



Vaginal Microbiome study of preterm labour in tertiary care centre.

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Abstract: OBJECTIVE:

- 1) To study the incidence of preterm labor in tertiary care centre
- 2) To study the association of common genital infections with preterm birth and its antibiotic sensitivity.
- 3) To study maternal and fetal outcome in preterm delivery

METHOD: This prospective observational study was conducted for 2 years at Government Medical College and Hospital, Aurangabad. High vaginal swab of cases with preterm labour collected and studied for presence of vaginal microbiome. Preventive measures to halt preterm labor were taken. Maternal and fetal outcomes studied and analyzed to draw various conclusions.

RESULTS: The incidence of preterm birth found to be 12.18%. In the present study, the incidence of sterile high vaginal swab report was found to be 24.5% (49), maximum microbime found was *Staphylococcus aureus* 37% (74) and minimum *Pseudomonas* 2% (4). Neonatal complications were Respiratory distress syndrome followed by Sepsis, Intraventricular haemorrhage, Jaundice, Pneumonia and Necrotising enterocolitis with cases found in 26%, 16%, 14.5%, 14%, 6% and 4%, respectively. 60% neonates had no complications.

CONCLUSION: High risk screening, vaginal microbiome study and treatment can prevent preterm labour by 40% and reduce maternal and neonatal mortality.

Index Terms – Preterm birth, Gestational age, Vaginal microbiome, Intrauterine infections, Neonatal complications.

I. INTRODUCTION

Preterm labor (PTL) is defined as a birth between 20 weeks of gestation and 37 weeks of gestation. The American College of Obstetricians and Gynecologists states that diagnosis of preterm labor is generally based on the clinical criteria of regular uterine contractions accompanied by a change in cervical dilation, effacement, or both, or initial presentation with regular contractions and cervical dilation of at least 2 cm.¹

As per the World Health Organization 2012 reports, the magnitude of this problem was highest in India with around 3,519,100 preterm birth followed by China (1,172,300), Nigeria (773,600), Pakistan (748,100), Indonesia (675,700), and Bangladesh (424,100).²The global theme for World Prematurity Day 2019 is: Born too soon: Providing the right care, at the right time, in the right place.¹¹

Preterm labor may occur as a result of multiple factors including history of preterm labor, inflammation or infection, bacterial vaginosis, candidiasis, decidual bleeding, maternal chronic disease, uterine over-distension, multiple pregnancies, and premature rupture of membranes.^{7,8} Genetic factors, nutritional status, and lifestyle of women may also play role in preterm labor.⁷ Preterm birth remains a major public health problem.

Bacterial infection of the lower genital tract and uterus are the major causes of preterm birth and worldwide the most common microorganisms responsible for this are *Streptococcus agalactiae*, *Ureaplasma urealyticum*, *Mycoplasma hominis*, *Fusobacterium* species, and *Gardnerella vaginalis* (Goldenberg et al., 2000)²⁸. It has been shown that bacterial vaginosis caused by polymicrobial species is one of the main factors contributing to preterm birth in India. Few other studies suggested Group B *Streptococcus* as one of the major etiological agent of preterm birth. 12-16% Indian women are found to be colonized with Group B *Streptococcus* (Stoll and Schuchat, 1998)¹⁴ and 31% of the GBS-colonized women have a history of foetal losses and neonatal deaths (Kuruvilla et al., 1999)¹⁵. Being a Tertiary Care Institute and high case load of deliveries around 18,000 per year, we also get many cases of preterm labour, there was an attempt to study the vaginal microbiome associated with preterm labour and their outcome.

2. RESEARCH METHODOLOGY

INCLUSION CRITERIA:

- Consented for participation in study.
- Singleton pregnancy
- Preterm pregnancy >28 wks and <37 wks.
- Preterm premature rupture of membranes

EXCLUSION CRITERIA:

- Not willing to participate in study
- Pregnancy <28 wks and >37 wks
- Multiple pregnancy
- Induced labor in view of associated medical condition
- Abruptio placenta, placenta previa, stillbirth, IUFD.

This was a prospective observational study, conducted at Government Medical College, Aurangabad for the duration of 2 years. Due to limited resources of logistics for diagnostic test 200 cases of preterm labor studied. Cases of preterm labor selected in the labor room with cervical dilatation >2cm and <4cm, effacement of cervix >80%, regular uterine contractions with or without pain at least one in every 10 minutes.. Detailed clinical history and examination done. All basic investigations like complete blood count, blood group, HIV, HbsAg, VDRL checked. After informed consent high vaginal swab collected and appropriately transported in amies transport medium to microbiology laboratory. All the samples inoculated in blood agar and MacConkey agar plates and incubated for 24hours. Gram stain smear of the sample prepared and evaluated according to standard guidelines. Antibiotic sensitivity of the microbes performed by modified Kirby Bauer disc diffusion technique. The sensitive and resistant drugs recorded. Females were treated with antibiotics (cefotaxime or amoxicillin and clavulanic acid) according to hospital protocols. Preventive measures like improvement in nutrition, rest, hydration, psychological support, tocolysis, steroid, neuroprotection with magnesium sulphate was given. Maternal outcome studied in the form of post partum haemorrhage, sepsis, febrile illness. Fetal outcome studied in the form of low birth weight baby, NICU stay, sepsis or any other fetal complications. All collected data analyzed to draw various informative conclusions.

3. RESULTS AND DISCUSSION

3.a RESULTS

In 200 cases of Preterm Labor, studied at Government Medical College and Hospital, Aurangabad from October 2017 to October 2019, the following observations were made.

Incidence:

Among 18,461 live births from the year 2018, 2,115 were found to be preterm births. The incidence of preterm birth found to be 11.4%.

Among 18132 live births from the year 2019, 2,350 were found to be preterm births. The incidence of preterm birth found to be 12.96%. Mean incidence of preterm birth during study period was 12.18%.

Table no.1: Distribution of cases of preterm labor according to clinical category

Age Group in Years	No. Of Cases (n=200)	Percentage (%)
18- 20	41	20.5
21-25	91	45.5
26-30	56	23
Parity		
Primigravida	60	30
Secondgravida	76	38
Thirdgravida	48	24

Fourthgravida	16	8
History of preterm birth		
Present	38	19
Absent	162	81
History of abortion		
No abortion	167	83.5
One abortion	29	14.5
Two abortions	4	2

Table no 1 shows that age group 21-25 (45.5%) had the maximum number of preterm cases whereas the least number was found above the age 30 (12%). Nevertheless percentage of preterm labor in multigravida (70%) was found higher as compared to primigravida (30%).

38 cases(19%)had history of previous preterm labor. 33 cases(16.5%)had history of abortion.

Table no2: Distribution of cases of preterm labor according to Gestational Age on admission

Gestational Age	No. of Cases n=200	Percentage (%)	Delivered preterm	Delivered term
28.1-30 weeks	17	8.5	16(8%)	1(0.5%)
30.1 -32 weeks	25	12.5	23(11.5%)	2(1%)
32.1-34 weeks	55	27.5	27(13.5%)	28(14%)
34.1-37 weeks	103	51.5	23(11.5%)	80(40%)

Table 2 represents the distribution of cases of preterm labor according to GA. The highest incidence of preterm labor on admission found in 34.1-37 weeks was 103 (51.5%). Out of which 23 (11.5%) cases delivered preterm and 80(40%) cases delivered at term.

Table no.3: Distribution of cases of preterm labor according to High Vaginal Swab report

HVS report	No of cases(n=200)	Percentage
Staph aureus	74	37
Sterile	49	24.5
Strepto pyogenes	29	14.5
Klebsiella	16	8
Acenetobacter	12	6
Enterobacter	10	5
E. Coli	6	3
Pseudomonas	4	2

From the above table 3, the incidence of sterile HVS report found 49 (24.5%), maximum microbiome found was Staph aureus 74 (37%) and minimum Pseudomonas 4 (2%).

Table no.4: Distribution of cases of preterm labor according to Microbial Sensitivity

Row Labels	CEFOXITIN	ERYTHROMYCIN	LINEZOLID	OFLOXACIN	CLINDAMYCIN	AMPICILIN	AMIKACIN	GENTAMYCIN	IMPENEM	PIRZO	TRIMETHOPRIM/SULFAMETHOXAZOLE	BACTRACIN
STAPH AUREUS	49	2	72	1	54	29	11	64	21	24	17	0
STREPTOPYOGENES	0	27	22	0	4	8	1	2	1	3	0	25
KLEBSIELLA	3	0	0	9	1	0	1	15	16	4	0	0
ACENETOBACTER	0	0	0	9	0	0	12	1	12	12	0	0

ENTERO BACTER	0	0	0	1	0	0		10	10	10	0	0
E.COLI	0	0	0	0	0	0	6	5	6	6	0	0
PSEUDO MONAS	0	0	0	4	0	0	0	4	4	4	0	0
STAPH CONS	1	0	1	0	1	1	0	1	0	0	1	0
STERILE	0	0	0	0	0	0	0	0	0	0	0	0

Table 4 shows the distribution of cases of preterm labor according to microbial sensitivity to the wide spectrum of antibiotics administered. From the above table, the sensitivity of Staphalococcus aureus is maximum for drugs like cefoxitin, clindamycin, gentamicin, ampicilin, piperacilin tazobactum, imipenum whereas coagulase negative staphylococcus aureus has the least sensitivity towards drug.

Table no.5: Distribution of cases of preterm labor according to Maternal and fetal Complications

Maternal Complications	No of Cases(n>200)	Percentage (%)
Fever	92	46
Breast Engorgement	60	30
UTI	57	26.5
PPH	10	5
Sepsis	8	4
Wound Infection	2	1
No complications	90	45
Neonatal Complications		
RDS	52	26
Sepsis	32	16
IVH	29	14.5
Jaundice	28	14
Pneumonia	12	6

NEC	8	4
No complications	120	60

Table no. 5 shows the cases of preterm labor with maternal and fetal complications out of which maximum cases had fever 92 (46%) and minimum cases had Wound infection 2 (1%). 45% cases were delivered uneventfully. From the above listed fetal complications the highest incidence of RDS was observed followed by Sepsis, IVH, Jaundice, Pneumonia and NEC with cases found 52 (26%), 32 (16%), 29 (14.5%), 28 (14%), 12 (6%) and 8 (4%), respectively. 60% of the delivered neonates had no complications.

3.b DISCUSSION

Of the enrolled patients in the present study maximum were from age group 21-25 yrs of preterm cases whereas the least number was found above the age 30 yrs. Similar study was performed by Chawanpaiboon S. et al⁵ and Achari et al²⁷ with a mean maternal age group of 25- 30 yrs.

The present study shows the preterm labor according to ANC registration among which 106 (53%) were unbooked and 94 (47%) were booked. Antenatal registration helps in detecting high risk factors and preventing PTL. Also a study proposed by Goldenberg, 2000²⁸ shows that Antenatal registration helps in detecting high risk factors and only 14.6% had registered as compared to 38% in controls.

In present study, there is increasing percentage of preterm labor in multigravida (35%) as compared to primigravida (15%) as previous history of preterm delivery is associated with 2.5 fold risk of preterm labor in next pregnancy²⁶. In a study by Jean Frederick and Anderson et al, 1976²⁹ the parity had no effect on the incidence of spontaneous preterm births whereas Carrhill and Hall et al²⁰ found there is 15% chance of next preterm delivery.

From the study, out 200 patients, 19% patients had a history of Previous Preterm Birth, 16.5% patients had a history of Abortion; whereas 35 patients had a history of both whereas in a study proposed by Carhill and Hall et al²⁰ study shows that there is significant number of previous preterm birth i.e. 75% patients. And Papaevangelou G et al²⁵ found that the incidence of prematurity is more than two fold in women with one or more induced or spontaneous abortions when compared with those who had no previous abortions.

Among 200 patients the incidence of urban residence found maximum 111 (55.5%) and least found in rural 89 (44.5%). WHO 2012 reports² shows that magnitude of this problem was highest in India with around 3,519,100 PTBs, followed by China, Nigeria, Pakistan, Indonesia and Bangladesh.

In the present study, the incidence of sterile HVS report was found to be 24.5%, maximum found Staph aureus 37% and minimum in Pseudomonas 2% such similarity was proposed by Lamont et al⁷ et al which concluded that infection is responsible in 40% of cases and earlier the abnormal genital tract colonization is detected the greater is the risk of adverse outcome; with maximum of 40% Staph aureus followed by Klebsiella.

A study by Thomson et al²⁴ and Minkoff et al, 1984²³ showed UTI accounted for 12.5% of all causes of preterm labor, whereas sepsis caused for 40% of patients.

The present study shows that highest incidence of RDS followed by Sepsis, IVH, Jaundice, Pneumonia and NEC with cases found 52 (26%), 32 (16%), 29 (14.5%), 28 (14%), 12 (6%) and 8 (4%), respectively. Fanaroff A.A. et al, 1955²¹ found Preterm infants had the following complications – RDS (53%), IVF (4%), Sepsis (25%), NEC (25%).

Preventive measures like improvement in nutrition, rest, hydration, psychological support, tocolysis, steroid, neuroprotection with magnesium sulphate was given. Maternal outcome studied in the form of post partum haemorrhage, sepsis, febrile illness or any other maternal complication. Fetal outcome studied in the form of low birth weight baby, NICU stay, sepsis or any other fetal complications.²²

The Early Neonatal Deaths were found maximum 10% in 28.1- 30 weeks of GA and minimum 1.5% in 32.1- 34 weeks of GA. Quilligan et al²² in USA, the incidence of prematurity varies from 8-19% of all deliveries yet, they contribute to 85% of neonatal Mortality.

4.CONCLUSION

There was mean incidence of 12.18% of preterm birth in the Tertiary Care Hospital. Regular and comprehensive antenatal care, prevention and treatment of vaginal infections, prompt management of any other risk factors detected, can be a major step towards reducing the incidence of preterm births. Preterm labor has got multifactorial etiology. However high risk screening, early identification and vaginal microbiome study can prevent preterm labor and have good maternal and foetal outcome. Future scope of study- Vaginal microbiome study with respect to sensitivity of antibiotics can be implemented for immediate treatment of preterm labour if the test results are obtained in 24 hours.

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