The Study of the Pattern of Maternal Vaginal Flora in Labour and Its Association with Neonatal Sepsis

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Research Article

Abstract: Infection continues to account for a major proportion of maternal, fetal and neonatal mortality and morbidity worldwide. The shared relationship between mothers and their newborns leads to common risk factors and etiologies of infectious diseases. Neonatal sepsis continues to be one of the leading causes of neonatal mortality in India. Neonatal sepsis is defined as a clinical syndrome of bacteremia with systemic signs and symptoms of infection in the first 4 weeks of life. The present study has been undertaken to understand the pattern of maternal vaginal flora in labour and its relation with neonatal sepsis. Knowledge of the significant organisms in the genital tract of the pregnant woman and the peripartal risk factors can help us to develop strategies to control the transmission from mother to child or from environment to the neonate. Prevention of these infections could be practiced through screening programs for mothers vaginorectal colonization, identification of pathogenic organisms and administration of antibiotic prophylaxis to mothers or high risk neonates. Early treatment with appropriate antibiotics would minimize the risk of severe morbidity and mortality besides reducing the emergence of multidrug resistant organisms in intensive care units by rational antibiotic use.

Keywords: maternal vaginal flora, neonatal sepsis, early onset sepsis, antibiotic prophylaxis.

Introduction

During the newborn period, infection remains an important cause of morbidity and mortality, despite increasing sophistication in infant intensive care and the use of broad –spectrum antimicrobial agents. Most neonatal bacterial infections occur in the first week of life as a result of exposure to maternal genital microorganisms during the intrapartum period.[1] Newborns may acquire early –onset neonatal infection 'vertically' (mother to newborn during birth) from endogenous bacteria in the mother's reproductive tract ,which may or may not cause disease in the mother but can cause disease in the newborn. The pattern of organism and the heaviness of colonization are usually closely similar between mother and the baby.[2] Intrauterine infection has been shown to play a major role in induction of preterm birth and neonatal sepsis.[3] According to the National Neonatal Perinatal Data (NNPD 2002-2003),the incidence of neonatal sepsis is 30/1000 live births.[4] When pathogenic bacteria gain access into the bloodstream ,they may cause overwhelming infection without much localization(sepsis) or may be predominantly localized to the lung (pneumonia) or the meninges (meningitis).[5] Neonatal sepsis may be classified according to the time of onset of the disease, early onset sepsis (EOS) and late onset sepsis (LOS) This distinction has clinical relevance, as EOS disease is mainly due to bacteria acquired before and during delivery and LOS disease due to bacteria acquired after delivery (nosocomial and community sources).[6] These definitions have contributed greatly to diagnosis and treatment by identifying which microorganisms are likely to be responsible for sepsis during these periods and the expected outcomes of infection. According to the National Neonatal Perinatal Database of India, Klebsiella pneumonia, Staphylococcus aureus and E .coli are the three most common organisms causing neonatal sepsis both in hospital and community[4].Common risk factors associated with increased severity of neonatal sepsis are birth weight and gestational age[5].In the newborn, septicemia, preterm birth, respiratory disorders as well as some neurological disorders seem to be consequences of such ascending genital tract infections in the pregnant woman[1]. There are not many studies which correlate such maternal genital colonization with neonatal sepsis. The magnitude of this problem in communities with poor maternal hygiene especially in the lower socio-economic strata needs to be assessed in greater detail in all developing countries. The pattern of organisms causing sepsis also differs from place to place and can change in the same place over time. Knowledge of the prevalence of the local isolates and their antimicrobial sensitivity is of utmost importance to prevent emergence of multi drug-resistant organisms in our neonatal intensive care units(NICU).

Aims and Objectives

To study the vaginal flora of pregnant women in labour and to determine if the specific vaginal microflora is associated with neonatal sepsis.

Materials and Methods

This was a prospective analysis conducted at the Department of Obstetrics and Gynecology and the Neonatal Intensive Care Unit of Krishna Institute of Medical Sciences, Karad during the period of April 2012 to March 2013. The present study was carried out on 353 pregnant women in labour. High vaginal swabs were taken from these women in labour at the first vaginal examination after taking informed consent. The vaginal swabs were taken from the posterior fornix with complete aseptic precautions using a speculum. The swabs were immediately transported to the microbiology laboratory in a sterile test tube .The vaginal swabs were cultured on blood agar, McConkey and chocolate agar and incubated at 37°C. Growth, if any was identified by the standard bacteriological techniques including Gram staining, colony characteristics, biochemical properties and slide agglutination where appropriate. Antibiotic sensitivity of strains was performed using Modified-Kirby-Bauer Disc diffusion method. A detailed history was obtained from the mothers which included the age, last menstrual period and expected date of delivery, ultrasonographic findings in antenatal scans, obstetric history ,presenting complains at labour, with premature rupture of membranes, associated disorders and drugs used during pregnancy. The type of bacteria isolated from mother's genital tract and their antibiotic sensitivity pattern was studied. The neonates born to these mothers were further evaluated during their duration of stay in the hospital. A detailed history regarding their sex, gestational age, birth weight, single or twins, mode of delivery, apgar score at delivery, further details of their general and systemic examination were all noted in the prepared proforma. All babies were examined daily during their stay for signs of sepsis such as poor feeding, lethargy, shock, fever, respiratory distress, skin pustules, eye and umbilical discharge, jaundice, convulsions and were admitted to the NICU for further investigations and management. They were further classified as early onset sepsis (EOS) within 72 hours of birth and late onset-sepsis (LOS) after 72 hours of birth. All signs, symptoms, investigations, diagnosis and outcome were recorded in the proforma.

Inclusion Criteria

Pregnant women of gestational age of 28-44 completed weeks on the basis of last menstrual period combined with ultrasonographic data were included. Pregnant women with normal vaginal delivery, cesarean section and instrumental delivery, primigravida and multigravida both, with premature rupture of membrane were included in the study. All neonates born to these women were also included in the study.

Exclusion Criteria

All mothers who were retropostive, mothers referred from outside hospitals where multiple vaginal examinations had already been done and who were on steroids or other immunosuppressant drugs were excluded from the study. All neonates with Rh isoimmunisation were also excluded from the study.

Laboratory Investigations

All neonates with clinical signs suggestive of sepsis were subjected to septic screening. The sepsis screen used in the study had the following: complete blood count, C-reactive protein(CRP), blood sugar levels (BSL) and serum bilirubin, blood culture and sensitivity. Investigations such as CSF examination, renal function tests, serum electrolytes were done as and when required. A total leukocyte count (TLC) range of less than 5000 and more than 20,000 was considered as suggestive of sepsis. All blood cultures were collected from a peripheral vein with proper aseptic precautions before starting any antibiotic therapy. Approximately, 2ml of blood was inoculated into Tryptose phosphate broth and bile broth and incubated at 37° C. Subcultures were made on blood agar, MacConkey agar and chocolate agar. Growth, if any was identified by the standard bacteriological techniques including gram staining, colony characteristics, biochemical properties and slide agglutination where appropriate. Antibiotic sensitivity of strains was performed using Modified-Kirby-Bauer Disc diffusion method.

Statistical Analysis

Qualitative variables were compared using Chi – square or Fisher's exact test. T-test was used where means of variables needed to be compared. P- value of <0.05 was considered to be significant.

Observations and Results

There were 353 mothers in labour and 357 neonates (4 pairs of twins) included in the study during this period.

Table 1: Maternal Characteristics.

Variables	N (n= 353)	Percentage (%)
Age		
18-25yrs	270	76.48 %
26-32yrs	72	20.39 %
≥33yrs	11	3.11 %
Parity		
Primigravida	95	26.9 %

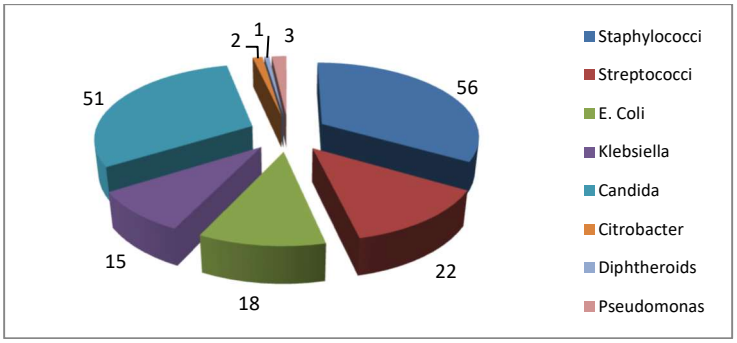
Multigravida	258	73.08%
Gestational Age		
≤ 36 wks	18	5.09%
37-42 wks	283	80.1%
≥ 43 wks	52	14.7 %
Associated disorders		
PIH	37	10.4 %
Hypothyroid	1	.28%
Hepatitis C	1	.28%
Diabetes	3	0.84%
Heart disease	1	.28%
Singleton /Doubleton pregnancy		
Single pregnancy	349	98.8%
Twin pregnancy	4	1.13 %

Out of a total of 353 mothers included in the study, 270 (76.48%) were in the age group of 18-25 years.258 (73.08%) mothers were multigravida. 283 (80%) were in the gestational age of 37-42(term)weeks. PIH was found in 37 (10.4%) mothers and was the most commonly associated disorder.349 (98.8%) mothers had singleton pregnancy. Out of a total of 353 mothers, 160 (45.3%) were culture positive and 193(54.6%) were culture negative. Among the 160 culture positive mothers 152 (95%) mothers had single organisms and 8(5%) mothers had two organisms found in their culture respectively. Therefore a total of 168 organisms were found in mothers having positive culture results.

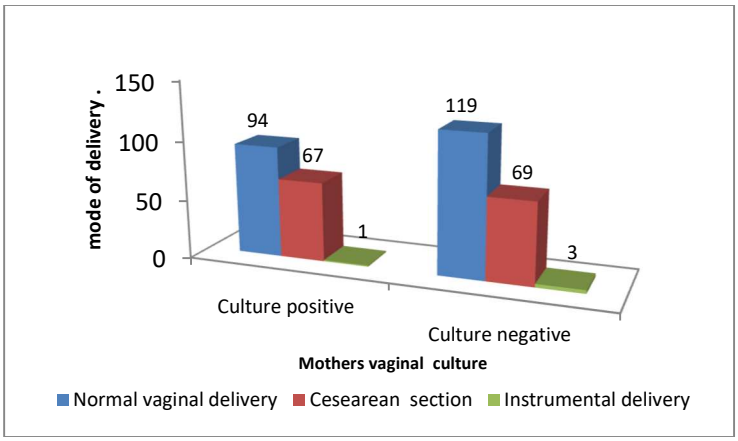
Table 2: Organisms Isolated in Culture Positive Mothers

Sr. No.	Microorganism	Culture positive cases	Percentage
1	Staphylococci	56	35 %
2	Streptococci	22	13.7%
3	E. Coli	18	11.2 %
4	Klebsiella	15	9.3%
5	Candida	51	31.8%
6	Citrobacter	2	1.25 %
7	Diphtheroids	1	0.62 %
8	Pseudomonas	3	1.8%
	Total	168	100 %

Amongst 168 organisms found in the vaginal culture of mothers, staphylococci (35%) and candida (31.8%) were the most commonly found organisms. The other frequently found organisms were streptococci (13.7%), E.coli (11.2%) and klebsiella (9.3%).



Graph 1: Organism Isolated in Culture Positive Mothers



Graph 2: Mode of delivery

213 mothers gave birth by normal vaginal delivery and was the most common mode of delivery. Out of them 94 (44.1%) and 119 (55.8%) were culture positive and culture negative respectively. The Chi-square value is 1.592 and the P value is 0.3253 .The mode of delivery does not have any association with the mothers vaginal culture. In mothers with normal vaginal delivery staphylococci (38.2%) was most commonly found followed by candida (26.5%). None of the organisms have any significant association with the mode of delivery (The Chi-square value is 9.785 and P value is 0.7777) There was a significantly higher proportion of babies with sepsis in culture positive mothers (7.36 %) compared to babies born to culture negative mothers (1.03 %). Out of the 357 babies included in the study, we had suspected sepsis in 32 babies. Only 14(3.92%) of the 32 babies in whom the investigations/clinical signs were supportive of sepsis had been diagnosed as septic babies.

Table 3: Incidence of Sepsis in Our Study and in Our Hospital during the Study Period

	Total number of babies delivered	**Number of babies with sepsis .**	**Incidence**
Present Study	357	14	3.9 %
In the hospital,KIMS during the study period	4743	50	1.05 %

Incidence of neonatal sepsis in our study and in our hospital,KIMS during the study period was 3.9 % and 1.05 %. respectively.

Table 4: Vaginal Culture Result And Sepsis In Newborn

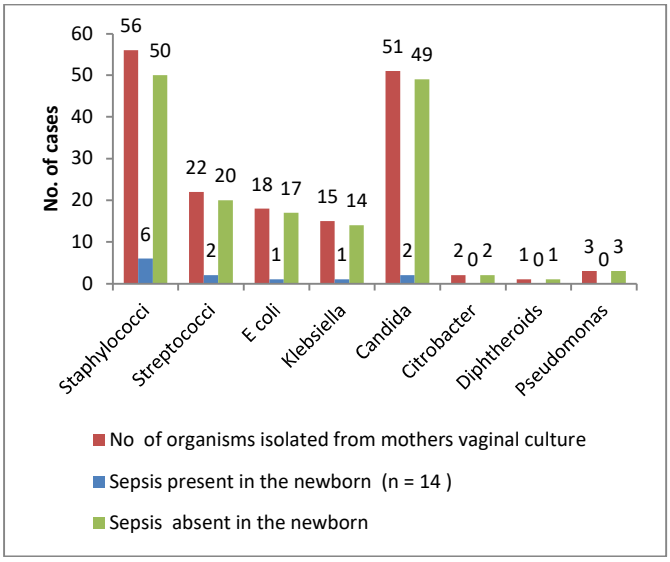
Mothers vaginal culture	**Sepsis present in the Newborn**		**Sepsis absent in the newborn**
	Blood culture positive	**Blood culture negative**	
Vaginal culture positive	8 (4.9%)	4(2.45%)	151(92.6%)
Vaginal culture negative	2 (1.03%)	0 (0%)	192 (99.48%)

Out of 163 babies born to mothers who were vaginal culture positive, 8 (4.9%) babies had blood culture positive proven sepsis while 4 (2.45%) had blood culture negative /clinically proven sepsis and sepsis was absent in 151 (92.6 %) babies. The presence of sepsis in the newborns had a very significant association with a positive vaginal culture in mothers (Fisher's exact –P value was 0.0023)

Table 5: Organism in Vaginal Culture and Sepsis in Newborn

Organism isolated from mothers vaginal culture	**No of organisms isolated from mothers vaginal culture**	**Sepsis present in the newborn (n = 14)**	**Sepsis absent in the newborn**	**Fisher's exact P- value**
Staphylococci	56	6 (42.8%)	50	0.0118
Streptococci	22	2 (14.2%)	20	0.2070
E coli	18	1 (7.1%)	17	0.5179
Klebsiella	15	1 (7.1%)	14	0.4541
Candida	51	2(14.2%)	49	1.0000
Citrobacter	2	0 (0%)	2	1.000
Diphtheroids	1	0(0%)	1	1.000
Pseudomonas	3	0(0%)	3	1.000
No organisms	193	2 (14.2%)	191	0.0044

The highest number (42.8%) of newborns with sepsis were born to mothers who had staphylococcus isolated in their vaginal swab culture. The highest number of organism isolated from the maternal vaginal culture was also staphylococcus.The presence of staphylococcus in the maternal culture was significantly associated with the presence of sepsis in the newborn (The Fisher's exact P value was 0.0118).Similarly the absence of organisms in their vaginal culture was very significantly associated with the absence of sepsis in their newborns (The Fisher's exact P-value was 0.0044)



Graph 3: Organism in Vaginal Culture and Sepsis in Newborns

Table 6: Vaginal Culture in Relation to Early –Onset and Late-Onset Sepsis

Mothers vaginal culture	**Early onset sepsis in newborn**		**Late onset sepsis in newborn**	
	Culture positive	Culture negative	Culture positive	Culture negative
Culture positive	5(41.6%)	2(16.6%)	3 (25%)	2 (16.6%)
Culture negative	1 (50%)	0 (0%)	1 (50%)	0 (0%)

Amongst 12 newborns with neonatal sepsis who's mothers were vaginal culture positive 7 (58.3%) had early –onset sepsis and 5(41.6%) had late onset sepsis. In the early onset sepsis cases 5(41.6%) and 2 (16.6%) newborns and in the late onset sepsis cases 3 (25%) and 2 (16.6%) newborns were proved blood culture positive and blood culture negative respectively.

Table 7: Comparison of Perinatal Factors in Early and Late Onset Sepsis

Perinatal factors	**EOS (n=8)**	**LOS (n= 6)**
Mean BW (SD) (g)	1.7 ± O.56	2.18 ±0.50
Mean GA (SD) (Weeks)	34.7 ± 3.4	38 ± 4.09
Male sex	2 (25%)	1 (16.6%)
Female sex	6 (75%)	5 (83.3%)
Maternal risk factors		
PROM > 12 hr	3 (37.5%)	1 (16.6%)
Maternal pyrexia	3 (37.5%)	1 (16.6%)
Meconium liquor	0 (0%)	0 (0%)
Vaginal culture positive	7 (87.5%)	5 (83.3%)
Mode of delivery		
Normal	5 (62.5%)	4 (66.6%)
LSCS	3 (37.5%)	2 (33.3%)
Instrumental	0	0

Birth weight was significantly lower in babies with EOS (1.7 ± 0.56) than babies with LOS (2.18 ± 0.50).The mean gestational age was also lower in babies with EOS (34.7 ±3.4) than babies with LOS (38 ±4.09).Females were predominantly affected with sepsis than males.3 newborns with early onset sepsis had mothers with a history of PROM >12 hrs and pyrexia before labour. 7 (87.5%) out of 8 newborns with early onset sepsis and 5(83.3%) out of 6 with late onset sepsis had mothers who were vaginal culture positive. The most common mode of delivery in early onset and late onset sepsis was normal vaginal delivery. According to the blood –culture results of babies with sepsis ,staphylococci was the most commonly found organism in newborns in both early

onset (57.14%) and late onset (33.3%) sepsis in newborns. E.coli (28.57%) in early-onset and streptococci (33.3%) in late onset sepsis were the second most common organisms.

Table 8: Antibiotic Sensitivity and Resistance Pattern of Organisms in Neonatal Sepsis

ORGANISM ISOLATED	TIME OF ONSET	NO OF BABIES	AMPI R/ S	AMIKA R/S	ERY R/S	CEF R/S	CEPH R/S	OXA R/S	PEN R/S	VAN R/S
Staphylococci Coagulase Positive	EOS	2	1/1	0/2	2/0	1/1	1/1	0/2	2/0	0/2
	LOS	2	2/0	1/1	1/1	1/1	1/1	0/2	2/0	0/2
Staphylococci Coagulase Negative	EOS	2	2/0	0/2	0/2	1/1	0/2	0/2	2/0	0/2
	LOS	0	-	-	-	-	-	-	-	-
Streptococci	EOS	0	-	-	-	-	-	-	-	-
	LOS	2	2/O	0/2	0/2	NT	1/1	NT	2/0	NT
E.Coli	EOS	2	2/0	0/2	NT	NT	1/1	NT	2/0	NT
	LOS	0	-	-	-	-	-	-	-	-

More than 85% of gram positive isolates showed resistance to ampicillin. Similarly, all gram negative isolates also showed resistance to ampicillin. Vancomycin, oxacillin, erythromycin and amikacin were found to be more effective in gram positive isolates. CoNs(coagulase-negative staphylococcus) was highly resistant to ampicillin and penicillin . 100% of isolates of CoNs as were sensitive to vancomycin., oxacillin and amikacin. Group B streptococcus isolates were 100 % sensitive to amikacin and erythromycin The gram negative isolate, E coli exhibited resistance to ampicillin and high sensitivity to amikacin. Most of the isolates were satisfactorily (50%) sensitive to cephalosporins. Out of 14 neonates with sepsis,7 (50%) were discharged after complete recovery ,5 (35.7%) neonates expired and 2 (14.2%) neonates took discharge against medical advice due to financial constraints. The causes of death of the 5 babies who expired due to sepsis were severe sepsis with sclerema, sepsis with congenital heart disease, sepsis with meningitis and sepsis with shock. 4 and 1 out of the 5 babies had early onset and late onset sepsis respectively.3 of the mother and baby-dyads had the same organism in their respective cultures.4 out of the 5 babies had low birth weight.

Discussion

The neonatal mortality rate, the number of newborns dying in the first 28 days of life per 1,000 live births, is estimated globally to be approximately 23.9.The shared relationship between mothers and their newborns leads to common risk factors and etiologies of infectious diseases and other complications. Intrauterine infection has been shown to play a major role in neonatal infection. Newborns may acquire early –onset neonatal infection ‘ vertically ‘ from endogenous bacteria in the mother’s reproductive tract which may or may not cause disease in the mother but can affect the fetus or the newborn.[2] This study was undertaken to determine the pattern of vaginal tract colonization during labour, the maternal risk factors and their association with neonatal sepsis. In our present study of the pattern of vaginal flora during labour the commonly found organisms were staphylococcus, streptococci, E. coli, klebsiella, candida , diphtheroids, citrobacter and pseudomonas. The most common of these were staphylococcus 56 (35%) in number closely followed by candida 51 (31.8%) . Our findings were almost similar to a study by CC Ekwempu *et al.* [9] where the commonly found organisms were candida, streptococci, staphylococcus , klebsiella, aeromonas, neisseria, lactobacillus. The most common organism was candida albicans (20.9%) in their study. J de Louvois *et al.* [16] in their study on microbial flora in the lower genital tract during pregnancy and its relationship to morbidity found a similar pattern of organisms but lactobacilli was the most prevalent organism .Studies by Bartlett *et al.* [17], Larsen and Galask [8] and Goperlud *et al.* [7] delineated a similar pattern of organisms that reside in the female genital tract. Among the 160 mothers who were vaginal culture positive, 94 (58.7%) had normal vaginal delivery and was the commonest mode of delivery. Early onset sepsis is associated with acquisition of microorganisms from the mother, through transplacental infection or an ascending infection from the cervix or may be caused by organisms that colonize the mothers genitourinary tract. Balaka *et al.* [10] said that newborns are mainly infected by passage through a colonized birth canal at delivery. So delineating the pattern of organisms in women with vaginal delivery should be of more concern. There was no significant association between any of the organisms and the modes of delivery. No other study has evaluated the association of the pattern of organisms in the genital tract during labour in women with different modes of delivery. The most common organism isolated in mothers with vaginal colonization was staphylococcus which was also the same organism commonly isolated in babies with blood culture positive sepsis indicating vertical transmission as also observed by other authors. The presence of staphylococcus in the maternal culture was significantly associated with the presence of sepsis in the newborn (The Fisher’s exact P value was 0.0118).

Similarly the absence of organisms in their vaginal culture was very significantly associated with the absence of sepsis in their newborns (The Fisher's exact P-value was 0.0044). There was a positive association between the organism found in the mothers high vaginal swab and the organism isolated from the blood culture of their newborns who had sepsis. The importance of vertical transmission was suggested by Vidya Ayengar *et al.* [11] as they had documented 24 (1.3 %) mother-baby dyads with vertical transmission of infection. Also important to be considered here is the fact that further typing of these bacteria could not be done due to limitations of our study. Typing of these bacteria would have been of great help in identifying the different strains of these bacteria and to understand which of the strains were more pathogenic. The incidence of sepsis in our hospital was 1.05%. The incidence of neonatal sepsis as reported by the National Neonatal –Perinatal Database report 2002-2003 was only 3.0 % [4]. An overview of the burden of neonatal sepsis in different countries has been provided in the table below

Incidence of neonatal sepsis ;An international perspective[6]

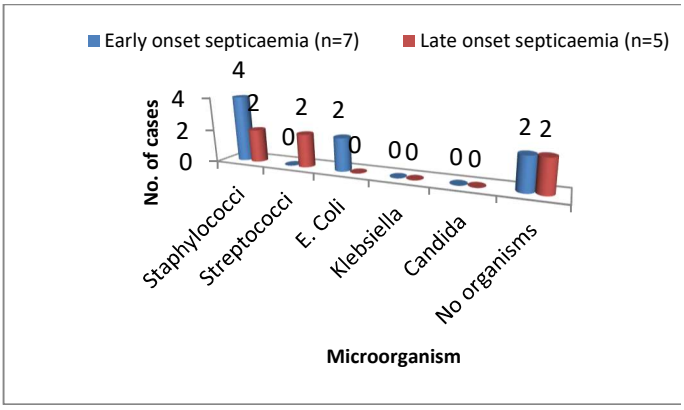
Country	Incidence per 1000/live births.
Asia	7.1 -38
Africa	6.5- 23
South America /Caribbean	3.5 – 8.9
Australia	1.5 - 3.5

Neonatal care settings and practices are very different in different countries. In most African studies, the neonatal population includes mainly term babies looked after in high dependency units, with scarce supportive and monitoring equipment, overcrowding, poor staffing levels and difficulty in providing even basic supportive treatment. In contrast, in many of the developing countries like India, South East Asia, Middle Eastern and Latin America the studies reported are generally carried out in level 3 nurseries ,having a population of mainly premature and lowbirth weight babies and many facilities similar to those in developed countries. These geographical differences are likely to be reflected in different patterns and burden of neonatal sepsis. Different practices of care are also likely to impact on these rates [6] In our study early onset sepsis was observed in 58.3% and late –onset sepsis in 41.6% of newborns of mothers with vaginal colonization .Other studies by Tallur *et al.* [18],report 83.4% and 16.5%,Kuruvilla *et al.* [15] 22.9% and 77.1%,Ayengar *et al.* [11] 73% and 27% for early and late onset sepsis respectively. Kuruvilla *et al.* [15] study group comprised of babies delivered at their institution and treated with prophylactic antibiotics in advance which explains the low incidence of early onset sepsis in their study. In the present study the birth weight was significantly lower in babies with EOS (1.7 ± 0.56) than babies with LOS (2.18 ± 0.50).The mean gestational age was also lower in babies with EOS (34.7 ±3.4) than babies with LOS (38 ±4.09) which signifies that birth weight and gestational age are both significantly associated with the early onset of sepsis. Prematurity and low birth weight are significant risk factors of sepsis as also reported by other studies .[12,19,20,21.] Sepsis is more likely to develop in male infants than females for reasons that are not clear. According to our findings there were 6 (75%) females and 2 (25%) males with early onset sepsis, 5 females (83.3%) and 1 (16.6%) male with late onset sepsis. Females were predominantly affected than males which is in contrast to findings by Vinodkumar *et al.* [19] and S Khurshid *et al.*[20]. Betty Chacko and InderpreetSohi [21] did not observe any increased incidence of sepsis in males in their study. Many authors[19,20,21] have concluded from their studies that PROM and maternal pyrexia were significant perinatal risk factors for neonatal sepsis. Our findings were dissimilar since in our study only 3 (37.5%) mothers of newborns with early onset sepsis had history of PROM > 12 hours and maternal pyrexia was present.7 (87.5%) out of 8 newborns with early onset sepsis and 5(83.3%) out of 6 with late onset sepsis had mothers who were vaginal culture positive. Vinodkumar *et al.* [19] and Grace J Chan[2] had observed the same in their study and concluded that maternal vaginal colonization was a significant risk factor for development of neonatal sepsis. The most common mode of delivery in early onset and late onset sepsis was normal vaginal delivery. Our finding was in concurrence with the studies by other authors [15,19].

Distribution of causative organisms in early and late onset sepsis in various studies

	Studied Cases Of Neonatal sepsis							
	Abdelrahman *et al.*[22]		**S. Khurshid *et al.*[20]**		**Kuruvilla *et al.* [15]**		**Present study.**	
Causative organism	EOS	LOS	EOS	LOS	EOS	LOS	EOS	LOS
Staphylococcus	6(15%)	18(60%)	6(8.8%)	12(29%)	5(17%)	12(12.6%)	4(57%)	2(40%)
Streptococcus	0	0	2 (2.94%)	0	2 (6.6%)	0	0	2 (40%)
E coli	-	1(3.3%)	4(5.8%)	2(4.8%)	7(23.3%)	4(4%)	2(28.5%)	0
Klebsiella	18(45%)	3(10%)	24(35%)	7(17%)	3(10%)	34(36%)	0	0
Pseudomonas	8(20%)	1(3.3%)	0	2(4.8%)	0	4(4%)	0	0

An analysis of distribution of causative organisms of sepsis revealed that gram positive organisms were more common than gram negative organisms. The major causative organism was staphylococcus ,similar to the findings of Abdelrahman *et al.*[22].Other authors [14,25] found Klebsiella and E coli as the most common organism in early onset sepsis. Gram negative organism, Ecoli was only isolated in 2 cases (28.5%) of early onset sepsis in our study. In the present study Staphylococcus and streptococcus were both found in equal numbers in late onset sepsis. Similar to our findings Khurshid *et al.*[20] and Abdelrahman *et al.* [22] reported Staphylococcus to be the common causative organism in late onset sepsis. Kuruvilla *et al.* [15] in contrast found klebsiella to be most common in late onset sepsis. Feng Ying *et al.*[23] in their study found that out of 122 cases of late onset sepsis 85(70 %) had streptococcus isolated from their blood.



Graph 4: Organism Found in Early and Late Onset Sepsis

As neonatal septicemia is considered as a life threatening emergency condition, prompt treatment with antibiotics is necessary. World health organization has recommended the use of penicillin or ampicillin plus an aminoglycoside for neonates. With the advent of third generation cephalosporin, the empirical use of antimicrobial approach for the neonatal septicemia has changed in many centers[25].The appropriate combination of these drugs is being followed in our hospital. In the present study, more than 85% of gram positive isolates showed resistance to ampicillin. Similarly, all gram negative isolates showed resistance to ampicillin. Probably, prolonged antepartum exposure to beta-lactamase (ampicillin and penicillin) may be related with this resistance .Vancomycin and amikacin were found to be more effective in gram positive isolates. The study by Narayan Gyawali [25] has also showed a similar pattern of resistance among gram positive isolates. Colonization of neonates with CoNs is unusual in the first 48 hours after birth. In areas with widespread beta-lactam resistance in CoNs and/or high prevalence of methicillin –resistant staphylococcus aureus, vancomycin is often preferred. Although not as bactericidal as oxacillin ,little resistance has been reported to vancomycin. Aminoglycosides are frequently used in addition and may have a synergistic antistaphylococcal effect when administered with penicillin or vancomycin[26].In our study CoNs was highly resistant to ampicillin and penicillin .Vancomycin was the drug of choice for CoNs as 100% of isolates were sensitive to vancomycin. These findings are in tandem with the study by Ghanshyam *et al.* [24] and the NNPD report 2002-2003[4]. Though all our CoNs isolates were sensitive to erythromycin also , it was not used due to lack of safety data. Group B streptococcus isolates were 100 % sensitive to amikacin and erythromycin. This was in concurrence to the study by Vinodkumar *et al.*[19]. Gram postive isolates were satisfactorily(50%) sensitive to cephalosporins. The gram negative isolate, E coli exhibited resistance to ampicillin and high sensitivity to amikacin. Similar findings have been reported by other authors [4,19].Tessy *et al.* [13] showed a shift of early onset E coli infection from a less fulminant disease caused by ampicillin-sensitive organisms to a more fulminant disease caused by ampicillin-resistant organisms. Increased use of maternal intrapartum ampicillin therapy may account for these changes. Our findings in the present study could also be explained on the same grounds. Low resistance was exhibited by most isolates in the study against the antimicrobials like vancomycin, oxacillin and erythromycin which are not so frequently used antibiotics in clinical settings for neonates.

Limitations of the Study

1 Anaerobic bacteria could not be identified in the study because of the unavailability of facilities for testing these bacteria in the microbiological laboratory.

2 Typing of the bacteria for further identification of different strains and variability of each

bacteria could not be done due to the unavailability of the antisera required for these tests in our microbiological laboratory.

Summary and Conclusion

1. Knowledge of the pattern of maternal genital flora could help us to identify neonates at risk of developing neonatal sepsis.
2. Staphylococcus, Streptococcus, E coli, Klebsiella and Candida were the most common organisms found in the vaginal culture of mothers in labour at our hospital.
3. Incidence of neonatal sepsis was 1.05 % during the study period at our hospital.
4. Early onset-neonatal infection among newborns of mothers with vaginal colonization was significantly higher compared to newborns of mothers without vaginal colonization.
5. Incidence of neonatal sepsis was higher in neonates delivered per vaginally indicating vertical transmission.
6. Low birth weight and prematurity were significant risk factors for developing neonatal sepsis.
7. Organisms causing neonatal sepsis in our (NICU) were Staphylococcus, Streptococci and E coli .
8. The high vaginal swab of most of the mothers of babies with sepsis due to staphylococcus also showed the same organism in their vaginal culture results indicating vertical or perinatal transmission.
9. According to the antibiogram profile of organisms causing neonatal sepsis in our NICU, most of the isolates were resistant to ampicillin. High sensitivity towards vancomycin and amikacin was shown by most of the gram positive isolates.E coli, a gram negative isolate showed high sensitivity only towards amikacin.

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