

A MICROBIOLOGICAL STUDY ON EARLY ONSET NEONATAL SEPTICAEMIA AND ITS CORRELATION WITH THE VAGINAL MICRO FLORA OF MOTHER IN LABOUR

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Abstract

Background: According to a report from the World Health Organization, an estimated 500,000 women suffers death annually due to complications of pregnancy and childbirth. Puerperal sepsis is the third most common cause of maternal mortality worldwide. Postpartum puerperal sepsis was also significantly associated with the occurrence of early neonatal mortality due to early-onset neonatal sepsis (EONS). The objective of the study was to find out about maternal colonization during active labor in low-risk women and its possible association with EONS and puerperal sepsis. **Materials and Methods:** A cross-sectional study conducted from July 2022 to June 2023 in the department of Microbiology, SKMCH, Muzaffarpur, Bihar after approval from the ethical committee of the Institute. A total of 521 samples could be obtained during this period. All consecutive antenatal women attending the outpatient or emergency department and getting admitted in the labour room were screened to be included in the study. We included low-risk pregnant women with a period of gestation of >28 weeks and in active labor (≥ 4 cm) with the intact membrane. Two high vaginal swabs were taken and transported to the microbiology lab immediately for processing. Out of the two HVSSs, one was cultured for six to eight hours on enrichment media followed by subculture on CHROM agar™ Strep B (CHROM agar, Paris, France) for Group B Streptococcus (GBS). Another swab was cultured on Mac-Conkey agar and Blood agar. The morphology of the colonies grown was studied and one representative of each morphologically distinct colony was picked up and sub-cultured onto new agar plates and was further identified by standard biochemical tests up to species level. **Result:** Out of 521 women, 284 (54.5%) showed vaginal colonization. Vaginal colonization was predominantly by gram-positive organisms in 125 (23.9%) followed by a mixture of organisms in 108 (20.7%) and gram-negative organisms in 67 (12.9%) of enrolled women. The majority of women yielded coagulase-negative Staphylococcus [CoNS] followed by Escherichia coli, methicillin-resistant S. aureus (MRSA) and methicillin-sensitive S. aureus (MSSA). **Conclusion:** Our study concludes that vaginal colonization with potentially pathogenic bacteria in low-risk women in labor usually does not lead to puerperal sepsis or EONS.

INTRODUCTION

According to a report from the World Health Organization, an estimated 500,000 women suffers death annually due to complications of pregnancy and childbirth.^[1] Hence, the care during labor is vital for the complete recovery of the woman and her newborn. It is known that sepsis, hemorrhage, and prolonged and obstructed labor are some of the factors at delivery that are responsible for increased maternal and neonatal morbidity.^[2,3] Puerperal sepsis is the third most common cause of maternal mortality

worldwide.^[4] In low- and middle-income countries, puerperal infection is the sixth leading cause of the disease burden in women during their reproductive years and accounts for 15% of total maternal deaths whereas in high-income countries, the death rate due to puerperal sepsis is less than 10% of all causes of maternal death.^[5] Puerperal sepsis can be caused by endogenous or exogenous bacterial inoculation of the uterine cavity. Endogenous bacteria are normal commensals in the vagina and rectum but can get carried into the uterus from the vagina by examining fingers or instruments during pelvic examinations

particularly following prolonged rupture of membranes, obstructed labor, and traumatic vaginal delivery. Exogenous bacteria can be introduced into the vagina by unclean hands or unsterile instruments. Postpartum puerperal sepsis not only leads to acute morbidity in women but also long-term morbidities like pelvic inflammatory disease and infertility. Postpartum puerperal sepsis was also significantly associated with the occurrence of early neonatal mortality due to early-onset neonatal sepsis (EONS).^[6]

EONS presents within 72 hr of birth, and accounts for 10.4% to 85% of total neonatal sepsis.^[7] The commonest causative organisms for EONS are endogenous bacteria that are acquired vertically during the process of birth through the mother's reproductive tract. The objective of the study was to find out about maternal colonization during active labor in low-risk women and its possible association with EONS and puerperal sepsis.

MATERIALS AND METHODS

A cross-sectional study conducted from July 2022 to June 2023 in the department of Microbiology, SKMCH, Muzaffarpur, Bihar after approval from the ethical committee of the Institute. A total of 521 samples could be obtained during this period. All consecutive antenatal women attending the outpatient or emergency department and getting admitted in the labour room were screened to be included in the study. We included low-risk pregnant women with a period of gestation of >28 weeks and in active labor (≥ 4 cm) with the intact membrane. Women with a history of any foul-smelling vaginal discharge, fever within seven days, antibiotics intake within seven days of admission, infected with HIV and active perineal infection, who had undergone five or more vaginal examinations during labor, undergoing cesarean section (lower segment caesarean section [LSCS]), or having any clinical features suggestive of chorioamnionitis were excluded from the study. Women who gave birth to the neonate with major congenital malformations non-compatible with life and Apgar score <3 at five minutes were also excluded from the study. Labor details for the first, second, and third stages of labor were recorded in a predesigned proforma. All of the mothers were observed for the first 72 hr in the hospital for clinical signs and symptoms of sepsis and

those with clinical signs were investigated further. They were followed up for two weeks afterward for clinical signs and symptoms of puerperal sepsis. Women were counseled regarding symptoms, for which they might require readmissions such as fever with purulent foul-smelling lochia, pain in the lower abdomen, episiotomy site infection/wound gape, abdominal distention, and other symptoms suggestive of puerperal sepsis.

Two high vaginal swabs were taken and transported to the microbiology lab immediately for processing. Out of the two HVSSs, one was cultured for six to eight hours on enrichment media followed by subculture on CHROM agar™ Strep B (CHROM agar, Paris, France) for Group B Streptococcus (GBS). Another swab was cultured on Mac-Conkey agar and Blood agar. These plates were incubated at 37 degree for 24 hr to allow for the growth of bacterial colonies. The morphology of the colonies grown was studied and one representative of each morphologically distinct colony was picked up and sub-cultured onto new agar plates and was further identified by standard biochemical tests up to species level. Microorganism's identification was followed by antibiotic sensitivity, performed by the standard procedure according to Clinical and Laboratory Standards Institute (CLSI) 2019 guidelines.^[8]

The data was collected in a Microsoft suite and analyzed using SPSS version 24.0 (IBM Corp., Armonk, NY). A p-value of <0.05 was taken as statistically significant.

RESULTS

Out of 521 women, 284 (54.5%) showed vaginal colonization. There was no statistically significant difference observed between vaginal colonization with maternal age, the period of gestation, term/preterm births, and birth weight of babies, whereas there was a statistically significant relationship between parity and vaginal colonization ($p=0.01$). [Table 1]

Vaginal colonization was predominantly by gram-positive organisms in 125 (23.9%) followed by a mixture of organisms in 108 (20.7%) and gram-negative organisms in 67 (12.9%) of enrolled women. The majority of women yielded coagulase-negative Staphylococcus [CoNS]) followed by Escherichia coli, methicillin-resistant S. aureus (MRSA) and methicillin-sensitive S. aureus (MSSA). [Table 2]

Table 1: Socio-demographic characteristics of the enrolled participants

Character	Isolated colony		Total (521)	P value
	Yes (284)	No (237)		
Maternal age	<20 years	12	16	>0.05
	20-30 years	231	183	
	>30 years	41	38	
Parity	Primigravida	132	151	<0.05
	Multigravida	152	86	
Mode of delivery	NVD	282	226	>0.05
	Assisted	2	11	
Period of gestation	28-32 weeks	2	6	>0.05
	33-37 weeks	10	6	

	>37 weeks	272	165	497	
Birth weight	≤1.5 kg	1	1	2	>0.05
	1.5 - 2.49	73	54	127	
	≥2.5 kg	210	182	392	

Table 2: Distribution of samples based on isolated aerobic bacteria in high vaginal swabs

Isolated organism	Number (%)
Gram positive organism	125 (23.9%)
CoNS	94
MRSA	13
MSSA	12
Enterococcus sp.	5
Group B Streptococcus	1
Gram negative organism	67 (12.9%)
Escherichia coli	48
Klebsiella pneumoniae	11
Enterobacter species	6
Pseudomonas aeruginosa	1
Acinetobacter baumannii	1
A mixture of ≥ 2 organism	88 (16.9%)
Contaminants	4 (0.77%)

DISCUSSION

In our study, 54.5% of studied women had vaginal colonization with pathogenic aerobic bacteria but none of the colonized women and their neonates developed puerperal or early-onset neonatal sepsis, respectively.

In pregnancy, the vaginal flora gets replaced by polymicrobial organisms, e.g. *G. vaginalis*, anaerobic gram-negative rods such as *Prevotella* species, *Peptostreptococcus* species, *Mycoplasma hominis*, *Ureaplasma urealyticum*, *Staphylococcus*, and often *Enterobacter* species. This change in bacterial flora can sometimes lead to asymptomatic or symptomatic bacterial vaginosis and to chorioamnionitis in pregnant women. Furthermore, the consequences are complications in the form of puerperal sepsis, EONS, and preterm labor.^[9] In the present study, we focused only on the isolation of aerobic bacteria in vaginal colonization in low-risk asymptomatic pregnant women with intact membranes who presented to the tertiary-level hospital for delivery. The objective was to find out any increased risk of complications like puerperal sepsis and EONS in these subsets of low-risk pregnant women. In the study, CoNS (gram-positive organism) was the most common single isolated organism whereas various studies have reported gram-negative bacteria predominantly.^[10,11] The rate of vaginal colonization with maternal age was not found to be significant. But few studies have reported increased colonization with age, predominantly in >30 years of age and some in <20 years of age.^[10-13] Multigravidas were at higher risk of colonization than primigravidas. This is similar to studies by Febriani et al., Akkanesermsaeng et al., and Sharmila et al.^[10,14] Multiparous women have had more contact with the health system and may have undergone more genital examinations in their reproductive tenure when compared to the primigravidas. This hypothesis is supported by Top et al., who observed an increase in the occurrence of MRSA and MSSA in multigravidas.^[15]

In our study, out of total births in mothers with vaginal bacterial colonization, 78.1% were term and 21.9% were preterm, comparable to others.^[12] Vaginal bacterial colonization did not affect the gestational age at the time of labor ($p=0.06$). Thus, we can suggest that vaginal colonization does not increase the risk of preterm deliveries. Our finding is possibly due to exclusion of subjects with premature rupture of membranes (PROM), preterm premature rupture of membranes (PPROM), and chorioamnionitis that are more likely to predispose to preterm labor.

CONCLUSION

Our study concludes that vaginal colonization with potentially pathogenic bacteria in low-risk women in labor usually does not lead to puerperal sepsis or EONS. It reinforces that low-risk mothers should not be given routine antibiotic prophylaxis during labor and postpartum for the prevention of puerperal sepsis and EONS. Multiple vaginal examinations should be avoided during active labor even in a low-risk woman as it increases the risk of vaginal colonization.

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