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VAGINAL FLUID pH FOR DETECTION OF BACTERIAL VAGINOSIS

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ABSTRACT

Background: Bacterial vaginosis (BV) is the most common cause of abnormal vaginal discharge and malodor in women of reproductive age. BV reflects a shift in normal vaginal flora to mixed anaerobic and gram negative flora increasing the vaginal pH, which eventually increases vulnerability to sexually transmitted diseases and complicates pregnancies. The objective of this study was to evaluate the accuracy of vaginal pH individually and its combinations with other factors of Amsel's criteria to diagnose bacterial vaginosis.

Methods: The study was carried out in gynaecology department in K.V.G. Medical College Hospital, Sullia over a period of 6 months. Vaginal swabs of women complaining of white discharge pv were collected using 2 sterile swabs. One, to prepare wet mount and to assess for amine test, other for Grams staining. Vaginal pH was measured using pH strips. A total of 105 patients were checked for the confirmation of Bacterial Vaginosis according to the Amsel's Criteria and Nugent's Scoring.

Results: The prevalence of bacterial vaginosis is 39% by Amsel's criteria and 33% by Nugent's scoring. Comparing with Nugent scoring methods, the clinical diagnosis by Amsel's criteria had sensitivity of 85.3%, specificity of 95%. Vaginal pH individually had a sensitivity of only 73.4%. Combination of vaginal pH test with clue cells had highest sensitivity while vaginal pH test with whiff test was comparatively less sensitive.

Conclusion: Amsel's criteria diagnosis can be simplified by using a combination of 2 criteria, pH of secretions and presence of clue cells but if lab equipment is unavailable, using vaginal fluid pH can still produce moderately accurate results.

KEYWORDS :

INTRODUCTION:

Bacterial vaginosis (BV) is the most common cause of vaginal discharge and malodor in women of reproductive age.[1] Bacterial vaginosis reflects a shift in normal vaginal flora from hydrogen peroxide producing lactobacilli to mixed flora like Gardnerella vaginalis, mycoplasmas and anaerobes such as *Mobiluncus*, *Peptostreptococcus*.[2] This polymicrobial superficial vaginal infection involving loss of the normal lactobacilli causes increase in vaginal pH (>4.5)

There is no known reason why it is so common. Though closely related to sexual intercourse it is not termed as a sexually transmitted disease because of its high prevalence in sexually inactive women.[4] Recent study shows association between high vaginal pH and cervicovaginal inflammatory cytokines that are implicated in increased vulnerability to HIV, sexually transmitted diseases, preterm delivery in asymptomatic women with BV.[5] BV has been linked to low birth weight infants, preterm delivery, chorioamnionitis, post hysterectomy cuff cellulitis, post-surgical endometritis, endometritis following vaginal delivery, and PID. Hence it is critical to diagnose and treat women affected with BV.

Risk factors include oral sex, douching, black race, cigarette smoking, sexual intercourse during menstruation, intrauterine device, early age of sexual intercourse, new or multiple sexual partners, sexual activity with other women.[2] Most of the patients are asymptomatic. Some patients show symptoms like increase in white discharge which appears homogeneous, is low in viscosity and evenly coats the vaginal mucosa and there is a characteristic fishy smelling vaginal odour. The patients may also experience itching and burning in the vaginal area.

The pH of vaginal secretions is maintained between 3.8 and 4.5 in healthy women and remains within range even during menstruation. A pH of 4.5 or less signifies the absence of vaginitis, whereas a pH of more than 4.5 is classified as vaginitis as the presence of BV leads to high pH of vaginal secretion. Other factors that are known to disturb this pH are antibiotic treatment,[7] inflammatory reactions and even intercourse[7] can lead to the elevated pH. In postmenopausal women the pH of vaginal secretion is increased, which is attributed to the decreasing level of estrogen.

Rapid screening with available resource is essential for a favorable health care outcome. Classical initial method of BV diagnosis was done by isolation of G. vaginalis from clinical specimen. Later on, with the advent of the anaerobic culture techniques other organisms were also detected from specimens. Amsel's clinical diagnosis and gram stain evaluation by Nugent methods are mostly used worldwide particularly in developing countries.

The Nugent scoring test requires health care experts, laboratory support, and also access to a high-power microscopy to obtain timely results for the diagnosis of BV. Since these necessities are not always available in rural places, it is important to have simple and reliable clinical criteria that clinicians can use in practice. Therefore, knowing the best diagnostic approach in a given area using the available resource helps in deciding the preferred method for diagnosis.

OBJECTIVES OF THE STUDY: To evaluate the accuracy of vaginal pH individually and its combinations with other factors of Amsel's criteria to diagnose bacterial vaginosis.

AMSEL'S CRITERIA: Amsel's criteria are used for the clinical diagnosis of BV, and a positive diagnosis is made when at least three of the four following criteria are present:
A thin homogeneous vaginal discharge
Vaginal pH ≥ 4.5
Positive whiff test
The presence of clue cells on wet mount.

SOURCE OF DATA:
This hospital based study was carried out in gynaecology department in K.V.G. Medical College Hospital, Sullia over a period of 6 months. Data was collected in a pre-designed form.

METHODS OF COLLECTION OF DATA:
Ethical committee clearance was obtained. Data was collected after taking written informed consent from the properly selected patients.

First, thorough history as per pre-prepared questionnaire, then clinical examination and laboratory tests were done. Those women complaining of white discharge per vagina were included in the study. Vaginal pH was measured using pH strips. Vaginal discharge was collected using 2 sterile cotton tip swabs. One, to prepare wet mount, then assess for amine test, other for Gram's staining. A total of 105 patients were checked for the confirmation of Bacterial Vaginosis according to the Amsel's Criteria and Nugent's scoring.

INCLUSION CRITERIA:
Patients complaining of excessive vaginal discharge in the age group of 18-45 years.

EXCLUSION CRITERIA:
Pregnant women
Known diabetics
On antibiotic treatment
History of intercourse on previous night
Per vaginal bleeding
Clinical diagnosis of bacterial vaginosis was considered positive if three of the four criteria in Amsel's diagnosis were met.

Gram stain diagnosis was based on a criterion score described by Nugent and considered positive if the score was 7–10. The Nugent criteria score vaginal flora as normal (0–3), intermediate (4–6), and bacterial vaginosis (7–10).

STATISTICAL ANALYSIS: - All relevant clinical information of the cases were recorded systematically in the predesigned clinical data sheet. Data was analyzed using SPSS (11.0).

A descriptive analysis was done and frequency, percentages and ratios were computed from the data.

Sensitivity, specificity, positive and negative predictive value of Amsel's criteria was calculated.

RESULTS:
The age range of the study participants was 18-45 with mean age was 32.5 years. [Table 1]

The prevalence of bacterial vaginosis was 39% by Amsel's criteria and 33% by Nugent scoring. Amsel's criteria had sensitivity of 85.3% and a specificity of 95.3% when compared using Gram stain evaluated by Nugent scoring method as standard. [Table 2]

When we compare individual Amsel's criteria with Nugent scoring, in the present study we found that presence of clue cells was the criteria with the highest sensitivity of 89.5% and specificity of 75.6%. [Table 3]

The sensitivity of vaginal pH and Whiff test was the same 73.7%. However, PPV and NPV of vaginal pH was higher in our study.

All criteria had high negative predictive value ranging between 88% - 91%.

When combination of two Amsel's criteria was considered, with vaginal pH along with presence of Clue cells, had the highest sensitivity of 81.6% with a PPV of 96% and a NPV of 94%. [Table 4] [Graph 10]

Combination of vaginal pH along with positive whiff test had a sensitivity of 73.7% with a PPV of 89% and a NPV of 87%. [Graph 11]

DISCUSSION:
The clinical diagnosis of bacterial vaginosis has long been made using Amsel's criteria or Nugent's criteria.

A study on the comparison of Pap smear and microbiological pattern in Bacterial Vaginosis done in Rajarajeshwari Medical College in Bangalore showed BV had a prevalence of 34.6% which is closely similar to ours which is 33% confirmed by Nugent's scoring method.

The flora of the female genital tract varies with the pH and estrogen concentration, which is dependent on the age of the host. In this study all women were within the reproductive age group, no one had diabetes, were not on any medication, and did not have sexual act on the previous night. These inclusion criteria were taken into consideration.

When we correlate any two of Amsel's criteria the presence of clue cells in vaginal wet mount and high vaginal pH had the greatest sensitivity (81.6%) and specificity (66.5%), respectively with high PPV and NPV.

That of vaginal pH with Whiff Test had a comparatively lower sensitivity, PPV and NPV.

In conditions where there is not enough time and gram stain procedure is cumbersome, combination of vaginal pH and clue cells detection can be used.

Therefore, combining pH tests with other symptoms can enhance the accuracy of the test in diagnosis of various infectious conditions. Various factors such as simultaneous infections of vagina and cervical mucus might have influence on these criteria.

From individual criteria for predicting the gram stain result, clue cells detection from wet mount microscopic examination is the most reliable predictor of bacterial vaginosis. It had a higher sensitivity(89.5%) , specificity(75.6%) and positive and negative predictive value. This is consistent with the study done by Mittal V et al.(2012).

Second most reliable predictor is alkaline vaginal pH with a significant sensitivity of 73.7% and higher PPV and NPV than that of whiff test. This is consistent with Dadhwal et al who also states that the decrease in sensitivity of whiff test may attribute to subjective nature of the test due sensation ability of the person doing the test.

Lack of acidity allows the proliferation of other organisms (microbial dysbiosis) which may be causative of underlying disease. As lactobacilli play a key role in acidifying the vagina, it is not surprising that vaginal pH remains part of Amsel's criteria in making the clinical diagnosis of bacterial vaginosis.

In a study by Mania Pramanik et al. done in KEM hospital in Mumbai, vaginal pH had a 100% PPV, as a tool for diagnosis of BV. However, in our study clue cells has the highest PPV and NPV in diagnosis of BV.

This difference in sensitivity and predictive value may be due to difference of study population, clinical case and higher prevalence of bacterial vaginosis.

CONCLUSIONS:

In patients complaining of white discharge per vagina, diagnosis by Amsel's criteria can be simplified by using a combination of 2 criteria, a high vaginal pH and the presence of clue cells on wet mount slide.

In our study high vaginal pH correlated well with the occurrence of BV. In developing countries microscopes are not readily available in all clinical settings and hence an easily performed pH evaluation in the OPD setting could be safely used to diagnose BV.

Table 1: Age Distribution Of The Patients Study.

Age Distribution	Number OF Patients	%
<25	7	6.66
26 -30	35	33.3
31 -35	37	35.23
36 -40	11	19.47
>41	15	14.28
Total	105	100.0

Table 2: Nugents Score

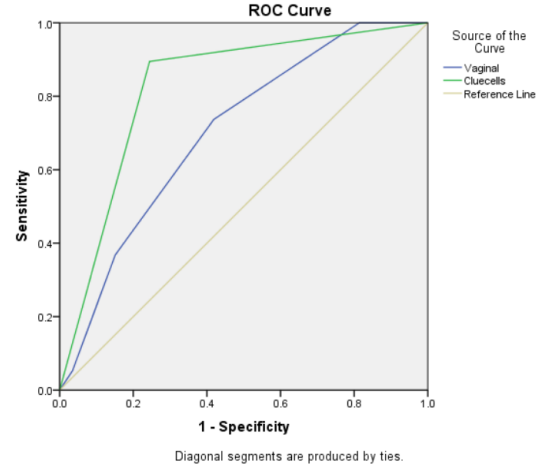
	NUGENTS SCORE	Sensitivity	Specificity	PPV	NPV	
AMSEL'S Criteria	35	03	85.36	95.31	92.1	91.5
	6	61				
Total	41	64				

Table 3: Diagnostic Accuracy Of Individual Clinical Criteria

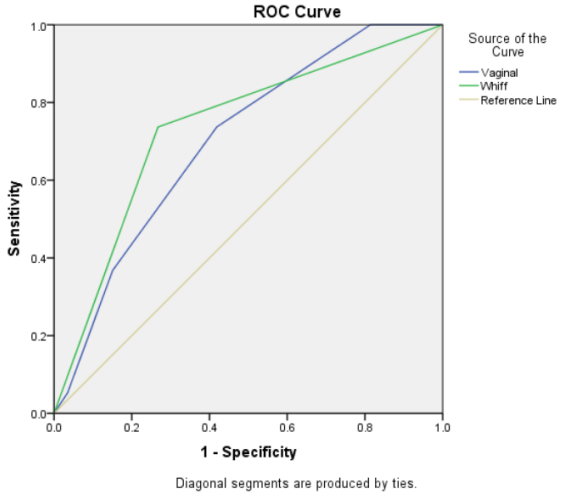
AMSEL Criteria	Sensitivity	Specificity	PPV	NPV
Vaginal pH	73.7	60	61	90
Whiff	73.7	73.3	60	88
Clue Cell	89.5	75.6	90	91

Table 4: Diagnostic Accuracy Of Combintion Of Two Criteria

AMSEL Criteria	Sensitivity	Specificity	PPV	NPV
Vaginal PH + Clue Cells	81.6%	66.85%	96.12	94.23
Vaginal PH + Whiff Test	73.7	65.65%	89.23	87.34



Graph 1: Diagnostic Accuracy Of Combintion Of Two Criteria (vaginal Ph + Clue Cells)



Graph 2: Diagnostic Accuracy Of Combintion Of Two Criteria (vaginal Ph + Whiff Test)

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