur in a normally acidic vaginal environment. This observation is supported by the study of McLaughlin et al. (2018), which found that elevated vaginal pH is associated with an increased risk of gonococcal infection.[13]

Cases categorized under non-specific urogenital infections and other causes showed a tendency towards higher vaginal pH levels, with 66.7% and 76.9% of cases, respectively, falling within the 5.0 to 6.0 pH range. This suggests that elevated vaginal pH may be a common feature in various non-specific or mixed infections, potentially due to the disruption of normal vaginal flora. Mania-Pramanik et al. (2008) emphasized that increased vaginal pH is an indicator of bacterial vaginosis and may be associated with other reproductive manifestations.[14]

Overall, the study reinforces the clinical utility of vaginal pH measurement as a diagnostic tool. Elevated vaginal pH levels are indicative of infections such as BV and trichomoniasis, whereas normal or acidic pH levels are more consistent with VVC and gonorrhoea. Incorporating pH assessment into routine gynecological evaluations can enhance diagnostic accuracy and inform appropriate treatment strategies.

LIMITATIONS OF THE STUDY

- **Limited Generalizability**: The study focused exclusively on symptomatic women presenting with vaginal discharge, which limits the applicability of the findings to asymptomatic populations. As noted by a study published in *Perspectives in Medical Research*, such a focus restricts the extrapolation of results to the broader female population.
- **Single-Centre Design**: Conducting the research in a single tertiary hospital may introduce selection bias and limit the

generalizability of the findings to other settings or populations. Multi-center studies are recommended to enhance the diversity and applicability of results.

- **Cross-Sectional Nature**: The study's cross-sectional design captures data at a single point in time, preventing the assessment of temporal changes in vaginal pH and microflora. Longitudinal studies are necessary to understand the dynamics and causality of these associations.
- **Incomplete Microbial Characterization**: The study did not perform biochemical characterization of anaerobes due to financial and logistical constraints, potentially missing key insights into the vaginal microbiota.

CONCLUSION

Authors found that vaginal pH test can help monitor vaginal pH levels to identify vaginitis, particularly when used alongside other diagnostic methods. This tool can function as both a point-of-care test in clinics and a self-test at home. It can boost motivation for vaginal health checks and contribute to improved vaginal health.These findings underscore the importance of vaginal pH as a diagnostic tool in identifying the type of vaginal infection. Monitoring pH levels can aid clinicians in early detection and appropriate management of vaginal infections, ultimately improving women's reproductive health outcomes.

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