HIGH VAGINAL SWAB STUDY IN PRETERM LABOUR AND PRETERM PREMATURE RUPTURE OF MEMBRANES AND ITS RELATIONSHIP WITH NEONATAL SEPSIS

Amina Beevi P[1], A. G. Girija Kumari[2], Sudhamani C[3]

[1]Senior Resident, Department of Obstetrics and Gynaecology, Government Medical College, Kozhikode, Kerala.
[2]Professor, Department of Obstetrics and Gynaecology, Government Medical College, Kottayam, Kerala.
[3]Associate Professor, Department of Obstetrics and Gynaecology, Government Medical College, Kozhikode, Kerala.

ABSTRACT

BACKGROUND

Preterm labour is one of the most challenging obstetric complications encountered by obstetricians. Preterm delivery affects one in ten births (11%) and even greater in developing countries. In the United States, preterm birth accounts for approximately 2/3rd of infant deaths. Prematurity is the cause of 80-85% of neonatal morbidity and mortality in developing countries. In India, prematurity is associated with 75% of perinatal mortality. Maternal infections precipitate preterm labour. One of the most important aetiology of preterm labour and preterm premature rupture of membranes is maternal lower genital tract infection and this leads to increased neonatal infections.

The objectives of the study were- 1) To find out the prevalence of vaginal infection in preterm labour and preterm premature rupture of membranes. 2) To find out the incidence of neonatal sepsis and its relationship with vaginal swab culture in preterm premature rupture of membranes and preterm labour. 3) To identify common organisms in high vaginal swab culture and its antimicrobial sensitivity.

MATERIALS AND METHODS

This is a prospective observational study conducted for 12 months from October 2016 to October 2017, in the department of Obstetrics and Gynaecology, Government Medical College, Kottayam. In a previous study by Taralekar Vaishali et al, incidence of genital tract infection in preterm labour was about 59%. For the present study, a conservative estimate of 50% is used in sample size calculation and it is n=105. Statistical analysis is done by using SPSS software.

RESULTS

Out of the 105 subjects included in this study, 74 were diagnosed with preterm premature rupture of membranes (70.5%), and 31 (29.5%) with spontaneous preterm labour. Majority were in the age group of less than 22 years. Regarding gestational age, 61.9% came under late preterm gestation, majority being primigravidae. Of the 20% positive cultures, most common organism isolated was E.coli (8.6%) and prevalence of genital infection in spontaneous labour and preterm premature rupture of membranes were 16.1% and 21.6% respectively. Most of the isolated organisms were resistant to Ampicillin and sensitive to Cephaperazone-Sulbactum. In this study, 34.3% had neonatal sepsis, 51.4% needed neonatal intensive care and 15.2% succumbed to neonatal death. In 21 positive swab cultures, 88% had neonatal sepsis and 47% died during neonatal period. Relation between the birth weight and neonatal sepsis was statistically significant.

CONCLUSION

Maternal genital tract infection is one of the causes of preterm labour and preterm premature rupture of membranes. The dictum "prevention is better than cure" applies very well to the management of preterm birth.

KEYWORDS

HVS, PPROM, NICU, WHO, FIGO, NND, CRP.

HOW TO CITE THIS ARTICLE: Amina Beevi P, Girija Kumari AG, Sudhamani C. High vaginal swab study in preterm labour and preterm premature rupture of membranes and its relationship with neonatal sepsis. J. Evid. Based Med. Healthc. 2018; 5(30), 2249-2254. DOI: 10.18410/jebmh/2018/466

BACKGROUND

World Health Organization (WHO) and the International Federation of Gynaecology and Obstetrics (FIGO) defined that the preterm infants are those delivered before 37 weeks of completed gestation. Preterm birth is subdivided in to (1) early preterm - birth before 33 weeks 6 days, (2) late preterm – birth between 34 - 37 weeks. Late preterm births comprise 70% of all preterm births. Preterm premature rupture of membranes (PPROM) is defined as spontaneous rupture of the fetal membranes before 37 weeks of gestation and before the onset of labour.[1]

Preterm labour is one of the most challenging obstetric complications encountered by obstetricians. Preterm delivery affects one in ten births (11%) and the percentage is even greater in developing countries. In United States,

Financial or Other, Competing Interest: None.
Submission 03-07-2018, Peer Review 07-07-2018,
Acceptance 13-07-2018, Published 23-07-2018.
Corresponding Author:
Dr. Sudhamani C,
'Gulmohar', Karanthur,
Kunnamangalam,
Kozhikode, Kerala.
E-mail: sudhamen@gmail.com
DOI: 10.18410/jebmh/2018/466

preterm birth accounts for approximately 2/3rd of infant deaths.[2] Prematurity is the cause of 80-85% of neonatal morbidity and mortality in developing countries.[1] In India, prematurity is associated with 75% of perinatal mortality.[3]

While most of the causes of preterm labour are not known, various conditions have shown to be associated with an increased risk of preterm delivery. Four main causes of preterm birth are (1) spontaneous preterm labour, with intact foetal membranes – 40%, (2) PPROM – 30%, (3) iatrogenic and delivery for maternal and foetal indications accounting for 20 – 30%, (4) twins and higher order multi foetal birth. Despite considerable advances in obstetric care, preterm labour continues to be a major cause of perinatal morbidity and mortality.[1] Studies indicate that one of the most important aetiologies of preterm labour and PPROM is maternal lower genital tract infection and the risk of neonatal infection is increased among mothers whose genital tract is colonized with Group B- Streptococci and other virulent microbial organisms. Infection in early gestation may lead to congenital anomaly, abortions and altered foetal growth whereas preterm labour, PPROM and neonatal sepsis are more prevalent in late onset group.[4] Significant progress has been made in the care of premature babies, but not in reducing the prevalence of preterm birth.[5] All neonates suspected to have sepsis, should undergo a sepsis screening to corroborate the diagnosis and include total leukocyte count (<5000/mm^3), absolute neutrophil count (low count), positive CRP (C- reactive protein) >2 mg/dl.[3] Antibiotics have become an important part of the recent treatment for PPROM and preterm labour, which prolong the pregnancy and decrease maternal and neonatal morbidity and mortality.[6]

This study is about the relation of preterm labour and PPROM to maternal genital tract infections and it's association to neonatal sepsis. Also, it aims to find out the common organisms in maternal lower genital tract infection in preterm labour and PPROM, and their antibiotic sensitivity.

Objective

1. To find out the prevalence of vaginal infection in preterm labour and PPROM
2. To find out the incidence of neonatal sepsis and it's relationship with vaginal swab culture in PPROM and preterm labour in present study
3. To identify common organisms in high vaginal swab culture and it's antimicrobial sensitivity

MATERIALS AND METHODS

This is a prospective, observational study conducted for 12 months from October 2016 to October 2017, in the Department of Obstetrics and Gynaecology, Government Medical College, Kottayam, Kerala. Antenatal women with gestational age between 28 weeks to 36 weeks 6 days, who got admitted with preterm labour or PPROM in labour room were selected. Above population with singleton foetus with cephalic presentation and no other medical, surgical or obstetric complications were included.

Pregnant women with gestational age <28 weeks and >37 weeks, those with obstetric and other medical complications like oligohydramnios, polyhydramnios, IUD, foetal anomaly, gestational diabetes, abnormal presentation, placenta praevia, gestational hypertension, multifoetal gestations and those induced for maternal and foetal indications were excluded.

Sample Size

From a previous study by Taralekar Vaishali et al, genital tract infection in preterm was about 59%. In this study, a conservative estimate of 50% is used in sample size calculation and it is n= 105. Approval from the Institutional Scientific and Ethics committee was taken, and informed consent was obtained. Gestational age was confirmed by LMP and first trimester USG. The study variables are maternal age, obstetric score, gestational age, spontaneous preterm labour or PPROM, culture positivity, antibiotic sensitivity of isolated organisms, baby birth weight, neonatal sepsis and neonatal death.

Statistical Analysis

Statistical analysis is done by SPSS software data variable. The categorical variables are analysed by the Chi square test and p value < 0.05 is taken as statistically significant.

Limitation of the Study

Micro-organisms like Gardnerella vaginalis, Fusobacterium, Mycoplasma hominis and Ureaplasma urealyticum causing female genital tract infections cannot be detected by conventional culture method. In the Department of Microbiology at Government Medical College, Kottayam, routine swab cultures are done in MacConkey's medium. Group B Streptococci will not grow in this medium. Hence prevalence of culture positivity in this group will be less.

RESULTS

Total of 105 subjects were included in this study and out of these 74 patients were diagnosed with PPROM (70.5%), and 31 (29.5%) patients were with spontaneous preterm labour.

Maternal Age			Obstetric Score		
Yrs.	**No**	**%**	**Gravida**	**No**	**%**
<22	31	29.5	G1A0	43	41.0
22-25	27	25.7	G2A1/G3A2	26	24.8
25-30	27	25.7	G2P1	30	28.5
>30	20	19.0	>G3P2	6	5.7
Total	**105**	**100**	**Total**	**105**	**100**

Table 1. Maternal Age and Obstetric Score

Gestational Age (Weeks)	No. of Cases	%
>28 – 33.6	40	38.1
>34 – 36.6	65	61.9
Total	**105**	**100**

Table 2. Gestational Age

Swab Culture Report	No	%
Sterile	84	80.0
E. coli	9	8.5
Klebsiella	5	4.8
Staphylococcus aureus	4	3.8
Candida	2	1.9
MRSA®	1	1.0
Total	**105**	**100**

Table 3. Vaginal Swab Cultures

®Methicillin resistant Staphylococcus aureus

Diagnosis		Positive Culture	Sterile Culture	Total
PPROM	No. of swabs	16	58	74
	% of swabs	15.23	55.23	70.5
Preterm Labour	No. of swabs	5	26	31
	% of swabs	4.7	24.76	29.5
Total	No. of swabs	21	84	105
	% of swabs	20	80	100

Table 4. Vaginal Swab Reports in Preterm Labour / PPROM

Antimicrobial Agent	Sensitivity	No. of Cases	%
Penicillin	No	18	85.7
	Yes	3	14.2
1st & 2nd Generation Cephalosporins	No	17	80.9
	Yes	4	19.1
3rd & 4th Generation cephalosporins	No	10	47.6
	yes	11	52.4
Erythromycin	No	9	42.8
	Yes	12	57.1
Gentamycin	No	6	28.57
	Yes	15	71.42
Amikacin	No	4	19.1
	yes	17	80.9
Clotrimazole	No	18	85.7
	yes	3	14.2
Ciprofloxacin	No	11	52.4
	yes	10	47.6
Cefaperazone-Sulbactum	No	3	14.2
	Yes	18	85.7

Table 5. Antibiotic Sensitivity in Positive Culture

Antibiotic Sensitivity	E. coli %	Klebsiella %	Staphylococcus aureus %	MRSA %	Candida %
Penicillins	11.1	0	50	0	0
1st & 2nd generation cephalosporins	11.1	0	50	0	0
3rd generation cephalosporins	50	0	75	0	0
Erythromycin	66.6	66	75	0	0
Gentamycin	66.6	100	75	100	0
Amikacin	88.8	100	100	100	0
Ciprofloxacin	33.3	60	50	100	50
Cephaperazone-Sulbactum	88.8	100	75	100	0
Clotrimazole	0	0	0	0	100

Table 6. Antibiotic Sensitivity of Respective Organism Isolated

Baby Weight		Sepsis Features		Total
		No	Yes	
<1 Kg	Count	0	5	5
	%	0.0	4.76	4.76
1 Kg-1.4 Kg	Count	7	14	21
	%	6.6	13.3	19.9
1.5-2.5 kg	Count	44	17	61
	%	41.9	16.19	58.1
>2.5 Kg	Count	18	0	18
	%	17.1	0.0	17.1
Total	Count	69	36	105
	%	100	100	100

Table 7. Association between Birth Weight and Neonatal Sepsis

$X^2 = 36.08$ p=.000; p <0.05

Analysing baby weight and neonatal sepsis, all the 5 babies with birth weight <1kg showed features of neonatal sepsis (100%) while 14 babies were affected in group with weight between 1-1.4 kg and none were affected with weight >2.5 kg.

		No. of babies	% of Total babies
Sepsis	Absent	69	65.7
	Present	36	34.3
NICU admission	No	51	48.6
	Yes	54	51.4
NND	Absent	89	84.8
	Present	16	15.2

Table 8. Sepsis / NICU Admission / NND

Incidence of neonatal sepsis, NICU admission and neonatal deaths in this study were 34.3%, 51.4% and 15.2% respectively. Most of the babies of NND were extremely preterm.

Maternal Age (Yrs.)		Membrane Status		Total
		Absent	Present	
< 22	No	24	7	31
	%	22.9	6.7	29.5
22 – 25	No	17	10	27
	%	16.2	9.5	25.7
25 – 30	No	17	10	27
	%	16.2	9.5	25.7
>30	No	16	4	20
	%	15.2	3.8	19.0
Total	No	74	31	105
	%	70.5	29.5	100

Table 9. Maternal Age and Preterm Labour

$X^2 = 3.17$; p=0.376 p >0.05

No statistical association between maternal age, preterm labour and PPROM.

Obstetric Score		PPROM	Preterm Labour	Total
G1 A0	No	31	12	43
	%	29.5	11.4	41.0
G2A1	No	20	6	26
	%	19.04	5.71	24.8
G2P1	No	18	12	30
	%	17.14	11.42	28.6
>G3P2	No	5	1	6
	%	4.76	0.95	5.7
Total	No	74	31	105
	%	70.46	29.5	100

Table 10. Obstetric Score and Preterm Labour

$X^2 = 2.62$; p=0.453 p>0.05

There is no association between obstetric score and preterm labour.

Neonatal Sepsis		Vaginal Swab Reports		
		Sterile	Positive	Total
Absent	No. of swabs	63	6	69
	%	60	5.7	65.7
Present	No. of swabs	21	15	36
	%	20	14.3	34.3
Total	No. of swabs	84	21	105
	%	80	20	100

Table 11. HVS Culture and Neonatal Sepsis

$X^2 = 23.03$; p=0.00005.

On analysing, the relationship between the HVS culture positivity and neonatal sepsis is statistically significant.

Sepsis		Swab Culture						Total
		Sterile	E. coli	Klebsiella	Staph. Aureus	Candida	MRSA	
Absent	No	63	1	2	1	2	0	69
	%	60	0.95	1.9	0.95	1.9	0.0	65.7
Present	No	21	8	3	3	0	1	36
	%	20	7.61	2.85	2.85	0.0	0.95	34.3
Total	No	84	9	5	4	2	1	105
	%	80	8.56	4.75	3.8	1.9	0.95	100

Table 12. Positive Swab Culture and Neonatal Sepsis

Neonatal sepsis was found in 88% of E coli and 100% of MRSA positive cultures.

NND		HVS		
		Sterile	Positive	Total
Absent	No. of swabs	78	11	89
	%	74.28	10.47	84.8
Present	No. of swabs	6	10	16
	%	5.71	9.52	15.2
Total	No. of swabs	84	21	105
	%	80	20	100

Table 13. NND and HVS Culture Reports

$X^2 = 21.76$; p=0.001 p<0.05.

Sixteen out of 105 babies died during early neonatal period. Of 21 swab positives, 10 babies died during neonatal period (47%). Of 84 swab negatives, 6 babies died in neonatal period (7.1%).

Baby Weight(kg)		NND		Total
		No	Yes	
< 1 Kg	No	0	5	5
	%	0.0	4.76	4.8
1 – 1.4 Kg	No	11	10	21
	%	10.47	9.52	20.0
1.5 – 2.5 Kg	No	60	1	61
	%	57.14	0.95	58.1
>2.5 Kg	No	18	0	18
	%	17.1	0.0	17.1
Total	No	89	16	105
	%	85	15	100

Table 14. Birth Weight and NND

X^2 =50.6 p=.000; p<0.05.

There is a statistically significant relation between birth weight and neonatal sepsis.

			Sepsis Features		Total
			Absent	Present	
Baby Sex	F	No	35	20	55
		%	33.3	19.0	52.4
	M	No	34	16	50
		%	32.4	15.2	47.6
Total		No	69	36	105
		%	65.7	34.3	100

Table 15. Baby Sex and Neonatal Sepsis

X^2=.070; p=.791 p>0.05.

Baby sex and neonatal sepsis is statistically not significant.

DISCUSSION

Total number of deliveries of the institution during the study period was 5349, of which 570 were preterm deliveries (10.66%). The percentage of preterm birth was 11.7 in UK in 2011 as per Mathews TJ et al.[2] The incidence of preterm birth has been estimated to be 12-13% in United States in 2011.[1] In India, preterm labour accounts to 7 – 10% of total births.[7] The percentage of preterm birth in the present study is comparable with other studies.

A study by Martin JA, Hamilton BE et al showed that in US, late preterm contributes to over 70% of preterm births.[8] In this study, out of 105 preterm deliveries, 40(38.1%) were early preterm and 65 (61.95%) were late preterm deliveries and comparable between studies.

In the present study, 29.5% were under the age group of 22 years and 19% were above 30 years of age and thereby showing no relation between maternal age and preterm birth. Primigravida were 43(41%), G_2A_1 26(24.8%), G_2P_1 23(28.6%), G_3P_2 or more 6(5.7%). No statistical association between obstetric score and preterm birth.

Analysing 105 vaginal swabs, 31 were obtained from spontaneous preterm labour and 74 from PPROM. Eighty-four (80%) were culture negative and 21(20%) were culture positive. In the present study, prevalence of maternal genital tract infection in preterm labour is 20% and the same is 21.6% and 16.1% in PPROM and spontaneous preterm labour respectively.

Among the 21 cultures found positive, the most common organisms isolated were E coli (8.6%), Klebsiella (4.8%), Staph. aureus (3.8%), Candida (1.9%) and MRSA (1%). Group B Streptococcus was not isolated. Study by Goncalves LF et al[9] showed that positive vaginal swab cultures were obtained for 10-40% of preterm labour. An Indian study by Taralekar et al[10] states that infection is 2-3 times more common in patients with PPROM as compared to PROM. HVS cultures showed microbial growth in 60% cases of threatened preterm labour and 56% cases of PPROM and concluded that vaginal infections increase the incidence of preterm labour and PPROM. Commonest organisms isolated in the study were comparable.

C. Karat et al[11] in a case control study found that E coli and Staph aureus were significantly associated with occurrence of PPROM. Group B Streptococcus and P Mirabilis were also isolated though not found in control group. Erich Hafner et al[12] found a significant association between Streptococci colonization and preterm birth. A study by Krychowska-Cwikla A et al[13] showed high virulence bacteria in vaginal swab culture and a statistical significance. Enterococci occurred significantly more frequently in pregnancies complicated with PPROM. Bacterial vaginosis and E coli are independent risk factors for preterm labour and PPROM.

Dr. T. Seshasai et al[14] in their study of endocervical swab cultures in preterm labour and PPROM, found 41.7% as culture positive. The organisms isolated were comparable with the present study, E coli being the commonest microbial isolated and sensitive to Amikacin and Gentamycin. Cephalexin was found to be effective against most of the organisms including E coli by Taralekar Vaishali et al.[10] In our study most of the strains were sensitive to Cefaperazone-Sulbactum, Klebsiella and MRSA being 100% sensitive. Taralekar et al[10] shows that 90% of threatened preterm cases are conserved by prompt treatment of infection. Most strains were resistant to Penicillins. From the more recent studies, Ampicillin resistant organisms are more common in maternal genital infections and associated with early neonatal sepsis.[10]

Some organisms like Gardnerella vaginalis, Fusobacterium, Mycoplasma hominis and Ureaplasma urealyticum are detected more frequently than others in amniotic fluid of women with preterm labour.[15,16] But these organisms cannot be detected by conventional culture methods. In the present study HVS reports didn't show Group B Streptococci as in our Department of Microbiology, routine swab cultures are done in MacConkey's medium and hence prevalence in this study will be less than the actual.

The incidence of neonatal sepsis is 34.3% (36) and 54(51%) babies needed NICU care. Among 21 culture positive mothers, 15 babies had sepsis (71.42%). E coli and MRSA positive cultures had associated neonatal sepsis in 88% and 100% babies respectively. Stoll BJ et al[17] found a reduction in Group B Streptococcal sepsis and increase in E coli with no change in the overall rate of early onset sepsis. Neonates with early onset sepsis were more likely to die, especially when infected with coliforms.[10,17]

There were 16(15.2%) neonatal deaths in this study and all had features of sepsis. Maternal genital tract infection was associated with 71.4% NND (10 /16 babies). David P. Vander Ham et al[18] observed 3.4% neonatal sepsis with PPROM. In this study, there is a relation between baby weight, neonatal sepsis and neonatal death. Extreme preterm and low birth weight babies are more susceptible to sepsis. All 5 babies with weight <1 kg. succumbed to neonatal death due to sepsis. Though study by Stoll BJ et al[19] have challenged this finding, evidences show that very small newborns are at increased risk for sepsis. On analysing this study, there is no statistically significant relation of baby sex with neonatal sepsis and death. Many studies showed that male babies have two fold increased risk for neonatal sepsis.

CONCLUSION

Maternal genital tract infection is one of the causes of preterm labour and PPROM. Prevalence of lower genital tract infection in the present study is 20% and is associated with early neonatal sepsis and neonatal death. Incidence of neonatal sepsis observed is 34.3% and 47.6% of this is found in babies whose mother had lower genital tract infections. Most common organism isolated in HVS was E. coli. Most of the organisms were sensitive to Cefaperazone-Sulbactum and resistant to Penicillin group of antimicrobials including Ampicillin.

REFERENCES

[1] Cunningham FG, Leveno KJ, Bloom SL, et al. Williams obstetrics. 24th edn. McGraw-Hill Education 2014.

[2] Matthews TJ, MacDorman MF. Infant mortality statistics from the 2010 period linked birth/infant death data set. Natl Vital Stat Rep 2013;62(8):1-26.

[3] Ali Z. Selection criteria in the NICU: who should get effective critical care? Indian J Med Ethics 2009;6(2):103-104.

[4] Cohen-Wolkowiez M, Moran C, Benjamin DK, et al. Early and late onset sepsis in late preterm infants. Pediatr Infect Dis J 2009;28(12):1052-1056.

[5] McCormick MC. The contribution of low birth weight to infant mortality and childhood morbidity. N Engl J Med 1985;312(2):82-90.

[6] Mercer BM, Miodovnik M, Thurnau GR, et al. Antibiotic therapy for reduction of infant morbidity after preterm premature rupture of the membranes. A randomized controlled trial. National Institute of Child Health and Human Development Maternal-Fetal Medicine Units Network. JAMA 1997;278(12):989-995.

[7] American College of Obstetricians and Gynecologists: Practice bulletin no. 130: prediction and prevention of preterm birth. Obstet Gynecol 2012;120(4):964-973.

[8] Martin JA, Hamilton BE, Sutton PD, et al. Births: final data for 2005. Natl Vital Stat Rep 2007;56(6):1-103.

[9] Gonçalves LF, Chaiworapongsa T, Romero R. Intrauterine infection and prematurity. Mental Retardation and Developmental Disabilities Research Reviews 2002;8(1):3-13.

[10] Vaishali T, Salil B, Girija W, et al. Vaginal infection as a cause for preterm labour, PPROM, PROM. Int J Adv Res 2014;2(7):1092-1096.

[11] Karat C, Madhivanan P, Krupp K, et al. The clinical and microbiological correlates of premature rupture of membranes. Indian J Med Microbiol 2006;24(4):283-285.

[12] Hafner E, Sterniste W, Rosen A, et al. Group B streptococci during pregnancy: a comparison of two screening and treatment protocols. Am J Obstet Gynecol. 1998;179(3 Pt 1):677-681.

[13] Krychowska-Ćwikła A, Dmoch-Gajzlerska E. Analysis of microorganisms isolated from cervical canal of pregnant women giving birth prematurely in Poland. Prog Health Sci 2011;1(1):63-72.

[14] Seshasai T, Sukanya S. Bacteriological study of endocervix in preterm labour and preterm premature rupture of membranes. IJSR 2015;4(7):915-919.

[15] Hillier SL, Martius J, Krohn M, Kiviat N, et al. A case-control study of chorioamnionic infection and histologic chorioamnionitis in prematurity. N Engl J Med 1988;319(15):972-978.

[16] Yoon BH, Romero R, Park JS, et al. Microbial invasion of the amniotic cavity with Ureaplasma urealyticum is associated with a robust host response in fetal, amniotic, and maternal compartments. Am J Obstet Gynecol 1998;179(5):1254-1260.

[17] Stoll BJ, Hansen N, Fanaroff AA, et al. Changes in pathogens causing early-onset sepsis in very-low-birth-weight infants. N Engl J Med 2002;347(4):240-247.

[18] van der Ham DP, van der Heyden JL, Opmeer BC, et al. Management of late-preterm premature rupture of membranes: the PPROMEXIL-2 trial. Am J Obstet Gynecol 2012;207(4):276-e1-10.

[19] Stoll BJ, Hansen NI, Bell EF, et al. Neonatal outcomes of extremely preterm infants from the NICHD Neonatal Research Network. Pediatrics 2010;126(3):443-56.