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Correlation of High Vaginal Swab Culture with Early Onset Neonatal Sepsis in Prelabour Rupture of Membrane

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ABSTRACT

Background: Prelabor rupture of membranes (PROM) refers to rupture of fetal membranes after fetal viability but before the onset of labor. PROM is associated with several maternal and neonatal complications, including chorioamnionitis, puerperal sepsis, and early onset neonatal sepsis (EONS). Ascending microbial infection from the cervico-vaginal canal is an important contributing factor. High vaginal swab (HVS) culture may help identify microorganisms associated with PROM and neonatal infection. This study aimed to evaluate the association between HVS culture positivity and the occurrence of EONS in mothers with PROM.

Methods: A cross-sectional study was conducted at Kalinga Institute of Medical Sciences, Bhubaneswar, from October 2020 to September 2022. A total of 334 pregnant women with PROM after 25 weeks of gestation were included. High vaginal swabs were collected before vaginal examination and antibiotic administration and were sent for culture and sensitivity testing. Neonates were followed for evidence of EONS based on clinical features, sepsis screening parameters, and blood culture results. Data were analyzed to determine the association between HVS status, PROM duration, gestational age, and neonatal sepsis.

Results: Among 334 mothers, 216 (64.5%) had positive HVS culture. EONS occurred in 12.6% of neonates. Neonates born to mothers with positive HVS had higher risk of sepsis compared to those with negative HVS (15.3% vs 7.6%; OR=2.18). PROM duration >18 hours significantly increased the risk of EONS, particularly in neonates born after 34 weeks of gestation.

Conclusion: Positive HVS culture and prolonged PROM are associated with increased risk of EONS. Early identification and close neonatal monitoring are essential to reduce neonatal morbidity.

Keywords: Neonatal sepsis. High vaginal swab, Premature rupture of membrane, Preterm labor

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INTRODUCTION

Prelabour rupture of membrane (PROM) refers to rupture of the membranes after the period of viability but before the onset of labor. It can be "preterm PROM" or "term PROM", depending on whether it occurs before or after 37 weeks of gestational age. [1] The incidence of prelabor rupture of membranes (PROM) is approximately 8% of term pregnancies. [2] The basic pathophysiology behind PROM is the release of matrix metalloproteases which alters the tensile strength of the membrane. [3] Bacterial invasion can cause inflammation which can trigger this process. [4] These microbes can ascend upward from the cervico-vaginal canal to uterus to infect the chorioamniotic membrane which surrounds the fetus. A significant number of maternal and neonatal complications are associated with PROM. Maternal complications include chorioamnionitis, postpartum hemorrhage and puerperal sepsis. Amongst neonatal complications, Early Onset Neonatal Sepsis (EONS) is one serious complication. [5] EONS can be defined as sepsis in neonates within 72 hours of life (few experts consider 7days). [6] It is associated with high mortality and long-term neurodevelopmental delay. High Vaginal Swab (HVS) may help the clinician to identify the microorganisms colonizing in upper part of the vaginal canal, which might be associated with PROM and may subsequently cause chorioamnionitis, puerperal sepsis and EONS.

The present study aimed to evaluate the association between HVS and EONS in PROM.

MATERIALS AND METHODS

The present cross-sectional study was conducted at Kalinga Institute of Medical Sciences, Bhubaneswar, over a period of two years from October 2020 to September 2022. Prior approval was obtained from the Institutional Ethics Committee of Kalinga Institute of Medical Sciences, Bhubaneswar (Letter No. KIIT/KIMS/IEC/452/2020 dated 03/11/2020).

Sample size: Considering 68% positivity among PROM with 5% level of significance and 5% precision, the calculated sample size was 334.[7]

Study Participants: Patients with PROM admitted in Obstetrics and Gynaecology(O&G) Department of Pradyumna Bal Memorial Hospital, KIMS were enrolled into this study after taking informed consent. Neonates born to the patients were followed up.

Inclusion criteria: Patients with PROM who has completed 25 weeks of gestation.

Exclusion criteria: The exclusion criteria included mothers who had received antibiotics or had undergone per speculum (P/S) and/or per vaginal (P/V) examination prior to admission; patients presenting with clinical features of chorioamnionitis such as fever, tachycardia, uterine tenderness, or foul-smelling liquor; cases with meconium-stained amniotic fluid or evidence of fetal distress at admission; pregnancies complicated by any medical or surgical disorders; multiple gestations, intrauterine growth restriction (IUGR), intrauterine fetal demise (IUD), or congenital fetal anomalies; and patients unwilling to participate in the study.

For diagnosing, sterile speculum examination was done under all aseptic precautions to check for the accumulation of liquor in the vaginal fornix or amniotic fluid leakage from the cervical OS. High vaginal swabs were taken prior to P/V examination and before starting prophylactic antibiotics which were sent for culture and sensitivity test and was checked for growth of organisms like Staphylococcus epidermidis, Staphylococcus aureus, Staphylococcus saprophyticus, Enterococcus, Acinetobacter, Klebsiella, Pseudomonas, Candida, E. coli, GBS mainly. If present, antibacterial testing was done as per Clinical and Laboratory Standards Institute (CLSI) guidelines. The Kuppuswamy classification has been used to stratify the patients in different socioeconoimic status for comparative analysis.

Diagnostic criteria for neonatal sepsis:

Neonatal sepsis was diagnosed in neonates based on clinical signs and symptoms consistent with sepsis, requiring either at least two positive parameters on a sepsis screen or a positive blood culture. The clinical features considered indicative of neonatal sepsis included absent spontaneous movement, reduced feeding ability, pyrexia or hypothermia (temperature ≥37.5°C or <35.5°C), severe chest indrawing, rapid breathing (respiratory rate >60/min), convulsions, jaundice appearing within the first 24 hours of life, or deep jaundice at any age as evidenced by yellow discoloration of the palms and soles.

The sepsis screen itself comprised five laboratory parameters: a total leucocyte count below 5,000/mm³, a low absolute neutrophil count as determined by the Manroe chart for term babies or Mouzinho's chart for very low birth weight babies, an immature-to-total neutrophil ratio greater than 0.2, a micro-ESR exceeding 15mm in the first hour, and a C-reactive protein (CRP) level above 1mg/dl.

Blood was drawn from a peripheral vein under aseptic precautions before starting any antibiotics. 2 ml of blood had been withdrawn and transferred to lab in Bactalert pediatrics blood culture bottle and in the lab, it will be processed automatically. It was examined for growth of organisms like GBS, E. coli, Klebsiella, Enterococcus, Staphylococcus, Acinetobacter and Pseudomonas etc. for 5 days and antibiotic sensitivity patterns were done.

RESULTS

Out of 334 mothers, 216 women (64.5%) had positive HVS and 118 women (35.5%) had negative HVS (table 1). In our study the mean (SD) age of study participants (mothers) was 28.74 (4.45) years. Majority of participants were in the age group between 25 to 30 years and belonged to upper lower socio-economic status. 64.6%

participants were primigravida and 80% participants' gestational age was more than 34 weeks. 57.7% of participants' LD duration was less than 18 hours and 58.1% had undergone LSCS. Participants with different LD duration as well as different mode of delivery were equally distributed across high vaginal growth swab status (p-value >0.05).

In neonates <34 weeks of gestational age, prevalence of sepsis was independent of the PROM duration (table 2). The neonates born at >34 weeks had higher incidence of sepsis in maternal PROM >18 hours, in comparison to maternal PROM <18 hours. The odds ratio of developing sepsis with leakage duration more than 18 hours rises with increase in gestational age. The risk which is 1.32 times at gestational age of <34 weeks, becomes 4 at 34-37 weeks and 11.67 at >37 weeks.

Table 1: Distribution of socio-demographic and clinical variables across high vaginal swab culture

Variables	HVS negative (n= 118) (%)	HVS positive (n= 216) (%)	p-value
Age in years, mean (SD)	28.07 (4.16)	29.11 (4.57)	0.040
Socio-economic Status			
Lower	21 (17.8)	23 (10.7)	0.270
Lower Middle	25 (21.2)	55 (25.5)	
Upper lower	59 (50.0)	111 (51.4)	
Upper middle	5 (4.2)	16 (7.4)	
Upper	8 (6.8)	11 (5.1)	
Gravida			
Primi	81 (69.2)	134 (62.0)	0.190
Multi	36 (30.8)	82 (38.0)	
Leaking to Delivery (LD) duration			
≥18 hours	47(39.84)	96(44.44)	0.41
<18 hours	71(60.16)	120(55.56)	
Mode of delivery			
LSCS	74 (62.7)	120 (55.6)	0.205
VD	44 (37.3)	96 (44.4)	

Table 2: Duration of prom and eons across various gestational ages

Gestational age	Total number	LD duration (n)	Septic neonates n (%)	Non septic Neonates n (%)	Odds ratio
<34 weeks	69	>18 Hours (n=44)	13(29.5%)	31(70.5%)	1.32 (95% CI: 0.43 to 4.01)
		<18 Hours (n=25)	6(24%)	19(76%)	
34-37 weeks	103	>18 Hours (n=48)	9(18.75%)	39(81.25%)	4 (95% CI: 1.03 to 15.6)
		<18 Hours (n=55)	3(5.45%)	52(94.54%)	
>37 weeks	162	>18 Hours (n=51)	9(17.65%)	42(82.35%)	11.67 (95%CI: 2.48 to 54.9)
		<18 Hours (n=111)	2(1.8%)	109(98.2%)	

Table 3: Distribution of EONS across HVS

HVS	Total	Septic neonates	Non septic neonates	Odds ratio
Positive	216	33 (15.3%)	183 (84.7%)	2.184 (95% CI:1.00 to 4.73)
Negative	118	9 (7.6%)	109 (92.4%)	

Women with positive HVS are at higher risk of developing EONS (table 3). HVS has sensitivity, specificity, Positive Predictive value and Negative predictive value of 78.6%,37.33%,15.28% and 92.4% to identify early onset neonatal sepsis. The odd ratio of having sepsis in neonates with positive HVS is 2.184 with 95% CI of 1.00 to 4.73 which is more than double of those with negative HVS.

DISCUSSION

A hospital-based observational cohort study was conducted with 334 patients to correlate the culture-positive HVS with EONS. In our study the incidence of sepsis amongst neonates born at <34 weeks, 34 – 37 weeks and >37 weeks with the presence of maternal PROM were 27.5%, 11.6% and 6.7% respectively and incidence of culture positive neonatal sepsis were 11.59%, 0.9% and 0.6% respectively. Ibishi VA et al[8] conducted a study between the year 2013 and 2015 in which they found that out of 200 neonates born to mother complicated with PROM, 13% had neonatal sepsis. Asindi AA et al[9] also found that incidence of neonatal sepsis following PROM was 14 %. Our study is congruent with their study with incidence of neonatal sepsis of 12.6%. However, Popwski T et al.[10] reported an incidence of only 4.3 % which is much lower than our study which might be because of consideration of participants with gestational age >34 weeks only. Study conducted by Wu J et al.[11] in China reported an incidence of 25 % which is much greater than our study. Alam MM et al.[12] conducted a study between 2007 to 2011 in which they reported an incidence of 4%. In one of the study by Linder N et al, among 135 neonates born to mothers complicated with PROM , the incidence of EONS was 8.1% and it was around 15 % in preterm neonates.[13] Similarly, Lee SY at al.[14] reported an incidence of 6.5% of culture-proven sepsis among neonates born from mothers having PROM. The relatively higher incidence of neonatal sepsis in our study could be explained by socioeconomic status, weather conditions being conducive for infection with India being a tropical country.

Neonates born at lower gestational age with risk factor of maternal PROM were significantly at higher risk of neonatal sepsis compared to neonates born at term gestational age.

Table 4: Comparison of our study with different studies

Study	Sample size	Gestational age	Incidence of Early onset Neonatal sepsis
Ibishi VA et.al.[8]	200	>28 weeks	13%
Popowski T et.al.[10]	399	>34 weeks	4.3%
Linder N et. al.[13]	135	>28 weeks	8.1% overall 15% in preterm
Lee SY et.al.[14]	306	25-35 weeks	6.5%
Our study	334	>25 weeks	12.6% overall

The variability in incidence of EONS following PROM might be due to geographical variations, use of antibiotics and different criteria used by various researchers to report the incidence of EONS. In subgroup analysis based on duration of PROM <18 hours versus >18 hours, we found neonates born at >34 weeks had higher prevalence of sepsis in maternal PROM >18 hours, in comparison to maternal PROM of <18 hours duration. Considering lower incidence of sepsis in term and late preterm neonates born to mother with PROM, the empirical antibiotics can be avoided if duration is <18 hours but needs close monitoring for the appearance of signs and symptoms of sepsis. Zhuang L et al conducted a study in which they included term neonates who were born to mothers complicated with PROM and they found that the risk EONI was higher if leaking to delivery duration was more than 16 hours [15]. In a study conducted in preterm neonates they found that the latency period was not a good predictor of EONS in mothers complicated with PPROM [16].

Neonates of mother with positive high vaginal growth swab status when compared to negative high vaginal growth swab status were more likely to be diagnosed with sepsis (15.3% vs. 7.6%, p-value <0.05). HVS culture had sensitivity, specificity, Positive Predictive value and Negative predictive value of 78.6%,37.33%,15.28% and 92.4% to identify EONS. In the study by Beevi PA et al [17], out of 21 mothers with positive high vaginal swab growth babies of 15 mothers developed early onset neonatal sepsis compared to 20 out of 80 babies who were born from mothers with high vaginal swab negative status. In the study by Anitha AK et al. [18], positive growth in high vaginal swab was associated with neonatal sepsis in 25.5% of babies compared to 4.8% in swab negative mothers. These studies partially support our observations as in all studies positive high vaginal swab culture was associated with more cases of neonatal sepsis but incidence varies which might be due to geographical variations and different criteria used to detect neonatal sepsis.

LIMITATION

Being a single-centered study, the findings may not be generalizable to the wider population. Additionally, the lack of specialized culture media in our hospital setup for organisms such as *Ureaplasma urealyticum*, *Chlamydia trachomatis*, *Mycoplasma* species, and anaerobic bacteria limited the ability to identify these pathogens among participants. Furthermore, high vaginal swab (HVS) samples were collected at the time of presentation rather than prior to delivery, which may have influenced the microbiological findings and limited the assessment of infection status during the antenatal period.

CONCLUSION

Neonates of the mother with positive HVS growth compared to negative HVS growth were more likely to be diagnosed with sepsis, however, in subgroup analysis, it was found that HVS significantly correlate with the chances of EONS if gestational age >37 weeks. For <37 weeks care should be taken even if the HVS is negative especially if the leaking to delivery duration is more than 18 hours. For neonates born at <34 weeks, clinicians should be careful about the occurrence of EONS irrespective of HVS culture and leaking to delivery duration and maternal antibiotics should not be stopped based on the HVS report.

Individual Author's Contribution: AG and **HS** contributed to study conception, study design, data collection, data analysis and interpretation, and manuscript preparation. **HB** contributed to study design, data analysis and interpretation, and manuscript preparation. **LP** contributed to study conception, study design, data analysis and interpretation, and manuscript preparation.

Availability of data: The data that support the findings of this study are available from the corresponding author on reasonable request.

Declaration of Non-use of generative AI Tools: This article was prepared without the use of generative AI tools for content creation, analysis, or data generation. All findings and interpretations are based solely on the authors' independent work and expertise.

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