Role of Bacterial Vaginosis in threatened preterm and preterm labour, and its screening

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**Abstract**

**Background:** 8-10% pregnancies end in preterm labour or threatened preterm labour which has maternal and foetal complications just due to infections such as bacterial vaginosis, which can be identified by simple tests like Amsel Criteria and treated at the earliest with appropriate antibiotics.

**Aim:** To establish the role of bacterial vaginosis in preterm labour and threatened preterm labour based on Amsel criteria.

**Materials and Methods:** 100 pregnant women with gestational age between 28 and 36 weeks, of which 50 women with symptoms of threatened preterm labour and preterm labour were kept as study group and another 50 pregnant women in the same gestational age without any symptoms of threatened preterm or preterm labour were kept as control group. Examination of vaginal discharge, vaginal pH, microscopy for clue cells and Whiff test, which are Amsel’s criteria, were done. Chi-square test was used to find out the association of variables and p value less than 0.05 was taken as statistically significant.

**Results:** The women in study group had higher incidence of vaginal discharge than control group. All the bacterial vaginosis cases had vaginal pH >4.5. The negative predictive value of vaginal fluid pH > 4.5 in our study is 100%. In our study 27% cases in all groups were found to have whiff test positive. 20% had clue cells on microscope examination. All the Amsel’s criterion was statistically highly significant between the study and the control group.

**Conclusion:** The strong correlation between presence of Bacterial Vaginosis and the incidence of threatened preterm and preterm labour is established. Amsel’s criteria are simple, inexpensive, easily reproducible method of diagnosing BV and can be used on a mass scale. By diagnosing and treating bacterial vaginosis in threatened preterm labour, preterm deliveries can be prevented to a certain extent, if not atleast in order to buy time for the inclusion of steroids which will decrease the perinatal morbidity and mortality by decreasing Respiratory Distress Syndrome and intraventricular haemorrhage.

**Keywords:** Bacterial vaginosis, Amsel’s criteria, Threatened preterm, Preterm labour, Whiff test, Clue cells, Vaginal pH

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**Introduction**

The dream of every woman is to deliver a healthy and mature baby capable of adapting satisfactorily to the extrauterine life. Even though the total Gestational period is 280 days or 40 weeks, atleast 37 completed weeks of gestation is necessary for the fetus to attain complete growth and maturity.

Almost 8% to 10% pregnancies end in preterm labour or threatened preterm labour which has maternal and fetal complications just due to infections such as bacterial vaginosis, which can be identified by simple tests.

Preventing preterm birth and its consequences is one of the obstetricians’ most important tasks. Neonatal morbidity and mortality and maternal morbidity is a major contributor in preterm births. Prevention of preterm labour (PTL) has assumed special importance in practice of obstetrics.

Almost 40% cases of PTL and threatened preterm labour are mostly caused by bacterial vaginosis and when treated with appropriate antibiotics it can be prevented in upto 70% to 80% cases. This can be detected by applying simple tests such as AMSEL CRITERIA.

Vaginal infections particularly bacterial vaginosis have consistently shown in many longitudinal population studies to be associated with preterm labour. Bacterial Vaginosis (BV) is a polymicrobial, vaginal infection caused by over growth of anaerobic bacteria like Gardenella vaginalis, Mycoplasma hominis, and Mobiluncus species, Ureaplasma urealyticum, and Chlamydia trachomatis, Peptostreptococcus, and Bacteroides in the vagina with a corresponding decrease in the number of Lactobacillus. 1,2,3,4,5,6,7 37 completed weeks of gestation is necessary for the fetus to attain complete growth and maturity.

Eschenbach et al⁸ and Gravett & colleagues⁹ were the first to implicate bacterial vaginosis as a risk factor for preterm labour. Bacterial vaginosis and its related organism have been implicated in higher rates of preterm premature rupture of membranes, late miscarriages,
spontaneous preterm, preterm birth, chorioamnionitis and postpartum endometritis.

The problem is that the half of the population of women with bacterial vaginosis is asymptomatic. The mechanism by which BV causes preterm labour and other conditions is exactly not known, but there is evidence that it causes infections of the upper genital tract which in turn causes preterm labour. BV is not a strong predictor of early miscarriage but may be a predictor in miscarriage after 13 week’s gestation. Uniform recommendations on effective screening methods to prevent preterm delivery are still lacking. Integrating a simple infection screening method into routine antenatal care can reduce the rate of preterm birth. This helps us to prevent the perinatal mortality and morbidity by preventing preterm labour. In clinical practice bacterial vaginosis is diagnosed using the Amsel criteria.

**Aims and Objectives**
1. Primary aim is to establish the role of bacterial vaginosis in preterm labour and threatened preterm labour based on Amsel criteria.
2. Secondarily to highlight the fact that the diagnosis of bacterial vaginosis can be done by simple and efficient procedures – Amsel criteria.

**Materials and Methods**

After obtaining the institutional ethics committee approval 100 pregnant women who are admitted in the labour ward with gestational age between 28 and 36 weeks are chosen for the study of which 50 women with symptoms of threatened preterm labour and preterm labour were kept as study group and another 50 pregnant women in the same gestational age without any symptoms of threatened preterm or preterm were kept as control group. Informed consent was obtained from all.

**Inclusion Criteria**
- Gestational age between 28 to 36 weeks
- Threatened preterm labour
  - Presence of menstrual like cramp
  - Low dull backache
  - Pressure symptoms
  - Excessive vaginal discharge
- Preterm premature rupture of membranes
- Previous history of premature rupture of membrane
- Established preterm labour
  - Regular uterine contractions
  - Dilatation > 2cm
  - Effacement of cervix > 80%
- Amnion test also known as Whiff test was done by addition of 10% potassium hydroxide to the discharge on the blade of the speculum and noting the emission of an offensive amine or fishy odour which indicates a positive amine test.

**Exclusion Criteria**
- Multiple pregnancy
- Pre eclampsia
- Fibroid uterus
- Polyhydramnios
- Ante partum haemorrhage
- Malformations of uterus
- Incompetent cervical os
- Foetal anomaly
- Medical complications like diabetes, hypertension, anaemia, pyrexia

**Statistical Analysis:** Data was entered into an Excel sheet and SPSS version was used to analyze the study. Chi-square test was used to find out the association of variables and p value less than 0.05 was taken as statistically significant.

**Study Tools**

Method = Amsel’s Criteria
1. Homogenous thin vaginal fluid that adheres to vaginal wall.
2. Vaginal pH greater than 4.5.
3. An amine odour when vaginal secretions are mixed with 10% potassium hydroxide - WHIFF TEST.
4. Presence of clue cells in the vaginal discharge, seen in wet mount.

The diagnosis of bacterial vaginosis is defined if three of the four criteria are fulfilled.

**Materials**
- Sims speculum, pH indicator paper, 10% KOH, sterile swab, slides, cover slips, normal saline, microscope, sterile gloves.

**Collection of specimen**

Selected patients were interviewed first reviewing obstetrical, medical and sexual history. A thorough general and systemic examination was done. A detailed obstetrical examination was done to note the fundal height, presentation and the uterine contraction. Informed consent was taken and the patient was asked to lie in the dorsal position. After separating the labia with fingers of left hand, a sterile sims speculum was gently introduced into the vagina. While performing speculum examination no lubricant was used as it can falsely increase the vaginal pH. Amount, colour and odour of the secretion was noted. The vaginal pH was measured with the pH strip held with the forceps and dipped into the secretion. Change in colour was noted. A pH of more than 4.5 was taken as positive for bacterial vaginosis. With a sterile cotton swab stick, a high vaginal swab from the posterior fornix was taken for vaginal smear. The smear of vaginal secretion was diluted with normal saline and examined microscopically for identification of clue cells.

**Results**

The study consisted of 100 cases; which includes 50 cases in study group and 50 cases in control group. All the subjects included in the study group satisfied the inclusion criteria. Vaginal pH strips test, Whiff test was
done and fishy odour noted by adding 10% KOH. Vaginal smear was taken and wet smear examined for presence or absence of clue cells.

**Table 1: Age Distribution**

<table>
<thead>
<tr>
<th>Age (Yrs)</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>20-24</td>
<td>19</td>
<td>22</td>
</tr>
<tr>
<td>25-29</td>
<td>16</td>
<td>23</td>
</tr>
<tr>
<td>30-34</td>
<td>11</td>
<td>3</td>
</tr>
<tr>
<td>&gt;35</td>
<td>4</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 1: Shows the age distribution. Majority of women in study and control groups were between 20 – 24 years. The concentration of women in this group may be due to early marriages which are common.

**Table 2: Gestational age of subjects**

<table>
<thead>
<tr>
<th>Gestational age (weeks)</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>28-30 weeks</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>31-33 weeks</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>34-36 weeks</td>
<td>41</td>
<td>41</td>
</tr>
</tbody>
</table>

Table 2: Shows the distribution of cases in both groups according to the period of gestation in weeks. Preterm or threatened preterm labour was seen in 34 – 36 weeks of gestational age at higher frequency.

**Table 3: Vaginal discharge in both groups**

<table>
<thead>
<tr>
<th>Vaginal Discharge (VD)</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>20 (40%)</td>
<td>2 (4%)</td>
</tr>
<tr>
<td>Negative</td>
<td>30 (60%)</td>
<td>48 (96%)</td>
</tr>
</tbody>
</table>

Table 3: Shows vaginal discharge in both groups. The difference was statistically significant difference in vaginal discharge positive women with p value < 0.001. This finding supports the opinion that many subjects with BV were symptomatic.

**Table 4: Distribution of cases according to pH**

<table>
<thead>
<tr>
<th>pH</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤4.5</td>
<td>26 (52%)</td>
<td>46 (92%)</td>
</tr>
<tr>
<td>&gt;4.5</td>
<td>24 (48%)</td>
<td>4 (8%)</td>
</tr>
</tbody>
</table>

Table 4: shows vaginal pH in both groups. The difference was statistically significant in vaginal pH > 4.5 group, p value < 0.001. Though this is a simple and economical test to perform, there are other factors like infection, semen blood, cervical mucus etc, which may alter the vaginal pH.

**Table 5: Whiff test in both groups**

<table>
<thead>
<tr>
<th>Whiff Test</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>24 (48%)</td>
<td>3 (6%)</td>
</tr>
<tr>
<td>Negative</td>
<td>26 (52%)</td>
<td>47 (94%)</td>
</tr>
</tbody>
</table>

Table 5 shows comparison of whiff test in both groups. The difference was statistically significant with p value < 0.001 in the Whiff Test positive group.

**Table 6: Clue cells in both groups**

<table>
<thead>
<tr>
<th>Clue Cells</th>
<th>Study Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Present</td>
<td>18 (36%)</td>
<td>2 (4%)</td>
</tr>
<tr>
<td>Absent</td>
<td>32 (64%)</td>
<td>48 (96%)</td>
</tr>
</tbody>
</table>

Table 6 shows comparison of clue cells in both groups. The difference was statistically significant with P < 0.001 between the study and control group where the clue cells were present. Detection of clue cells is the
most sensitive and specific criteria of the diagnosis of BV, but it may be mistaken by debris, degeneration of cells to which lactobacilli are adherent. The quality of microscope and the adequacy of specimen are additional factors which affect the accuracy with which the clue cells are detected.

Fig. 3: Distribution of cases according to the presence of clue cells

Table 7: Incidence of bacterial vaginosis

<table>
<thead>
<tr>
<th></th>
<th>Study Group n=50</th>
<th>Control Group n=50</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Women with BV</strong></td>
<td>24 (48%)</td>
<td>3 (6%)</td>
</tr>
<tr>
<td><strong>Women without BV</strong></td>
<td>26 (52%)</td>
<td>47 (94%)</td>
</tr>
</tbody>
</table>

Table 7 shows the incidence of BV in both the groups. The incidence of BV was higher among the study group compared to control group. The difference was statistically significant with p value <0.001.

As most of the patients in high risk group were included in present study, the incidence of threatened preterm and preterm labour was more in women with BV, when compared to women without BV.

Discussion

Bacterial vaginosis is the most common lower genital tract disorder among the women of reproductive age group and the most prevalent cause of vaginal discharge and malodour. It has been associated with the significant number of obstetric and gynaecological complication such as preterm labour and delivery, preterm premature rupture of membranes, spontaneous abortion, chorioamnionitis, postpartum endometritis, post cesarean wound infection, post-surgical infections and subclinical pelvic inflammatory diseases.

In our study a total of 100 women were enrolled who presented with threatened preterm labour and preterm labour. At the time of admission to the labour room, nature of vaginal discharge was noted. Vaginal swabs were obtained and subjected to pH test, whiff test and were examined for presence of clue cells under the microscope.

In our study 28% of primigravidae and 25.5% of multigravidae showed BV. In Darwish et al11 study, 47.1% multigravidae and 10% primigravidae had BV. In Hiller et al12 study BV is associated with primigravidae. In our study, out of 27 cases of BV, 11 were multigravidae and 16 were primigravidae, which is not significant statistically.

In our study 22% cases of BV had white discharge per vaginum. In Darwish et al11 study also only 49% cases had white discharge. Less than half the 57 cases had white discharge per vaginum. This suggests that it is not a significant criterion in diagnosing BV.

In our study 28 cases with Vaginal fluid pH >4.5 were found to have BV. All the bacterial vaginosis cases had vaginal pH >4.5. The patient with pH >4.5 who were negative for BV would have vaginal infection other than Bacteriodes species. In Darwish et al11 study negative predictive value and sensitivity for pH >4.5 were 100%. In our study also negative predictive value of vaginal fluid pH > 4.5 is 100%.
In our study 27 cases were found to have positive whiff test. 20 cases had clue cells on microscope examination. In Darwish et al. study whiff test is positive in 75.6% and 83.3% had clue cells. Despite being positive in 75.6% in this study, the detection of fishy odour on the addition of 10% KOH is not an ideal diagnostic test because of personal variation in perceiving the odour. In our study, Whiff test was positive in 27% of cases in all groups.

Detection of clue cells in west mount is the most sensitive and specific test for the diagnosis of BV, but it may be mistaken for debris, degeneration of cells or epithelial cells to which lactobacilli are adherent.

Eschenbach et al. studied that 49% women with preterm delivery or low birth weight (LBW) babies were found to be associated with bacterial vaginosis. They diagnosed bacterial vaginosis by gas liquid chromatography. Paul et al. found that there is an association of bacterial vaginosis and preterm birth. He diagnosed bacterial vaginosis by Gram stain of vaginal smear, wet mount and 10% KOH preparation.

Table 8: Prevalence of BV in various studies

<table>
<thead>
<tr>
<th>S.No</th>
<th>Study</th>
<th>Prevalence of BV</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Atef M Darwish et al.</td>
<td>33.3%</td>
</tr>
<tr>
<td>2.</td>
<td>Sharon L Hillier et al.</td>
<td>16%</td>
</tr>
<tr>
<td>3.</td>
<td>Saharan SP et al.</td>
<td>37.5%</td>
</tr>
<tr>
<td>4.</td>
<td>Nelofer Saleem et al.</td>
<td>55.38%</td>
</tr>
<tr>
<td>5.</td>
<td>Desai Veena et al.</td>
<td>18.7%</td>
</tr>
<tr>
<td>6.</td>
<td>Kumar Aruna et al.</td>
<td>30.35%</td>
</tr>
<tr>
<td>7.</td>
<td>Our study</td>
<td>27%</td>
</tr>
</tbody>
</table>

The preterm prediction study done by Meis PJ et al. evaluated the association of BV, trichomonas and monilial vaginitis with spontaneous preterm birth at <35 weeks. They studied at 24 and 28 weeks of gestation. By gram stain of vaginal smear west mount and 10% KOH preparation to detect vaginal infections. The rates of detected infections at 24 and 28 weeks respectively were, BV 23.4% and 19.4%, trichomonas 3.3% and 2.7%, and monilia 21.1% and 19.5%. Presence of BV at 28 weeks gestation is associated with an increased risk of spontaneous preterm birth.

Conclusion

The strong correlation between presence of Bacterial Vaginosis and the incidence of threatened preterm and preterm labour is established. Amsel’s criteria is simple, inexpensive, easily reproducible method of diagnosing BV and can be used on a mass scale. Vaginal fluid pH >4.5 can be taken as an initial criteria to screen Bacterial vaginosis. A positive whiff test with pH >4.5 verify the diagnosis. Only if the whiff test is negative microscopy has to be performed in order to screen for clue cells. By diagnosing and treating bacterial vaginosis in threatened preterm labour, preterm deliveries can be prevented to a certain extent, if not atleast in order to buy time for the inclusion of steroids which will decrease the perinatal morbidity and mortality by decreasing Respiratory Distress Syndrome and intraventricular haemorrhage.

References