

Original Research Article

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Bacterial Vaginosis: One of the Causes of Preterm Labor

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ABSTRACT

The aim of this study was to evaluate the role of bacterial vaginosis in preterm labor. Preterm labor has increasing evidence of infectious causes especially ascending infections from lower genital tract. Preterm delivery is a leading cause of neonatal morbidity and mortality. Laboratory evaluation and clinical assessment were performed for 100 female patients. Bacteriological diagnosis were done using direct Gram staining for clue cells, bacterial culture and amsel's criteria, scoring was done by Nugent score system. We carried our study on 100 pregnant women, divided into two groups. First group delivered preterm and other group at full term. 33 patients out of 50 pts with preterm group had BV (66%) while 24 patients out of 50 patients had BV (48%) in full term group, so the incidence of BV was significantly more in preterm group. The commonest isolated pathogen was *G. vaginalis*, followed by beta hemolytic streptococci and staph aureus. It was concluded that bacterial vaginosis is considered as one of the most common vaginal infection in pregnancy and it have role in preterm labor.

Keywords

Vaginitis,
Gardenella
Vaginalis,
Preterm labor.

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Introduction

Preterm labor is presence of contraction of sufficient strength and frequency to effect progressive effacement and dilatation of cervix between 20 and 37 weeks of gestation. Preterm labor has increasing evidence of infectious causes especially ascending infections from lower genital tract. Bacterial vaginosis has been associated with late miscarriage, preterm labor, premature rupture of membranes and chorioamnionitis (6).

One of the leading causes of neonatal morbidity and mortality is Preterm delivery. Preterm infant with a birth weight below 2000

grams was in the main focus (17). Preterm labor represents 9.6% of all births worldwide. In which 11 million of these preterm were in Africa and Asia while about 0.5 million occurred in each of Europe and North America while 0.9 million occurred in Latin America (2).

Strongly association was found between intrauterine infection including BV and spontaneous preterm labor that occur before 35 week of gestation and before 32 week in particular have been (11).

Although 30 – 40% of Bacterial vaginosis (B.V.) are asymptomatic it remains the commonest cause of lower genital tract infection among women in child bearing period. No single microorganism is detected in all women with B. V. *Gardnerella vaginalis* (G.V), *Bacteroids* species and *Mycoplasma hominis* (*M. hominis*) were detected in most studies (12).

The most common cause of vaginal discharge in women attending gynecological clinics is bacterial vaginosis (5). Infection ascending from the lower genital tract in pregnancy is associated with preterm labor (23).

Bacterial vaginosis is diagnosed in up to 23% of pregnant women. It is linked to pregnancy complications including miscarriage, preterm labour, pre-mature rupture of membrane, amniotic fluid infection and postpartum infection (21).

Preterm labor is commonly associated with intrauterine infection and inflammation and at least 40% of all preterm birth delivery has been estimated to occur with mothers who have intrauterine infection. Colonization by *Trichomonus vaginalis*, *Bacteroids* species or *Urea plasmaurelyticum* had a statically increased risk of subsequent preterm birth, preterm delivery and PROM (20).

The risk of preterm labor and PROM increased when associated with bacterial vaginosis or vaginal pH > 5 at 16 – 36 weeks of gestation (1).

Screening and treatment of bacterial vaginosis in high risk women with previous preterm birth may prevent these women from having another preterm birth (3).

A detailed explanation about the nature of the early symptoms and signs of excessive uterine activity should be told to the patient. Treating

infections in preterm labor is an important preventive measure.

Intra-amniotic infection may be developed in women with bacterial vaginosis. So vaginitis or chronic cervicitis should be treated immediately (13).

The aim of this study is to evaluate the role of bacterial vaginosis in preterm labor.

Materials and Methods

The present study is a case control study of 100 pregnant women including 50 women in preterm labor (study group) and 50 women in full term labor (control group). Their ages ranged from 20 to 35 years. They were collected from gynecological and obstetric department of the Mansoura Faculty of Medicine, Egypt. All patients were enrolled in the study after informed consent. The study was approved by the ethical committee of the Faculty of Medicine, Mansoura University.

Inclusion criteria

The study group: females with preterm labor in pregnant females 20-36 weeks of gestation by last menstrual period or ultrasound guided.

The control group: full term pregnant women in labor

Exclusion criteria

Women less than 20 weeks of gestation

Overt vaginal infection caused by candida or chlamydia.

Placental abruption and placenta previa and other causes of antepartum hemorrhage.

Associated septic focus.

Congenital anomalies in the uterus.

Incompetence of the cervix and premature

Rupture of membrane.

Multiple pregnancies and polyhydramnios.

For each woman a full history, general and local examinations were done. Vaginal speculum was inserted in the vagina at admission without lubrication and the appearance of vaginal discharge was noted. Vaginal swabs were collected from the posterior vaginal fornix with sterile swabs for direct smear and cultures.

Vaginal smears were heat fixed, gram stained, examined for presence of clue cells and Nugent's scoring. On microscopic examination of the discharge, demonstration of 3 of the following 4 Amsel criteria is considered necessary to diagnose BV most accurately:

Demonstration of clue cells on a saline smear (the most specific diagnostic criterion)

A pH greater than 4.5 (up to 90% of patients)

Characteristic thin, gray, and homogeneous discharge

Positive whiff test (up to 70% of patients) (27): was done near the patient putting several drops of KOH 10% solution on a vaginal discharge sample and to see if a strong fishy odor is produced.

Nugent's criteria used to quantify or grade bacteria via Gram staining of vaginal samples. These criteria evaluated the following 3 types of bacteria and assign scores to each *Lactobacillus* (score, 0-4), *Bacteroides/Gardnerella* (score, 0-4), *Mobiluncus* (score, 0-2). Total scores are calculated and interpreted as follows, 0-3: Normal, 4-6: Intermediate bacterial count, 7-10: BV.

Vaginal swabs were inoculated at the lab onto: (i) Columbia blood agar: Aerobically

incubated at 37°C for 24 to 48 hours for isolation of *Gardnerella vaginalis* which were identified by beta hemolytic colonies. Gram negative pleomorphic coccobacilli were seen by gram stained film. *Gardnerella* was urease positive after performing catalase test, oxidase test and urease tests for more confirmation. (ii) Blood agar plate: Incubated at 37°C for 24 to 48 hours in 10-15% CO₂ for isolation of lactobacillus which appeared as pinpoint α hemolysis colony and as large gram positive rods by gram stain film. (iii) Another two blood agar plates: One is incubated anaerobically at 37°C for 7-9 days for isolation of anaerobic bacteria (gram negative *Bacteroids*) and the other incubated aerobically at 37°C for 24 to 48 hours for isolation of gram positive cocci (coagulase positive staphylococci and beta hemolytic *Streptococci*). (iv) MacConkay's agar plate: aerobically incubated at 37°C for 24 to 48 hours for isolation of gram negative bacilli. (v) Sabouraud's agar plates, which were incubated microaerophilically at 37°C for 24 to 48 hours to isolate candida confirmed by gram stain from colonies after growth and germ tube test.

Statistical analysis

These data were analyzed using SPSS version 16. Inter-group comparison of categorical data was performed by using chi-square test and Z test for proportion.

Results and Discussion

Hundred pregnant women were included in our study then were divided into two groups of patients, first is preterm and second is full term delivered. 33 patients out of 50 pts in preterm group had BV (66%) while 24 patients out of 50 patients in full term group had BV (48%) so the incidence of BV is significantly more in first group than second group (Table 1).

BV symptoms are fishy odor, itching and sometimes dysuria. In preterm women only 23 patients out of 33 cases with BV complained of BV symptoms representing 69.7% while only 9 patients out of 24 cases with BV in full term women complained of symptoms (Table 2). It had clinical and statistically significant by using Z test at level of 1.67 but by chi square test it nearly significant > 0.7 because of small sample size.

By using the Nugent score (18), preterm group were divided into 14 (28%) normal women with a (0-3) score, 3(6%) women with intermediate a (4-6) score and 33 (66%) women having bacterial vaginosis with a (7-10) score, while in 50 women of (full term group) 20(40%) women were normal, 6 (12%) women were intermediate, and 24 (48%) women having bacterial vaginosis (Table 4).

Diagnosis of BV was done by Amsel's criteria. All 4 criteria were found in 12 patients out of 33 in preterm group, (vaginal $\text{pH} \geq 4.5$, thin discharge and fishy odor) were found in 3 patients only, ($\text{pH} > 4.5$, fishy, odor and clue cell) were found in 12 patients and ($\text{pH} \geq 4.5$, thin discharge clue cell) were found in 6 patients. While all 4 criteria were found in 10 patients, ($\text{pH} \geq 4.5$ thin discharge and fishy odor) were positive in 2 patients, ($\text{pH} > 4.5$, fishy odor and clue cell) were positive in 8 patients and ($\text{pH} > 4.5$, vaginal discharge and clue cell) were positive in 4 patients in full term group (Table 5).

The incidence of preterm labor is more in early G.A in patients who complain from B.V. 75% Preterm labor in G.A > 20 week – 24 week, (42%) 25 week – 28 week, (37%) 29 week – 32 week and (3.0%) 33 week – ≤ 37 week and because of small number of cases this was not statistically confirmed (Table 6). Bacterial vaginosis (BV) is one of the commonest cause of vaginal discharge caused

by imbalance in normal vaginal bacterial flora and reduced *Lactobacillus* and increase in *Gardenella*, *Mycoplasma* and anaerobic bacteria (24). The most prevalent lower genital tract infections in women and the most common cause of vaginal discharge in reproductive age is BV (12).

Preterm labor onset is initiated by multiple mechanisms including intrauterine infection, uterine over distention, retroplacental hematoma stress or other immunological mediated process. The treatment of preterm labor target to prevent uterine contraction (26), because it is the leading cause of neonatal morbidity and mortality (17).

Preterm group incidence of preterm labor in this study was 33 (66%) patients out of 50 patients which had BV (Table 1) and this agreed with the study by Shahgeibie *et al.*, (18). In Krauss *et al.*, (9) the exposure group was 136 women had BV and 397 pregnant women without BV as non-exposure group. Spontaneous preterm labor that occur before 35 week of gestation have been strongly associated with intrauterine infections including BV and this agree with our study.

In study done by Kalinka *et al.*, (8) the preterm labor rate was about 15.7% suffering from BV compared with 9.1% who didn't have bacterial vaginosis and it concluded that early diagnosis and treatment of BV could decrease preterm labor.

This study also agreed with Camarge *et al.*, (4) study that has detected the impact of treatment of bacterial vaginosis in preterm labor. In Brazilian 785 women were studied in 3 different groups of women were identified as 580 women without bacterial vaginosis and 134 women with BV treated during pregnancy and 71 with bacterial vaginosis. The frequency was 5.5% of preterm labor in patients without bacterial

vaginosis and 3.7% in pretreated from bacterial vaginosis during pregnancy and 22.5% in women with bacterial vaginosis. There's no significant difference in preterm labor, PROM or low birth weight group without BV and treated group. On other hand there was significant difference between treated and untreated group.

vaginalis was the commonest cause of bacterial vaginosis. Azarگون and Darvishezadeh, (1), agreed with this study. This cohort comprised 1223 pregnant women with gestational ages of 16 – 36 weeks admitted to Amir-Almomnein General Hospital in Semnan, Iran, from March 2002 through March 2003.

The commonest isolated pathogen in the present study, was *G. vaginalis*, followed by beta hemolytic streptococci and staph aureus (Table 3). Reports differ in pathogens associated with vaginal discharge and their prevalence. This comes in agreement with Maysaa *et al.*, (15) who reported that *G.*

Women with systemic diseases were excluded, also patients with placenta previa, abruption, uterine anomalies, incompetent cervix, twin pregnancies, preterm labor pain, prior use of antibiotics in the preceding two weeks or use of corticosteroid and to colytic agents during the current pregnancy.

Table.1 Incidence of BV in full term and preterm group

	Preterm women		Full term women		P value
	No	%	No	%	
Number of BV cases	33	66	24	48	< 0.05

Table.2 Symptomatic and asymptomatic cases with positive BV

	Positive BV cases in preterm gr (n = 33)		Positive BV cases in full term gr (n = 24)		P value
	No	%	No	%	
Symptomatic	23	69.7	9	37.5	> 0.05
Asymptomatic	10	30.3	15	62.5	> 0.05

Table.3 Frequency of isolated pathogens from patients

Isolated organism	Preterm group		Full term group	
	No	%	No	%
	33	66	24	48
<i>Gardenerella vaginalis</i>	14	42.4	11	45.8
B hemolytic Streptococci	7	21.2	5	20.8
<i>Staphylococcus aureus</i>	6	18.2	4	16.8
<i>E. coli</i>	2	6.1	2	8.3
<i>Candida albicans</i>	4	12.1	2	8.3

Table.4 Prevalence of BV in both groups (by using Nugent Score)

	Nugent Score					
	Normal (0-3)		Intermediate (4-6)		BV (7-10)	
	No	%	No	%	No	%
Preterm group	14	28	3	6	33	66
Full term	20	40	6	12	24	48

Table.5 Amsel's criteria in both study groups

Amsel's criteria	First preterm group		Second full term	
All 4 criteria	12	36%	10	41.6%
Vaginal pH > 4.5, thin discharge and fishy odour	3	10%	2	8%
pH > 4.5, fishy odour and clue cell	12	36%	8	34%
pH > 4.5, thin discharge and clue cell	6	18%	4	16%

Table.6 Incidence of BV in preterm group according to GA

Groups	N	BV	%	
≥ 20 week – 24 week	4	3	75%	
25 week – 28 week	7	5	71%	
29 week – 32 week	16	11	68%	
33 week – ≤ 37 week	23	14	60%	
Full term	24	48%	24	48%

No history of alcohol consumption or smoking was reported in their patients. Bacterial vaginosis was diagnosed if at least three of four Amsel's criteria are present. Tests were negative for BV at the first time, specimens were again obtained at 34 – 36 weeks of gestation or at the time of admission for labor. In 33% of patients with gestational age 16-20 week and 66% of patients at 37 weeks of gestational bacterial vaginosis was positive. 24% from those women who had BV develop preterm labor compared to 4.9% in uninfected women.

Our study didn't agree with the results of Vogel *et al.*, (25) it was a population based prospective cohort study to detect the effect of bacterial vaginosis on pregnancy. The participant was 2.927 pregnant women visiting Odense University Hospital Denmark. They excluded multiple gestations and antibiotic received 2 weeks ago. Each patient fill a questionnaire (dealt with socioeconomic state, medical, obstetric history and gestational age) at enrolment at 30 week and at delivery. The diagnosis of BV was done as defined by Amsel's criteria. They assessed the prevalence outcome (PTL, LBW) in this sample, 3.3 per 100 live births was the rate of

spontaneous onset of preterm labor but they didn't detect an increased risk of preterm labor with BV.

The present study didn't agree with the results of Povlsen *et al.*, (19) which was done on 484 pregnant women came to the maternity ward of Odense University Hospital. Only 10 patients had BV (12%) out of 84 women at preterm labor and only 60 patients have BV (15%) out of 400 women at full term, so in this study no effect of BV in preterm labor.

We used 2 methods in diagnosis of bacterial vaginosis in our study, which were Amsel's criteria (pH, fishy odour, presence of clued cell, excessive vaginal discharge) and gram stain by using scoring system as that both methods have the same results and this was in agreement with Hogan *et al.*, (7).

Modak *et al.*, (16) agreed with our study in which the bacterial vaginosis prevalence was 24% by Nugent's score (Table 4). In comparison, Amsel's criteria had 66.67% sensitivity and 94.74% specificity, 80% positive predictive value and 90% negative predictive value (Table 5). Presence of clue cells was best correlated with a positive diagnosis by

Nugent's score while the whiff test had the lowest correlation.

In this study it was found that about 32% (56%) women from 57 women who had BV complain of symptom of BV as vaginal discharge fishy odor or itching. And this gets in co ordinance with Sobel, (22), where 50% of patients had a symptomatic BV. The incidence of patient who had bacterial vaginosis in our study and complains from preterm labor is more in early gestational age preterm labor in GA \geq 20 week – 24 week (75%), 25 week – 28 week (71%), 29 week – 32 week (68%) and 33 week \leq 37 week (60%).

Uata, (24) study greed with our study (Table 6). Total 200 women attending the antenatal clinic, the incidence of preterm deliveries who had BV at 11-20 week was 14 patients from 62 patients (23%) while the incidence of patients who had BV in 21-30 week were 19 patients from 90 patient (20%) and incidence of patients who had BV from 31-37 week 8 patients from 43 patients (18%).

Laudanski *et al.*, (10) detected that infection is considered a causative factor of preterm labor in around 50% of cases. It is more likely associated with deliveries that occur before 30 weeks and is relatively rare in late preterm labor (34-36) weeks. 80% of women deliver before 30 weeks have evidence of bacterial infection in amniotic fluid compared to 30% only at 37 weeks and this agreed with our study. The present study not in agreement with study done by Mark *et al.*, (14) although BV was associated with an increased risk of preterm labor the gestational age at which BV was screened didn't influence the increase.

Bacterial vaginosis is one of the commonest causes of lower genital tract infection among women in child bearing period. No single microorganism is detected in all women with

B.V but a polymicrobial condition with deficient lactobacilli and overgrowth of mixed vaginal flora.

Bacterial vaginosis is not considered a serious disease by itself, but it appears to be a risk factor for the occurrence of many complications including preterm labor. Spontaneous preterm labor before 37 week accounts for 10% of all birth and 75% of neonatal death.

Preterm labour is a major cause of perinatal morbidity and mortality throughout the world, causing significant financial burden, and any program that reduces preterm labor will provide great benefits. This study has proved the association between bacterial vaginosis and preterm labor with its adverse outcome.

It was concluded that bacterial vaginosis is considered as one of the most common vaginal infection in pregnancy and it have role in preterm labor.

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