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ASYMPTOMATIC BACTERIURIA IN PRETERM AND TERM PROM DELIVERY IN A TERTIARY HEALTHCARE IN NORTH-EAST INDIA

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ABSTRACT **Background:** In order to promote safe outcomes for both the mother and the neonates, managing Premature Rupture of Membrane (PROM) is a crucial aspect of obstetric care. Causal relationship between the presence of Asymptomatic bacteriuria (ASB) and the types of PROM (preterm and term) and subsequent neonatal outcome in a tertiary healthcare in North East India is studied in this research work. **Methodology:** This research work is a Prospective Observational study in the Department of Obstetrics and Gynaecology, Central Referral Hospital (CRH), Gangtok conducted over a period of 1 (One) Year (January 2019 - December 2019). The present study was based on a primary sample of 250 women who were admitted with PROM (either preterm PROM or term PROM). Urine Routine Examination/ Microscopic Examination (RME) and Urine Culture Sensitivity (CS) were used for the study. Descriptive statistics, percentage, mean and standard deviation (SD) were calculated wherever found suitable. To establish association of each categorical variable with the type of pregnancy i.e., preterm and term PROM, $\chi 2$-test was used. In order to assess how well is the relationship between each parameter with the type of PROM, Spearman's Rank Correlation Coefficient "ρ" which is a non-parametric test using a monotonic function was also used. **Results:** The present study identifies that out of the 250 PROM samples, the incidence rate of preterm PROM is 58.4% against the 41.6%, for term PROM. The prevalence of ASB was found to have significant correlation with preterm PROM and term PROM deliveries according to the study, especially the presence of microscopic pus cells (1-4 HPF), epithelial cells and mixed growth of microorganisms. The type of PROM (preterm or term) was found to have significant correlation with adverse neonatal outcomes such as low birth weight, asphyxiated neonates, low APGAR score etc. **Conclusion:** Preterm and term PROM were found to be substantially linked with ASB. Urine RME and Urine CS are useful, accessible, and affordable methods for determining whether ASB is present in the early antenatal period. Diagnosing and treating ASB may reduce the likelihood of both preterm and term PROM occurrences as well as avoid adverse neonatal outcomes.

KEYWORDS : Asymptomatic Bacteriuria, PROM, PPROM, ASB

INTRODUCTION

The persistent bacterial colonization of the urinary system but without the presence of any symptoms is known as asymptomatic bacteriuria (ASB). It is a condition where the patient does not exhibit symptoms of a urinary tract infection (UTI), such as burning micturition, increased frequency of urination, or lower abdominal pain, yet a urine culture shows a significant number of pathogens, above 105 bacteria/ml [1]. It is prevalent to only 5–6% of non-pregnant women, but it is significantly prevalent during pregnancy since pregnant women are prone to have lower immunity, which seems to promote the proliferation of commensal and non-commensal microbes [11]. Urine concentration decreases due to the physiological increase in plasma volume during pregnancy, and up to 70% of pregnant women experience glycosuria, which promotes bacterial growth in the urine [7]. Pregnancy enhances the progression from asymptomatic to symptomatic bacteriuria, which may result in pyelonephritis and unfavorable obstetric outcomes like low birth weight, prematurity, and increased rates of fetal death [6,8,4].

Treating asymptomatic bacteriuria could avoid between 40% and 80% of pregnancy problems caused by acute pyelonephritis. About 25% of infected women will experience symptomatic infection during pregnancy if asymptomatic bacteriuria is left untreated [19]. Therefore, testing during pregnancy enables the provision of medication to lower the risk of Pyelonephritis progression and prevent the associated perinatal outcome. Due to the advantages of timely treatment, a number of organizations support screening pregnant women for asymptomatic bacteriuria, particularly during the first trimester [12].

According to Deering et al., 2007 [4], premature rupture of membrane (PROM) is the rupture of the membrane and loss of amniotic fluid before labor begins. PROM occurs in about 10% of pregnancies [1]. According to estimates, PROM is one of the most common underlying causes of perinatal death, accounting for 30 to 40% of premature deliveries [5]. Mal-presentation, cord compression, Oligohydramnios, necrotizing enterocolitis, cognitive impairment, intraventricular hemorrhage, and respiratory distress syndrome are among the effects that surviving newborns could suffer if PROM happens too early [13,16]. When PROM occurs before 37 weeks of gestation, it is referred to as preterm premature rupture of membrane, or preterm PROM [1,5]. Premature delivery is one of the most frequent adverse effects of preterm PROM [18]. The gestational age at which PROM occurs is typically negatively correlated with the latent period, or the interval between membrane rupture and birth [18]. Up to 3% of pregnancies result in preterm PROM, which complicates 30% to 40% of preterm births [10]. Respiratory distress, sepsis, intraventricular hemorrhage, necrotizing enterocolitis, and perinatal death are among the serious hazards it can cause for neonates [2,15]. A higher incidence of intraamniotic infection is linked to both preterm and term PROM [20]. The intra-amniotic fluid infection negatively affects perinatal morbidity and mortality of newborns.

Finding a causal link between the presence of ASB and neonatal outcome is an important goal in preterm and term PROM deliveries. In light of this, we conducted this prospective observational study in a tertiary healthcare in North-East India with the goal of determining the causal relationship with ASB and preterm and term PROM deliveries, their significant effect on her neonatal outcomes, using urine Routine Examination/Microscopic Examination (RME) and Urine Culture Sensitivity (CS) tests.

METHODOLOGY

This research work is a Prospective Observational study in Central Referral Hospital (CRH), Gangtok conducted over a period of 1 (One) Year (January 2019 - December 2019). This study was approved by the ethical review committee. A total of 250 (Two-Hundred and Fifty) patients with PROM (either preterm or term) cases admitted to CRH were included by purposive sampling technique using the selection criteria as detailed below.

Study Population

The diagnosis of PROM was made based on the patient's history, patients complain of sudden gush of fluid per-vaginally and sterile speculum examination to inspect the liquid escaping out through the cervix with coughing and straining and detection of pH by litmus or Nitrazine paper test.

Inclusion Criteria: All preterm and term PROM pregnant women.
Exclusion Criteria: Patients with following features were excluded from the study:

- Symptomatic bacteriuria (cystitis or pyelonephritis) at the time of urine collection,
- previously treated bacteriuria in current pregnancy,
- past medical history of known congenital or acquired urinary tract anomaly,

- o pregnant women on antibiotic treatment,
- o pregnant women with pre-existing renal anomalies and renal calculi and
- o women lost to follow up.

Data Collection Procedure

After receiving Institutional Review Board (IRB) permission, all patients who meet the inclusion and exclusion criteria and admitted during the study period were included in the study. Informed written consent was taken from each patient prior to enrollment. Their relevant medical history, physical and clinical examinations were done. Women included in the study were asked to collect clean catch, midstream urine samples in containers. The urine samples were sent for urine RME and CS. Women with asymptomatic bacteriuria were followed up. The Pretested Semi-structural Proforma, which was created specifically for this purpose, was used to gather information. The information, so collected, was first transferred to Microsoft Excel Worksheet. Then after thorough scrutiny and checking of the data, statistical analysis was performed by using IBM: SPSS Statistics Version 21. Mean±SD (Standard Deviation) was used to convey numerical and continuous variables that adhere to normality and equality of variances, whereas percentages and the number of instances were used to explain qualitative and categorical variables. To establish association of each categorical variable with the type of PROM i.e., preterm and term, χ2-test was used. In order to assess how well the relationship between each parameter varies with type of pregnancy, Spearman's Rank Correlation Coefficient "ρ" which is a non-parametric test using a monotonic function was used.

RESULTS

The purpose of this study was to establish a correlation between the occurrence of ASB and the two types of PROM, viz. preterm and term and their subsequent neonatal outcome. The results of the study are discussed below.

Incidence Rate of Type of PROM

The incidence of preterm or term PROM is illustrated in Table 1. It was found that, out of the 250 PROM women, there were 146 preterm and 104 term cases. This indicates that, among PROM cases, preterm cases had a slightly higher incidence rate (58.4%).

Table 1: Patient Profile.

Sl No.	Type of PROM	No. of Patients	Percentage
1	Preterm	146	58.4
2	Term	104	41.6

[1] Total Sample of Patients = 250

Routine Examination/ Microscopic Examination (RME)

RME is a quick, all-purpose urine health screening test that can identify possible UTIs, renal illness, or diabetes very quickly. The test consisted of urine examination for Albumin, sugar, RBC, Epithelial Cells, Casts cells and microscopy pus cells.

Albumin: The findings of the urine examination for Albumin is illustrated in Table 2. It is observed in the study that 230 patients did not have presence of albumin in urine, while only 20 patients indicated presence of albumin. Out of the 20 cases with albumin, 17 had less than 2mg/dL and only 3 had more than 2mg/dL. The pattern is quite in conformity within each type of PROM which is again supported by insignificant P-value (P = 0.957). Thus it may be deduced that the presence or absence of albumin in urine examination is akin to both the preterm and term PROM.

Sugar: The presence of sugar may signify the development of glycosuria, which may encourage bacterial growth in urine and further develop urinary tract infection. However, all the patients did not have the presence of sugar in urine (100%). This is true in both the types of PROMs. Therefore, the study for the test is overruled in this case.

***RBC*:** Presence of RBC may signify bleeding or infections, and hence included in this study. For RBC analysis, 4 (Four) groups, viz. Nil, 1 – 4 million/µL, 5 – 8 million/µL, and 9 & above million/µL were categorized. The findings are illustrated in Table 3. It may be observed from the table that RBC was not found in 175 (70%) of the cases, among which 56.6% in preterm and 43.4% in term cases. The nature and pattern of observance of RBC over the PROM types is found to be more or less the same despite some visible variations. This statement is further endorsed by the insignificant p-value (P = 0.838). This is true in both the types of PROMs

Table 2: Distribution of PROM Cases with Respect to Urine Examination-Albumin.

Albumin (mg/dL)	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
NIL	134 (58.3)	96 (41.7)	230 (92.0)	.087	2	.957
<2	10 (58.8)	7 (41.2)	17 (6.8)			
>=2	2 (66.7)	1 (33.3)	3 (1.2)			

Table 3: Distribution of PROM Cases with Respect to presence of RBC.

RBC (million/µL)	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
NIL	99 (56.6)	76 (43.4)	175 (70.0)	.847	3	.838
1 - 4	32(62.7)	19(37.3)	51 (20.4)			
5 - 8	9(64.3)	5(35.7)	14 (5.6)			
9 and Above	6(60.0)	4(40.0)	10 (4.0)			

Table 4: Distribution of PROM Cases with Respect to Presence of Epithelial Cells.

Epithelial Cells (HPF)	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
NIL	128 (61.2)	81 (38.8)	209 (83.6)	5.056	3	.168
1 - 4	12 (40.0)	18 (60.0)	30 (12.0)			
5 - 8	3 (60.0)	2 (40.0)	5 (2.0)			
9 and Above	3 (50.0)	3 (50.0)	6 (2.4)			

Table 5: Distribution of PROM Cases with Respect to the Presence of Casts Cells.

Casts Cells (HPF)	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
NIL	139(57.9)	101(42.1)	240 (96.0)	.989	2	.610
1	3(60.0)	2(40.0)	5 (2.0)			
2	4(80.0)	1(20.0)	5 (2.0)			

Table 6: Distribution of Cases with Respect to Presence of Microscopy Pus Cells.

Microscopy pus cells (HPF)	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
NIL	2(33.3)	4(66.7)	6 (2.4)	9.272	3	.026
1 - 4	141(60.0)	94(40.0)	235 (94.0)			
5 - 8	-	5(100)	5 (2.0)			
9 and Above	3(75.0)	1(25.0)	4 (1.6)			

Epithelial Cells: Epithelial cells in the urine may indicate irritation and may indicate infection in the urinary tract, hence included in this study. The same classification and analysis as done with RBC is made for epithelial cells and is illustrated in Table 4. It is observed from the table that, regardless of the type of PROM, the majority of the patients, that is 209 cases (83.6%) don't have epithelial cells in the RME test. Among the 41 remaining cases, 30 (12%) belong to the range of 1-4/HPF, 5 (2%) belong to the 5-8/HPF range and 6 (2.4%) belong to 9/HPF and above category. With an insignificant P-value= 0.168, the trend is persisted in both the PROM types and therefore it may be inferred that there is no disparity of either presence or absence of epithelial cells in the urine with regard to the type of PROM.

Cast Cells: It may be observed from Table 5 that the majority (96.0%) of the patients in the present study shows absence of cast cells in the urine and the minority (4%) shows presence of cells in the urine. Out of the presence cases, 2% each belong to 1/HPF category and 2/HPF category respectively. The pattern is almost consistent in both PROM types considered. It is observed that there is no variation between the types of PROM with respect to the presence of cast cells in the RME (P = 0.610).

Microscopy Pus Cells: The result of presence of Microscopic pus cells in urine RME is illustrated in Table 6. As evident from Table 6, for both the PROM types, the presence of microscopy pus cells is highest at 1 - 4 cells per high-power field (HPF) with 94%. It may be worth noting that, the cases with 5 and above cells per HPF, which may indicate mild infection to UTI is only 9% of, and only 2.4% of the cases with no microscopic pus cells. The P-value indicates that there is a significant correlation between the presence of pus cells in urine and the type of PROM with a P=0.036 which is significant at 5% probability level. Therefore, it may be deduced that the presence of microscopic pus cells highly correlates with the occurrence of preterm PROM.

Urine Culture Sensitivity Test

The urine culture sensitivity test, which cultivates bacteria in a lab for 48 hours to detect the presence and growth of microorganisms, was used as a diagnostic test. χ2-test is used to establish the association with the type of PROM

Table 7: Type of Pregnancy-wise Distribution of Cases with Respect to Urine CS for Presence of Micro Organisms.

Presence of Micro Organisms	PROM		Total (%, n=250)	χ^2-value	df- value	p- value
	Preterm (%)	Term (%)				
Candida species	1 (100.0)	-	1 (0.4)	3.693	4	.449
E.coli colony	1(25.0)	3(75.0)	4 (1.6)			
Klebsiella	1(100.0)	-	1 (0.4)			
Mixed Growth	10(66.7)	5(33.3)	15 (6)			
Sterile	133(58.1)	96(41.9)	229 (91.6)			

The findings of the study for the presence of micro organisms, viz, Candida species, E-coli colony, Klebsiella, Mixed growth and sterile, is illustrated in Table 7. From the table, it is observed that there were 229 PROM cases which are sterile, which is 91.6% of the overall cases analyzed. Out of these 229 cases, 133 cases were of preterm and 96 were of term cases. Sterile cases in a urine CS report generally indicate a normal, healthy urine sample with no significant infection or inflammation. Sterile means no bacteria grew in the culture, indicating no significant bacterial infection. Out of the 146 preterm cases in the table, 10 are of mixed growth and 1 each of candida species, E.coli colony, and klebsiella respectively. But within the 104 term cases, only 5 of mixed growth, 3 of E.coli colony, and no case is detected for candida species and klebsiella respectively. With an insignificant P-value=0.449 no significant correlation could be deduced regarding the type of micro organisms in the urine CS test and the type of PROM, except for the fact that preterm cases have a slightly higher presence of mixed growth of microorganisms.

Establishment of Relationship

The nature of direct relationship between the type of PROM and the parameters studied were analyzed using the Spearman's ρ, as shown in Table 8. There are three parameters which have a direct/ positive relation with the type of PROM while another two parameters have a negative relation ascribed by +ve and –ve values of ρ respectively. The positive relations consist of the presence of microscopy pus cells, epithelial cells, and presence of micro organisms whilst the negative encompasses RBC, and cast cells. Among the positive parameters, presence of epithelial cells is found to have a slightly significant relationship with type of PROM.

Table 8: Spearman's rho "*ρ*" Between Each Parameter with Type of PROM.

Sl. No.	Relationship Parameter	Spearman's ρ
1	Microscopy Pus Cells	.024
2	RBC	-.055
3	Epithelial Cells	.126
4	Cast Cells	-.049
5	Presence of Microorganisms	.020

The presence of microscopy pus cells and micro organisms show little significant relationship with type of pregnancy. Out of the negative relationship observed, none found to be significant statistically as none of the corresponding ρ-value is less than 0.05, which is the significance level adopted for this study.

Neonatal Outcomes

The neonatal outcomes studied in this work consisted of sex, parity, birth weight, APGAR score, history of asphyxia and NICU admission of neonates. Table 9 and 10 illustrates the distribution of type of PROM cases according to neonatal outcomes.

Sex: It is observed from Table 10 that the percentage of male babies among the preterm mothers is 60.7% as against 39.3% among term PROM. The corresponding figures for female babies are 55.0% and 45.0%. The total number of PROM cases either preterm or term was found to be slightly higher for male babies (150) than that of female babies (100). However, the test value suggests that there is no significant variation of the sex of the baby between preterm and term cases (P=0.373).

Parity: Parity patterns for parity-0 for preterm and term cases are found to be 96.2% and 3.8% while for parity-1, the figures are 1.4% and 98.6% respectively as seen from Table 9.

Table 9: Distribution of PROM Cases with Respect to Neonatal Outcomes.

Parameters		PROM		Total (%, n=250)	χ^2-value	df. value	p- value
		Preterm (%)	Term (%)				
Sex	Male	91 (60.7)	59 (39.3)	150 (60)	.793	1	.373
	Female	55 (55.0)	45 (45.0)	100 (40)			
Parity	0	4 (3.8)	102 (96.2)	106 (42.4)			
	1	142 (98.6)	2 (1.4)	144 (57.6)	226.039	1	<.001
Birth weight	2.0 - 2.5 kg	55 (98.2)	1 (1.8)	56 (22.4)			
	2.6 - 3 kg	86 (95.6)	4 (4.4)	90 (36)	210.633	3	<.001
	3.1 - 3.5 kg	4 (4.8)	80 (95.2)	84 (33.6)			
	3.6 – 4.0 kg	1 (5.0)	19 (95.0)	20 (8)			
APGAR score	< 5/10	25 (100.0)	-	25 (10)			
	>5/10	121 (53.8)	104 (46.2)	225 (90)	19.787	1	<.001
History of Asphyxia	Yes	14 (82.4)	3 (17.6)	17 (6.8)			
	No	132 (56.7)	101 (43.3)	233 (93.2)	4.308	1	.038
Admission to NICU	Yes	9 (81.8)	2 (18.2)	11 (4.4)	2.597	1	.107
	No	137 (57.3)	102 (42.7)	239 (95.6)			

The above discrepancy is tested by $\chi 2$ and found to be very highly significant statistically (P<0.001). This signifies that, preterm cases are more prone to parity-1 cases, whereas term cases tend to occur in parity-0 cases.

Birth Weight: There is a great discrepancy in terms of the birth weight of the baby which is highly significant at P<.001. Out of the 146 PROM cases for birth weight below 3kg, 141 cases belong to preterm cases (55 in 2-2.5kg category, and 86 in 2.6-3kg category), with only 5 babies were term PROM delivery. On the other hand, only 4 babies were preterm PROM delivery were found for the category above 3 kg, and the majority of the cases were term delivery (99). In brief it may be concluded that the birth weight of the baby of the preterm mothers is significantly low in comparison with the birth weight of the baby of the term mothers.

APGAR: Regarding the APGAR score, all the neonates of term PROM mothers have above 5/10 of APGAR score while 82.9% of the neonates of preterm mothers have above 5/10 of APGAR score. This also signifies high correlation between the APGAR score with the type of PROM (P<.001), whereby babies of preterm PROM mothers are prone to score less in APGAR score, signifying unhealthy babies.

Asphyxia: In case of incidence of asphyxia among the delivered babies, there is still significant variation between the types of pregnancy considered as evident by P=0.038 which is significant at 5% probability level. 82.4% of the babies of preterm PROM mothers have a history of asphyxia while 17.6% only of the babies of term PROM mothers.

NICU Admission of Neonates: It is worthwhile to mention that out of the 11 babies who required NICU admission, 9 (81.8%) babies are of preterm PROM mothers, while only 2 (18.2%) babies are of term PROM mothers. Nevertheless, the visible correlation is not significant enough statistically as p=0.107, which is greater than 0.05.

DISCUSSIONS

Although the exact origin of PROM is largely unknown, it has been reported that the causes might be a reduction in membrane strength or an increase in intrauterine pressure or both. It may be associated with an incompetent cervix, an unstable lie, polyhydramnios, bacteriuria particularly beta-streptococci infection [13]. The fetal membranes and the mother's uterine lining (decidua) have been shown to react to a variety of stimuli, such as membrane stretching and reproductive tract infections [13]. This prospective observational study found that the incidence rate of preterm PROM is 58.4% against the 41.6%, for term PROM among the sample of 250 PROM pregnancies. Preterm cases had a slightly higher incidence rate (58.4%) among PROM cases. As per urine RME tests, presence or absence of albumin, sugar, RBC, epithelial cells and cast cells in urine examination has no significant relationship with the causality of PROM, both for the preterm and term PROM. If present, the presence of epithelial cells is slightly correlated with the term PROM, with absence more correlated with preterm PROM. Presence of microscopic pus cells signifies the presence of PROM, especially in the range of 1-4 per HPF epithelial cells in the urine RME. This is in support of the earlier reports. However, this presence does not discriminate between the preterm or term PROM. On the other hand, the presence of microscopic pus cells highly correlates with the occurrence of preterm PROM with a p-value=0.026, which solidifies the earlier reports linking PROM with bacteriuria. However, no significant correlation could be deduced regarding the type of micro organisms in the urine CS test and the type of PROM with an insignificant P-value=0.449. It may be noted that, preterm cases have a slightly higher presence of mixed growth of microorganisms, if present. As per Spearman's correlation "ρ", presence of epithelial cells is found to have a slightly significant relationship with type of PROM. The presence of microscopy pus cells and micro organisms show little significant relationship with type of pregnancy.

Induction of labor, as opposed to expectant management, has been advised during term PROM as it may lower the risk of maternal infections without raising the risk of cesarean delivery [9]. However, the ideal gestational age for delivery has been reported to be unknown and varies by regional practice when preterm PROM occurs during the late preterm period. [9,17]. The significance of practice in North East India as derived by this study heavily correlates with that of term PROM cases.

Earlier studies reported respiratory distress, sepsis, intraventricular hemorrhage, necrotizing enterocolitis, higher incidence of intraamniotic infection, and perinatal death among the serious hazards preterm deliveries can cause for neonates [2,15]. With respect to neonatal outcomes, the study did not find perinatal deaths, The study

also found that there is no significant variation of the sex of the baby between preterm and term cases (P=0.373). However, parity-1 cases are more prone to preterm PROM deliveries, whereas term cases tend to occur in parity-0 cases. Also, the birth weight of the baby of the preterm mothers is significantly low in comparison with the birth weight of the baby of the term mothers. High correlation was found between the APGAR score with the type of PROM (P<.001), whereby babies of preterm PROM mothers are prone to score less in APGAR score, signifying unhealthy babies. There is a significant correlation between the types of PROM as evident by P=0.038, being that babies of preterm PROM mothers are prone to have a history of Asphyxiation. Also, the study found that 4.4% of the babies studies required NICU admission, out which majority were the babies of the preterm PROM mothers.

CONCLUSION

A prospective observational study was conducted to assess the prevalence of ASB in pregnancy with preterm and term PROM deliveries and its associated neonatal outcomes. ASB and its unwanted complications over perinatal outcomes have been correlated with PROM, especially with the preterm PROM. PROM and its complications could be avoided by properly evaluating early during the first prenatal care visits to identify any source of infection through urine RME and CS. In turn, the procedure might avoid complications for both the mother and the fetus. Antibacterial medication may help in longer pregnancies and lower rates of neonatal complications when used in the expectant management of preterm and term PROM. Proper diagnostic facilities, adequate monitoring facilities, and a standard protocol of management can be of utmost help in this regard. Further studies may be conducted with a bigger sample size. The relationship between the types of PROM and other parameters, including socioeconomic profile and occupation of the pregnant mothers may also be studied.

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