

Prevalence of Bacterial Vaginosis in Preterm Labour and Its Fetal and Maternal Outcome

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ABSTRACT

Introduction. Preterm births accounts for about 11.1 % of births worldwide. Infection accounts for 20% – 40% of these cases and Bacterial vaginosis has been found in 15 % of preterm labor group.

Objective. To find the prevalence of Bacterial vaginosis among the cases who presented with preterm labour and to determine its maternal and neonatal outcome.

Methodology. A prospective cross- sectional analytical study on 100 preterm labour cases conducted over a year in the obstetric ward in Manipal Teaching Hospital. Three high vaginal swabs from posterior fornix were taken and tested in Microbiology laboratory. The maternal and neonatal outcome of women with preterm labour due to bacterial vaginosis were noted. The data were analyzed using SPSS 21. Chi² test was used for comparing maternal and neonatal complications positive or negative with Bacterial Vaginosis.

Results. In the study period, 584 women had preterm deliveries. The total deliveries during that period was 2,531. The prevalence of Bacterial vaginosis in PTB was found to be 17 %.The accuracy of Gram stain was found to be 90% (sensitivity-57%, specificity-92%, PPV-36%, NPV-96%). The accuracy of wet mount was 90 % (sensitivity-42.85%, specificity-93.5%, PPV-33.3%, NPV-95.6%). There were increased cases of chorioamnionitis among women who were BV positive (p-value=0.02). Newborn born to women with BV positive had increased chance of poor Apgar score at 5 minutes (p-value <0.05).

Conclusion. Maternal and neonatal complications were more common in women with preterm labour associated with Bacterial Vaginosis. Gram stain and wet mount are fairly accurate and can be used as an adjunct to culture for the diagnosis of Bacterial Vaginosis.

Keywords: Bacterial vaginosis, Chorioamnionitis, Preterm labour, Preterm prelabour rupture of membranes, Neonatal sepsis

INTRODUCTION

Preterm labor (PTL) is defined by the World Health Organization as the onset of labor after the gestation of viability (20 to 28 week, depending on definition) and before 37 completed weeks. In our study, 28 weeks was taken as age of viability as there is increased chance

of survival beyond this gestational age in low income countries like ours.¹ which is the leading cause of perinatal morbidity and mortality in developed countries. Infants are born preterm at less than 37 weeks' gestational age after: (1

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Preterm birth (PTB) accounts for 75% of neonatal deaths and 50% of long term neonatal morbidity.¹ which is the leading cause of perinatal morbidity and mortality in developed countries. Infants are born preterm at less than 37 weeks' gestational age after: (1 About one-fifth of babies born under 32 weeks cannot survive the first year compared with 1% born at 33 to 36 weeks which is further decreased to 0.3% at term.² Severe morbidity like intra-ventricular hemorrhage, Respiratory distress syndrome, bronchopulmonary dysplasia, necrotizing enterocolitis, is common at 30 to 32 weeks. The common maternal complications of PTL are chorioamnionitis and postpartum endometritis.³

Almost 20 to 40% of PTL and 25% of PTB are attributable to infection³ with bacterial colonization rates as high as 79% for birth at 23 weeks, declining to 11% at 31 to 34 weeks.⁴ Bacterial Vaginosis (BV) amongst other infections has significant association with PTL. The prevalence of BV in PTL is found to be 11 to 26 % and has more adverse maternal and neonatal outcome.⁵⁻⁷ BV is diagnosed clinically by appearance of greysish-white vaginal discharge. In laboratory, it is diagnosed by Amsel's criteria , Gram staining and vaginal cultures .⁸

Although there have been many studies on association of BV with PTL, this study is conducted because it has not been frequently studied in our part of the world. Hence, this study is conducted with the aim to find the prevalence of Bacterial vaginosis among the cases who presented with preterm labour and to determine its maternal and neonatal outcome.

METHODOLOGY

This study was conducted in Manipal Teaching Hospital, Department of Obstetrics and Gynecology. Women diagnosed as preterm labour and meeting the inclusion criteria constituted the study population. It was a prospective cross – sectional study with a sample size of 100. It was collected over one year duration from January to December 2018 . There were three laboratory tests for the diagnosis of BV. Among the three laboratory tests for BV, if any one of the test was found to be positive, the patient was considered as positive for BV.

Inclusion criteria: Patients admitted in Antenatal Care ward with gestational age between 28 to 36+6 weeks

1. Singleton pregnancy
2. Patients with preterm labor, defined by contractions occurring at regular intervals, four or more in 20 minutes or eight or more in 1 hour, and each should

last more than 40 seconds

3. Cervical dilatation >1 cm and effacement >80%
4. Patients willing to sign a consent form

Exclusion criteria:

1. Multiple pregnancy
2. Previous history of antepartum hemorrhage
3. Severe anemia
4. Hypertensive disorders in pregnancy
5. Known medical disorders in pregnancy
6. Structural and functional abnormalities of the uterus
7. Urinary tract infection
8. Polyhydramnios
9. Intrauterine fetal death

Pregnant women meeting the inclusion criteria were enrolled in the study after informed consent and detailed history was taken. Specimens were collected using three sterile cotton swabs from the posterior vaginal fornix. Two were kept in a sterile glass tube and third one was transported on protease peptone No.3 medium. They were immediately transported to microbiology laboratory. Following per speculum examination, per vaginal examination was done. Of the three swabs, one was used for wet mount, second swab was used for Gram stain and third swab which was transported in protease peptone medium was used for culture. All the mothers were followed up. Their gestational age at delivery was noted. The neonates after delivery were followed till discharge. All these information were filled up in predesigned proforma.

Data were first entered in MS - excel and later transferred to SPSS software version 21. Chi² test was used for comparing maternal and neonatal complications in women with PTL who were BV positive and negative. p – value <0.05 was considered statistically significant. McNemar tests was used to calculate the Sensitivity , Specificity , Positive Predictive value and Negative Predictive value of wet mount and Gram stain taking culture as gold standard.

RESULTS

Our total sample size was 100 which was collected within a period of a year, amongst which 17 patients were positive for bacterial vaginosis. The prevalence of Bacterial vaginosis was found to be 17% in our study. There were 11 cases who were positive for BV according to Gram stain results. Amongst these, four were positive for both Gram stain and culture. The sensitivity of Gram stain was 57% and specificity was 92%. The PPV and NPV were 36% and 96% respectively. The accuracy of Gram stain was found to be 90%.

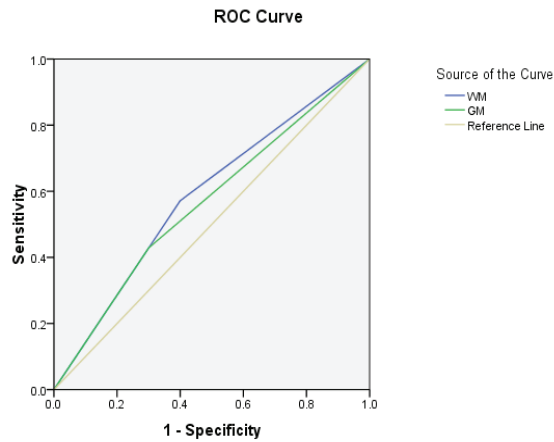


Fig. 1. ROC curve showing sensitivity and specificity of GM stain and wet mount

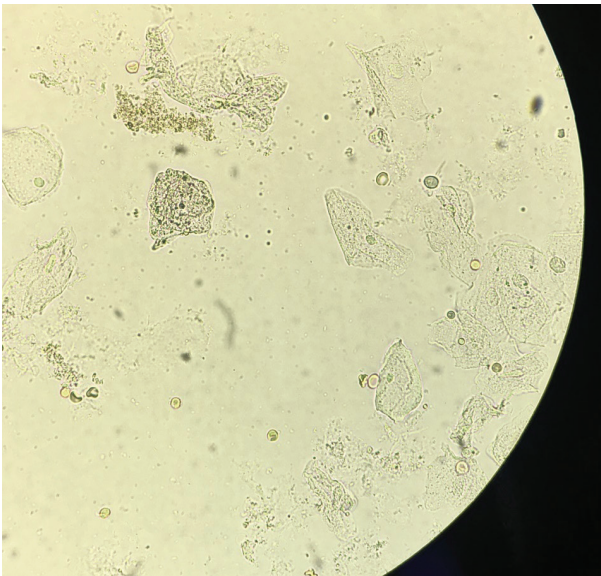


Fig 2. Clue cells on wet mount

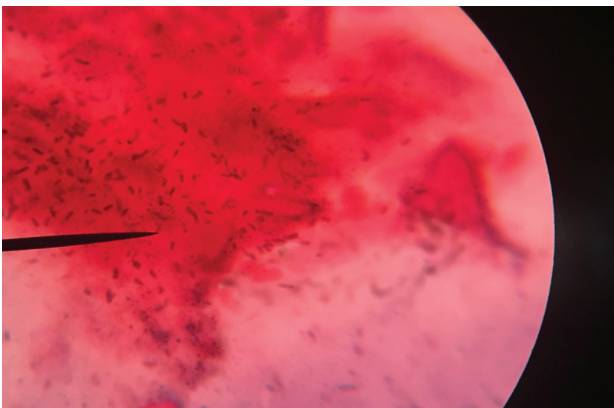


Fig 3. Coccobacilli in Gram stain

Taking culture as the gold standard the sensitivity of wet mount for clue cells was 42.85 % in this study .The specificity of wet mount was found to be 93.5 %.The PPV for wet mount was 33.3%and NPV was 95.6%. The accuracy of wet mount was 90 %.

The maternal complications of women with PTL due to BV is presented in table 1.

Table 1: Types of Maternal Complications in Patients with and without Bacterial Vaginosis

COMPLICATIONS	BV POSITIVE (n=17)	BV NEGATIVE (n=83)	P value
PPROM	11 (64.70%)	42 (50.60%)	0.09
PTB	14 (82.35%)	59 (71.08%)	0.13
Chorioamnionitis	2(11.74%)	1 (1.20%)	0.02
Puerperal Pyrexia	0	2(2.40%)	0.51

**Figures in the parentheses indicate percentage.*

Maternal complications were more in BV positive group than in BV negative group but this was not statistically significant. In patients with PTL who were positive for BV, PPROM occurred in 65% of the cases and PTB in 83% of cases though the results were statistically not significant. Nearly 12% of cases who were BV positive developed chorioamnionitis whereas only 1.2 % of BV negative cases had chorioamnionitis; this difference noted was found to be statistically significant.

The neonatal outcome of the women with PTL due to BV is presented in the table no 2.

Table 2 :Neonatal Parameters in Bacterial Vaginosis positive and negative Patients				
Neonatal Parameters		BV positive	BV negative	p value
Birth weight	>2.5 kg	2 (11.76%)	29(34.93%)	0.26
	1.5-2.49 kg	13 (76.47%)	46(55.42%)	
	< 1.5 kg	2 (11.76%)	8(9.63%)	
Apgar Score at 1 minute	7-10	9(52.94%)	10 (58.82%)	0.108
	4-6	7(41.17%)	6 (35.29%)	
	0-3	1 (5.88%)	1 (5.88%)	
Apgar core at 5 minute	7-10	10 (58.82%)	79(95.10%)	<0.05
	4-6	7(41.17%)	2(2.40%)	
	0-3	0	2(2.40%)	
POG at delivery	28 – 33+ 6 weeks	2 (11.76%)	23(27.7%)	0.26
	34 – 36+6 weeks	13 (76.47%)	35(42.16%)	
	37 + weeks	2 (11.76%)	25(30.12%)	
NICU Admission	Respiratory Distress Syndrome	4 (44.44%)	15(38.46%)	0.04
	Neonatal sepsis	4 (44.44%)	2(5.12%)	
	Nectrotisingenterocolitis	1 (11.11%)	2(5.12%)	
	Neonatal Hyperbilirubinemia	0	20(51.28%)	

*Figures in the parentheses indicate percentage.

Neonatal complications were more common in patients who were BV positive (53%) than in those who were BV negative (23%) which was significant statistically. Low birth weight in BV positive group was 76.47 % and 55.42 % in BV negative group. There was 11.76% of very low birth weight neonate in BV positive and 9.6 % in BV negative group. Analysis of Apgar score at 5 minutes showed that 41.17% had poor Apgar in BV positive whereas only 4.9 % of neonates from BV negative group had poor Apgar score and this was statistically significant. PTB were more common in BV positive patients (87%) than in those who were BV negative (66.5%). Among the 17 newborns from BV positive group, 9 (53%) required admission in Neonatal Intensive care Unit (NICU) whereas only 23% of neonates in BV negative groups were admitted in NICU, this difference was found to be statistically significant. There were no cases of neonatal mortality during in this study.

DISCUSSION

In our study , the prevalence of bacterial vaginosis was 17 % among the cases of preterm labour, which was similar to another study conducted by Aduloji OP et al. ⁵ The prevalence of BV was 13 % in a study conducted by Rohit C et al. ⁹ and 26 % in a study conducted by Thompson JL et al . ¹⁰

In our study, majority of the patients (88.2%) who were BV positive developed maternal complications but only 78.3% of those patients who were BV negative developed complications. Different complications like PTB, PPROM and chorioamnionitis is occurred in patients with PTL. Maternal complications like chorioamnionitis was present in 11.76 % of the patients positive with BV. Only 1.2 % of the patients negative with BV had chorioamnionitis in this study and this difference was found to be significant statistically. Dingens AS et.al also found the increased incidence of chorioamnionitis in preterm labour group who were BV positive than BV negative. ¹¹

Preterm birth is a major factor in neonatal outcome. Increased rate of preterm birth in BV positive group in our study could have resulted in increased association of that group with neonatal complications. In our study, 53% of the neonates suffered complications and required admission in the NICU for various indications. Likewise, in a prospective cohort study, among BV exposed and BV unexposed cases, BV-exposure was associated with an increased risk for NICU admissions with relative risk of 1.24. Even among full-term infants, BV was associated with an increased risk of NICU admission.¹¹

Various indications for NICU admission in our study were Respiratory Distress Syndrome (RDS), neonatal sepsis and necrotizing enterocolitis. The indications for NICU admission in another study were more or less similar.¹¹ In our study, the number of neonates admitted with a diagnosis of RDS was 44.44 % in BV positive which was higher compared to that of BV negative group comprising of 38.46 %. This difference was statistically significant. Similar finding was obtained in a study done by Laxmi U et al. where the incidence of RDS was higher in BV positive mothers compared to in BV negative mothers.¹ with or without bacterial vaginosis (BV² The reason for the increase incidence of RDS can be attributed to complications of prematurity itself. The number of neonates admitted with a diagnosis of NEC was 2.2 % in BV positive group compared to 1.4 % in BV negative group in our study. NEC is also a known complication of prematurity.

In our study, among 17 patients who were diagnosed as BV positive, 76.47 % were low birth weight whereas 55.42 % of neonates from BV negative group had low birthweight. Similar results were documented by Hiller SL et al. where an increased rate of low-birth-weight infants was common among women colonized by BV.¹³

A 5-minute Apgar score of 0 to 3 correlates with neonatal mortality in large populations but does not predict individual future neurologic dysfunction. Population studies have uniformly reassured that most infants with low Apgar scores will not develop cerebral palsy. However, a low 5-minute Apgar score clearly confers an increased relative risk of cerebral palsy, reported to be as high as 20-fold to 100-fold over that of infants with a 5-minute Apgar score of 7 to 10. Although individual risk varies, the population risk of poor neurologic outcomes also increases when the Apgar score is 3 or less at 10 minutes, 15 minutes, and 20 minutes.¹⁴ In our study, five - minute Apgar was lower in the neonates born to women with PTB positive for BV. However, as the long term follow up was not done, the outcome of the babies could not be done in our study.

Various tests can be done and diagnostic criteria can be used for diagnosis of BV. In our study, wet mount for clue cells, Gram stain for presence of Mobiluncus/ Gardnerella using Hay/Ison criteria and culture were done to test for

BV. The case was considered positive for BV if any one of the test was positive. Presence of Gram variable coccobacilli (*Gardnerella/Mobiluncus*) may at times mimic normal vaginal microflora and hence may not be confirmatory of BV. Hence, in order to check for accuracy of the test, culture was taken as the gold standard.

For analyzing Gram stain, various criteria can be used. Although Nugent's criteria in Gram stain has been regarded as the gold standard, the field size of the microscope has a bearing on the results which is another issue of concern.⁸ Moreover, the score intervals are very narrow, with a difference of only a few bacteria and the real number of Lactobacillus morphotypes recognized may be influenced by the variability of the methods. The homogeneity and thickness of the sample may vary in different ways of spreading the sample on the glass slide. Hay / Ison criteria was preferable alternative to this and therefore Hay/Ison criteria was used in our study to interpret the Gram stain results.⁹

The sensitivity and specificity of Gram stain was 57% and 92% in this study. The PPV and NPV was 36% and 96% respectively. Since the specificity and NPV of this test was high, if this test is negative, there is high probability that the subjects truly do not have the disease. It is thus a good test in ruling out the disease. The accuracy of Gram stain was 90 %, therefore, there is less chance of systematic error or bias in a test as per our study.

Thomason JL et al. conducted a study on statistical evaluation of diagnostic criteria for bacterial vaginosis where they observed the Gram stain criteria (bacterial morphologic types) to be less accurate predictor of the disease with a sensitivity of 97.0%, specificity of 66.2%, positive predictive value of 57.2% and negative predictive value of 97.9%.¹⁰

In our study, taking culture as the gold standard, the sensitivity of wetmount for clue cells was 42.85 %. The specificity was found to be 93.5 %. The PPV for wet mount was 33.3% and NPV was 95.6%. The accuracy of wet mount was 90 %.

However, in a study done by Thomason JL et al, wet mount for clue cells was found out to be the single most reliable indicator of bacterial vaginosis with high sensitivity.¹⁰ difference may be due to different gold standard used in different studies. Amsel's criteria was used as the gold standard in these studies whereas culture was the gold standard in our study, resulting in this difference in results.

High specificity of wet mount in our study taking culture as gold standard indicate that the absence of clue cells in the vaginal swab rules out the presence of BV. Therefore the absence of clue cells could give good prediction of absence of BV.

This study had small sample size and was of shorter duration. The study population may not be representative of the larger community. The results of the study can therefore not be generalized. Larger scale studies in large sample of patients would be more conclusive.

CONCLUSION

The prevalence of preterm births was 23%. The prevalence of Bacterial Vaginosis in preterm labour was 17%. There was increased maternal and neonatal complications in women who had preterm labour associated with bacterial vaginosis. Gram stain using Hay/Ison criteria and wet mount for clue cells were accurate test and were more likely to rule out the presence of disease taking culture for *Gardnerella* and *Mobiluncus* as the gold standard.

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