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RESEARCH ARTICLE

Burden of aerobic organisms among women presenting with preterm labor (PTL) or preterm premature rupture of membranes (PPROM) in a tertiary care hospital in Bangalore, Karnataka, India

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ABSTRACT

Objective: To determine the proportion of aerobic organisms among patients presenting during pre term labour (PTL) or preterm premature rupture of membranes (PPROM). **Methods:** A cross -sectional study was conducted in the Department of Obstetrics and Gynecology of ESIC MC and PGIMSR, Bangalore from July 2017 to June 2022 on 130 pregnant women with preterm labor or PPROM ≤37 completed weeks of gestation. The sample was collected by inserting a sterile swab stick into the posterior and lateral fornix and cultured to determine the bacterial flora. **Results:** In our study, 49 patients (37.69%) were aged between 21-25 years and 44 (33.8%) between 26-30 years. 100 patients (77%) delivered between 34-37 weeks gestation and 30 patients (23%) < 34 weeks. A positive culture report was obtained in 35 patients (26.9%) – symptomatic Escherichia coli infection in 20(15%). 67 patients (51.5%) delivered vaginally, and 62 (47.7%) underwent LSCS. 54 patients (41.53%) had no comorbidities and among the rest, reduced liquor in 27(20.8%), hypertensive disorders in pregnancy in 28 (21.5%), and fetal growth restriction in 15(11.5%). **Conclusion**: Knowing the burden of aerobic infection in this group can provide an opportunity to deliver specific treatment and hence prevent morbidity in the mother and neonate.

Keywords: Preterm labour, preterm premature rupture of membranes, vaginal flora, aerobic organisms, Escherichia coli.

Preterm birth is defined as one that occurs after 20 weeks of gestation, before 37 completed weeks of gestation irrespective of the birth weight [1]. Premature rupture of membranes refers to the spontaneous rupture of membranes that occurs before the onset of labour [2]. Rupture of membranes occurs near term in most pregnant women. Preterm prelabour rupture of membranes (PPROM) is defined as the spontaneous rupture of fetal membranes before the onset of labour and before 37 weeks of gestation [3]. Preterm prelabour rupture of membranes complicates about 3% of pregnancies and contributes to 30 – 40% of preterm deliveries [4].

Ascending choriodecidual infection is an important factor in the causation of preterm labour and preterm premature rupture of membranes [5, 6]. Preterm labour leads to higher maternal and neonatal morbidity and mortality and is associated with a longer duration of hospital stay. Compared with term infants, preterm infants have a higher chance of developing cerebral palsy, respiratory distress, hypoglycemia, jaundice, kernicterus, seizures, periventricular leukomalacia, NICU admissions, and neonatal death [7]. This being an idiopathic disorder, one of the etiologies is an intrauterine infection characterized by early rupture of membranes and cervicovaginal discharge. Other

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risk factors for preterm labor include (1) genitourinary factors including uterine anomalies ; (2) behavioral factors like smoking, substance abuse, and malnutrition; (3) genetic and racial factors; (4) prior pregnancy history – prior preterm birth, prior abortions (5) current pregnancy risks – vaginal bleeding in the first trimester, assisted reproductive technique conception, multiple pregnancy, polyhydramnios, gestational diabetes, pre-eclampsia, fetal growth restriction; (6) other comorbidities – connective tissue disorders like Marfan's syndrome, Ehler Danlos syndrome, renal diseases like pyelonephritis, immunological disorders like SLE (Systemic Lupus Erythematosus), APLA (Anti Phospho Lipid Antibody syndrome) [1].

Normal vaginal flora in pregnancy is typically consisting mainly of Lactobacillus [8]. Abnormal vaginal colonization altering the normal flora and characterized by aerobic organisms like *Escherichia coli*, *Streptococcus* species, and *Enterococcus* species can result in preterm labour [9, 10]. If we identify the patterns of the cervical and vaginal flora that exists in this group of obstetric population with preterm labour, we can tailor the treatment by providing chemoprophylaxis to such women who stand at a higher risk of preterm labour (PTL) and/ or Preterm Premature Rupture of Membranes (PPROM) and hence reduce the burden of a preterm baby needing NICU care, financial constraints and prolonged stay for the mother and the baby. There is a scarcity of data regarding the identification and management of this obstetric subgroup. Therefore, we undertook this study to know the burden of the aerobic flora and so, eventually the measures that could be taken to reduce maternal and foetal morbidity and mortality.

Aims and objectives of the study**:** To determine the proportion of aerobic organisms among patients presenting during preterm labour (PTL) or preterm premature rupture of membranes (PPROM).

Materials and methods

The present study is a cross sectional study conducted in the Department of Obstetrics and Gynaecology from July 2017 to June 2022. Pregnant women between ≥24 weeks to ≤37 completed weeks of gestation with preterm labour or preterm premature rupture of membranes were considered. On admission, detailed history followed by general physical examination, obstetric examination, and per speculum and per vaginal examination was performed after requesting the woman to lie down in the dorsal lithotomy position. The posterior vaginal wall was retracted by inserting Sims' double bladed vaginal speculum. The anterior vaginal wall was also retracted.

A sterile swab stick was inserted into the posterior and lateral fornix and rotated by 360° to obtain the sample. This sample was sent immediately to the microbiology department for culture and sensitivity to determine the bacterial flora. This swab was cultured on blood agar, MacConkey agar, and Sabouraud’s Dextrose agar. Bacterial colonies are identified by their morphology and biochemical reactions like catalase, coagulase for gram-positive organisms and testing for motility, catalase, oxidase, sugar fermentation, nitrate reduction, citrate utilization, urea hydrolysis, methyl red and Voges-Proskauer test for gram-negative organisms.

To estimate the accurate gestational age we included the last menstrual period and calculated the expected date of delivery as per Naegele’s rule. In case this data was not available the previous obstetric ultrasound evaluation and or further obstetric ultrasonographic evaluation were performed to establish the gestational age.

Preterm labour was diagnosed based on the presence of regular, painful uterine contractions along with cervical dilation and/or effacement [1]. These patients were managed as per obstetric indications.

Inclusion criteria:

- All pregnant women are in labor above 24 weeks until 37 completed weeks of gestation.
- All pregnant women with preterm labour and or preterm premature rupture of membranes.

Exclusion criteria:

1. Women with intrauterine fetal demise
2. Women with ultrasonological evidence of fetal anomalies
3. Cord prolapse
4. Cervical polyp or growth

Statistical analysis: The proportion of patients with aerobic organisms among patients presenting with Preterm labour or premature rupture of membranes was found to be 30% based on a study done by Jigyasa et al in 2019 in Uttar Pradesh [11]. We estimated the sample size with a 95% confidence interval, 80% power, and relative precision of 8%. The sample size for the study was found to be 127. We have included 130 patients in our study. Data was entered into Microsoft Excel and statistical analysis was carried out in SPSS software version 24.0. Frequency and percentages were used to represent qualitative variables. Quantitative variables were represented as mean.

Results and observations

Out of 130 pregnant women included in our study, 66 women (50.7%) were primigravidas and the remaining 64 (49.23%) were multigravidas. 98 women (75.4%) had preterm labour and the remaining 32 women (24.6%) had PPROM. In our study, 49 women (37.69%) were aged 21 – 25 years and 44 women (33.8%) were between 26 – 30 years. The mean age was 27.36 years with a range of 19 to 38 years. Majority of the patients delivered as late preterm – 100 women (77%) between 34 to 37 weeks gestational age and the remaining 30 women (23%) delivered as early preterm < 34 weeks gestational age.

Table 1: High vaginal swab culture report

Organism grown		Number of cases(n=130)	Percentage
No growth		47	36.1%
Normal vaginal flora		48	36.9%
Gram-positive	Coagulase-negative *Staphylococcus* species	3	2.31%
	Enterococcus species	9	6.9%
Gram-negative	*Escherichia coli*	20	15.38%
	Klebsiella pneumoniae	2	1.54%
	Pseudomonas aeruginosa	1	0.77%

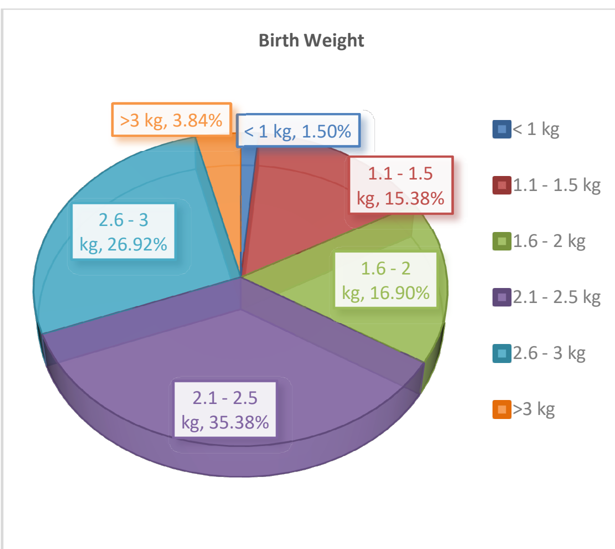

Figure 1: Distribution of birth weight

On the culture of the high vaginal swab, a positive report was obtained in 35 women (26.9%), normal vaginal flora was isolated in 48 women (36.9%) and there was no growth in the remaining 47 women (36.1%). The most frequently isolated organism among the positive reports was *Escherichia coli* (*E. coli*) in 20 women (15.38 %), followed by *Enterococcus* species in 9 women (6.9%), Coagulase negative *Staphylococcus* species in 3 women (2.31%), *Klebsiella pneumoniae* in 2 women (1.54%) and *Pseudomonas aeruginosa* in 1 woman (0.77%) (table 1).

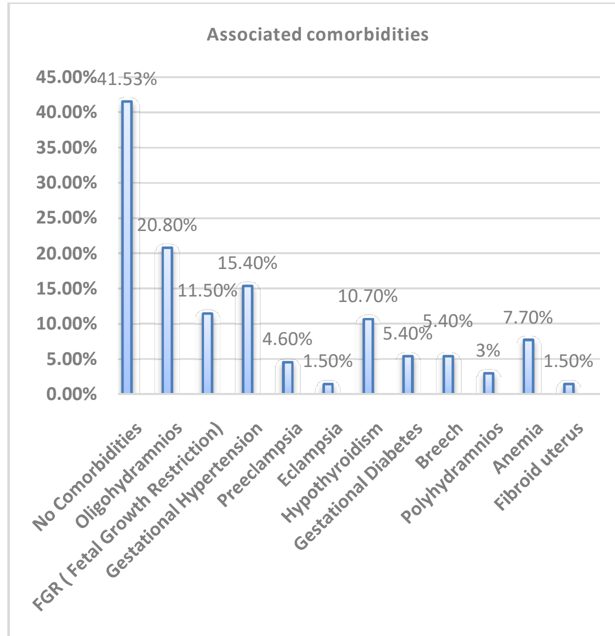

Figure 2: Associated comorbidities

67 women (51.5%) underwent preterm vaginal delivery, 62 women (47.7%) were delivered by LSCS, and 1 woman (0.77%) by forceps assisted vaginal delivery. Previous LSCS with an absolute indication for repeat LSCS was noted in 26 women (20%). Primary LSCS for fetal distress was done in 18 women (13.8%) and for breech presentation in 7 women (5.38%).

46 of 130(35.38%) babies weighed between 2.1 – 2.5 kilograms and 35 of 130(26.92%) babies weighed between 2.6 – 3 kilograms.

Among the 130 women, 54 women (41.53%) had no comorbidities, and among the rest, reduced liquor was seen in 27 women (20.8%), hypertensive disorders in pregnancy in 28 women (21.5%), and fetal growth restriction in 15 women (11.5%) (figure 2).

Discussion

Worldwide, approximately 1.1 million neonates die from complications related to prematurity [12]. Preterm labor accounts for approximately 12% of all births [13]. In our study, there was no difference in preterm labor and PPROM with parity – 50.7% primigravida and 49.2% multiparous. This correlates with the studies by Pandey et al (primigravida 52% and multipara 48%) [14] and Tavassoli et al (primigravida 55.9% and multipara 44.1%) [15]. 71.49% of women in our study were aged between 21- 30 years which correlates with the study by Chatterjee et al (61%) [16].

In resource limited settings, it is not possible to perform microbiological analysis of amniotic fluid. In such situations, high vaginal swab culture can be considered an alternative. In our study, a positive high vaginal swab culture was obtained in 35 cases (26.9%). The most frequently isolated organism was *E. coli* followed by *Enterococcus* species and Coagulase negative *Staphylococcus* species. This correlates with a retrospective study conducted by Li Y et al in Hong Kong in 2019 which showed that among patients of PPROM, positive high vaginal swab culture of gram-positive bacteria was found in 18.4% and gram-negative bacteria in 12.8% of which *E. coli* was the most common [17]. A prospective study conducted by Jigyasa et al in 2018 at Uttar Pradesh University of Medical Sciences showed positive high vaginal swab culture in 25.8% of women with PPROM. *E. coli* was the most common pathogen. 11 In this study similarity was found between the microorganisms isolated from amniotic fluid cultures - Peptostreptococcus, Bacteroides, Fusobacterium, Lactobacillus, Ureaplasma urealyticum, and the normal vaginal flora. This pointed the origin of infection towards normal bacterial vaginal flora.

A cross-sectional study was conducted by Rani et al in 2010 in Chandigarh in India on 100 women with PPROM. The culture of high vaginal swabs showed normal flora in 22% of cases. Pathogenic organisms grew in 18% of cases with *E. coli* being the most common (6%), followed by *Staphylococcus aureus* (4%) [18]. Similarly in the present study, *E. coli* was grown in 20 women (15.38%), *Enterococcus* species in 9 women (6.9%), and Coagulase-negative *Staphylococcus* species in 3 women (2.31%). A study by Chhabra and Patil showed a cervical colonization rate of 28% in preterm labor [19]. A study by Verma et al in Ludhiana showed a high vaginal swab positivity rate of 21.15% among women with preterm labor [20] similar to our study with a positivity rate of 26.9%.

Gram-negative bacilli behave as placental pathogens and lead to subclinical chorioamnionitis and preterm birth [21, 22]. Maternal vaginal infection with *E. coli* can be transmitted to the newborn after colonization or infection of the amniotic fluid or during vaginal delivery leading to neonatal sepsis [23]. In comparison with the faecal *E. coli* strains, the vaginal strains contain more virulence factors thus posing a higher risk of complications [24]. Similarly, in the present study, we had a burden of 20 women (15.38%) with *E. Coli*, and 15 women (11.5%) had vaginal discharge with soreness.

Limitations of our study: (1) Sample size is small; (2) We did not study the role of anaerobic organisms, trichomoniasis, candidiasis, and bacterial vaginosis which are also associated with preterm labour.

Conclusion

Due to the paucity of Indian data regarding the vaginal aerobic flora in preterm labour and PPROM, the identification of the burden of the aerobic organisms in 26.9% of preterm pregnant women through high vaginal swab culture, reiterates the need for evidence-based treatment in this subset of pregnant women to prevent maternal and neonatal suboptimal outcomes.

Conflict of interest: None. **Disclaimer:** Nil.

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