Early- and Late-Onset Neonatal Sepsis: Risk Factors and Outcome Study

Assistant professor Dr. Zuhair Omran Easa

**Medical college-Karbala University*

Abstract

Background: Early-onset neonatal infections are acquired before or during delivery (vertical mother-to-child transmission). Late-onset infections develop after delivery from organisms acquired in the hospital or the community. The age at onset depends on the timing of exposure and virulence of the infecting organism. Very-late-onset infections (onset after 1 mo of life) may also occur, particularly in VLBW preterm infants or term infants requiring prolonged neonatal intensive care.

Aims of the study: To study the effect of some risk factors, laboratory investigations and outcome on early and late onset neonatal sepsis and their relation with early and late onset neonatal sepsis.

Patients and methods: A prospective study was carried out in Karbala teaching hospital from the first of January 2011 through the first of January 2012 for a group of 150 neonates who were subgrouped into 86 (57.3%) with early onset neonatal sepsis and 64 (42.7%) with late onset neonatal sepsis. Information were taken about the patients regarding the age in days, gender, perinatal and maternal history and blood samples were aspirated for complete blood count, blood culture, and C-reactive protein.

Results: We found that the neonate is more susceptible to early onset neonatal sepsis in comparison to late onset neonatal sepsis. Premature neonates were more liable for late onset neonatal sepsis than full term ones, 93 vs. 57 neonates (62% vs. 38%). Neonates of mothers with prolonged rupture of membranes were more liable for early onset neonatal sepsis than late onset neonatal sepsis, 27 neonates vs. 11 neonates (31.3 vs.17.2) respectively. Also we found that the blood culture result highly affecting by antibiotics used before culture. The number of neonates with history of maternal fever during pregnancy or shortly before labor was associated with higher number of early onset neonatal sepsis than late onset sepsis, 20 vs. 16 neonates (13.3% vs. 10.7%) respectively. The mortality rate is more in ENS in comparison to late type.

Conclusion: Prematurity and prolonged rupture of membranes had significant association with early onset neonatal sepsis while gender, antenatal care and maternal fever had no difference between early onset and late onset neonatal sepsis. Incidence and mortality of early onset neonatal sepsis was higher than late onset neonatal sepsis. Immature: total neutrophils count, and platelets count had high sensitivity to the diagnosis of neonatal sepsis

Introduction

Early-onset neonatal infection is usually due to vertical transmission by ascending contaminated amniotic fluid or during vaginal delivery from bacteria colonizing or infecting the mother's lower genital tract. As a result, the risk for sepsis increases from 1 to 4 percent in neonates born to mothers with chorioamnionitis. Maternal GBS bacteriuria during the current pregnancy, prior delivery of an infant with GBS disease, and maternal colonization are risk factors for early-onset GBS sepsis [1]. Late-onset neonatal infections can be acquired by the two following mechanisms: Maternal vertical

transmission, result-ing in initial neonatal colonization that evolves into later infection. Horizontal transmission from direct contact with care providers or environmental sources. Disruption of the intact skin or mucosa, which can be due to invasive procedures (eg, intravascular catheter), increases the risk of late-onset infection. Late-onset sepsis is un-commonly associated with maternal obstetrical complications. Risk factors can include use of forceps during delivery or electrodes placed for intrauterine monitoring, which penet-rate the neonatal defensive epithelial barriers of the skin and mucosa [2].Metabolic factors, including hypoxia, acidosis, hypothermia, and inherited metabolic disorders (eg, galactosemia), are likely to contribute to risk for and severity of neonatal sepsis. These factors are thought to disrupt the neonate's host defenses (ie, immunologic response) [2, 3].The aim of this work is to study the relationship and some risk factors associated with early onset and late onset neonatal sepsis regarding gender, prematurity, mode of delivery, midwife interfere-nce, prolonged rupture of membranes, maternal fever, antenatal care, incidence of blood culture and causative organisms, antibiotics, some laboratory investigations, WBC, absolute neutronphil count, platelets count, C-reactive protein, immature: total neutrophil counts and study the outcome between the two presentations.

Patients & Methods

This is a prospective, hospital-based, single- centre study. It has been carried out in the neonatal care unit and in the hospital wards of Kerbala'a Teaching Hospital for children during the time period from January 1, 2011 through January 1, 2012.A total of 150 neonates (less than 30 days old) were selected with clinical features sugges-tive of sepsis such as poor feeding /inability to feed, respiratory distress, lethargy , apnea, cyanosis , or presence of risk factors of sepsis such as prematurity, home delivery, maternal fever during pregnancy or during labor, prolonged rupture of membranes (rupture of membranes of more than 18-hour duration), meconiumstaining. Neonates were divided according to age of onset of presumed sepsis into two groups: EONS group of age range from birth to 7 days, with a mean age of 3.3 days and total number of 86 (57.3%) of total. LONS group of age range from 8 to 30 days, with a mean age of 17 days and total number of 64 (42.7%) of total. We asked about patient's name, age in days, gender, mode of delivery, delivery place, maturity (depending on new Ballard criteria for assessment of gestational age and on ultrasonography monitor-ing), meconium stained amniotic fluid, and maternal history of fever (fever had occurred during pregnancy or shortly before labor), history of prolonged rupture of membranes (18 hours or more), birth weight, whether antibiotics has been used or not before investigations and other information regarding results of blood tests and other important points from the focused history and physical examin-ation. Also we followed up the patients and recorded the outcome. From 171 neonates initially included in our study, 21 neonates were excluded because they were presented with the following clinical pictures: Nine neonates resumed breast feeding successfully within 5-9 hours after admission, i.e. there is dramatic improvement within a short time. Seven neonates were jaundiced and presented with poor feeding and were admitted for phototherapy and improved within 24-hours of returning of serum bilirubin to normal level. Five patients were hypothermic during admission, and that picture was confusing to us and gave us false impression about neonatal sepsis and they were improved a short time after admission in the incubator. Two samples of blood were collected and sent for CBC (blood with anti-coagulant), and for CRP

(clotted blood).Under strict aseptic measures (using of gauze after wearing gloves with the use of povidone iodine, and then we sterilized (three times) the site from which we need to aspirate the sample for blood culture under direct supervision, samples of blood (2-2.5 ml) were added to a bottle containing 8ml of Brain-Heart infusion broth. The cut off values of the studied parameters for positive tests were: Platelets count >450000/cmm or < 150000/cmm; White blood cell count (WBC) < 5000/cmm or > 21000/cmm. Lymphocyte percent normal range is 31%; neutrophil percent normal range is 40-80%; absolute neutrophil count (ANC) an age adjusted normal reference range was used and neutrophilia were considered abnormal. Immature: total neutrophil ratio I:T of 20% or more was considered abnormal. Data analysis was computer aided. An expert statistical advice was sought for. Statistical analyses were done using SPSS version 13 computer software (Statistical Package for Social Sciences P value of < 0.01 is considered is highly selective; <0.05 is considered as significant; > is considered as not significant.

Results

There was no significant difference in gender, maternal fever and antenatal care, between EONS and LONS groups, (*p* value > 0.05), table 1. Prematurity, as a risk factor was higher in EONS group compared to LONS group (44.2%, vs. 29.7%) respectively; and the difference was significant (*p* = 0.042), table 1.Mode of delivery showed no effect in EONS group (NVD = C/S = 50%); while it seems to have higher effect on LONS group as (73.4% vs 26.6%) of patients are C/S, (*p* = 0.003), table 1.

Table 1. Frequency distribution of possible risk factors and theoutcome of early and late onset neonatal sepsi

Characteristics	Values	Early onset neonatal Sepsis N= 86 (percent)	Late onset neonatal Sepsis N=64 (percent)	P Value
Gender	Male	52 (60.5%)	35 (54.7%)	>0.05
	Female	34 (39.5%)	29 (45.3%)	
Prematurity	Premature	38 (44.2%)	19 (29.7%)	<0.05
	Full term	48 (55.8%)	45 (70.3%)	
Mode of delivery	NVD	43 (50.0%)	47 (73.4%)	<0.05
	C/S	43 (50.0%)	17 (26.6%)	
Place of delivery	Hospital	75 (87.2%)	47 (73.4%)	<0.05
	Home	11 (12.8%)	17 (26.6%)	
Midwife interference	Yes	16 (18.6%)	23 (35.9%)	<0.05
	No	70 (81.4%)	41 (64.1%)	
Prolonged rupture of membranes	Yes	27 (31.3%)	11 (17.2%)	<0.05
	No	59 (68.7%)	53 (82.8%)	
Maternal fever	Yes	20 (23.3%)	16 (25%)	>0.05
	No	66 (76.7%)	48 (75%)	
Antenatal care	Good	20 (23.2%)	8 (12.5%)	>0.05
	Poor	36 (41.9%)	35 (54.7%)	
	None	30 (34.9%)	21 (32.8%)	
Outcome	Died	16 (18.6%)	2 (3.1%)	<0.01

Only 11 (12.8%) of neonates with EONS were born at home, while 17 (26.6%) of neonates with LONS were home delivery, (p = 0.048). Midwife interference was more effective in LONS subjects than EONS subjects, (35.9%, vs. 18.6%) respectively, (*p* = 0.019), table 1.Prolonged rupture of membrane was significantly more frequent in EONS subjects than LONS subjects (31.3% vs. 17.2%) respectively, (*p* = 0.028), table 1.Death was more frequent in EONS subjects than LONS subjects; (18.6%, vs. 3.1%) respectively, (*p* = 0.004), table 1 There was a significant difference in the type of causative organisms between the two groups; the most common organism in EONS subjects was the Enterobacter (29.1%), while the least common was the *Escherichia coli* species. In comparison, the most common organism in the LONS group was the *Staphylococcus epidermidis* species (20.3%), (*p* = 0.007), table 2.

Table 2. Frequency distribution of causative organisms in early and late onset neonatal sepsis

Type of Organism	Early onset neonatal sepsis	Late onset neonatal sepsis	P Value
	No.=86 (percent)	No.=64(percent)	
E-coli	0 (0.0%)	4 (6.3%)	0.007
Proteous	1 (1.2%)	0 (0.0%)	
Serratia	2 (2.3%)	0 (0.0%)	
Pseudomonas	6 (6.9%)	0 (0.0%)	
Klebsiella	9 (10.5%)	6 (9.4%)	
Staph aureous	9 (10.5%)	8 (12.5%)	
Staph. Epidermidis	9 (10.5%)	13 (20.3%)	
Enterobacter	25 (29.1%)	10 (15.6%)	
Negative Blood Culture	25 (29.1%)	23 (35.9%)	
Total positive Blood Culture	61 (70.9%)	41 (64.1%)	

Antibiotics before culture and sensitivity were equally given in both groups, (p = 0.399). CRP titer was also not significantly different in both groups, (p = 0.48), table 3

Table 3. Frequency distribution of antibiotics before blood culture & sensitivity and CRP titer in early and late onset neonatal sepsis

Characteristics	Value	Early onset neonatal sepsis No.==86 (percent)	Late onset neonatal sepsis No.=64 (percent)	P Value
Antibiotics before blood culture and sensitivity	Yes	47 (54.7%)	33 (51.6%)	0.399
	No	39 (45.3%)	31 (48.4%)	
CRP titer	+ve	54 (62.7%)	39 (60.9%)	0.48
	-ve	32 (37.3%)	25 (39.1%)	

There was no significant difference in mean WBC, neutrophil percent, lymphocyte percent, ANC, P value > 0.05, table 4.Mean platelet count was significantly higher in LONS group than EONS group, P =0.01, table 4.

Table 4. Mean ± SD values of age, and hematological parameters in Early onset neonatal sepsis group (n = 86) and Late onset neonatal sepsis group (n = 64).

Characteristics	Early onset neonatal sepsis	Late onset neonatal sepsis	P value
Age (days)	3.29 ± 2.19	17.03 ± 7.64	< 0.01
WBC count ($\times 10^9$)	13.26 ± 9.17	12.14 ± 7.07	> 0.05
Neutrophil percent	53.37 ± 18.319	53 ± 17.06	> 0.05
Lymphocyte percent	39.37 ± 18.22	37.07 ± 15.77	> 0.05
Absolute neutrophil count (×1000)	5.37 ± 3.65	4.56 ± 2.77	> 0.05
Platelet count ($\times 10^9$)	162.51 ± 111.71	228.61 ± 181.82	< 0.05

The number of neonates with I:T neutrophils count of >20% in both groups was 87 of total 150 (58%), and was significantly higher in LONS group than EONS group, 61(70%) and 26 (30%) respectively.

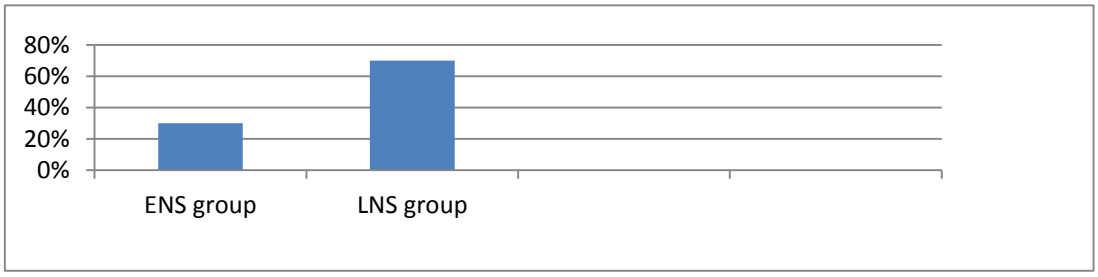

Figure 1. EONS and LONS groups of I:T neutrophils >20%

Discussion

In this study, prematurity as a risk factor was higher in EONS group compared to LONS group (44.2% vs. 29.7%), but in LONS, prematurity as a risk factor was less than full term neonates, (29.7 vs. 70.3), while Stoll BJ Hansen study had prematurity results of (23% vs. 22%) [4] and Jordan HT, etal (birth at<37 weeks gestation was common among case-infants 49%) [5]. In our study and in other studies, this may be due to premature infants may have immune dysfunction. Premature infant often require prolonged intravenous access, endotracheal intubation or other invasion procedure make him more susceptible to infection [4, 6].In our study, the number of neonates who were delivered by vaginal delivery (VD) was equal to the number of those delivered by cesarean section (C/S) in EONS, but there is significant difference in mode of delivery between VD, and C/S in LONS group, (73.4 vs.26.6). This difference in number of neonates with LONS regarding vaginal route may be attributable to a relatively less aseptic environment, frequent vaginal examinations, passing through birth canal which may harbor pathogenic organisms and midwife interference. [7,8].The incidence of neonatal sepsis is probably more when there is midwife interference, but in our study, there was relatively small number of midwife interference. Adefinitive diagnosis of neonatal sepsis is established by a positive blood culture. In this study, 61 neonates (70.9%) of EONS group, and 41 neonates (64.5%) of LONS group have positive blood culture. Garcia-Prats JA et al identify 94% culture positive at 48 hours of incubation in aerobic

cultures obtained from both term and preterm infants using a computer-assisted, automated blood culture system [9]. While Auravg Zeb et al, has reported (55%) culture positive of neonatal sepsis [10]. This variation in blood culture positivity may depend on the criteria of studied groups, volume of sample, the number of cultures obtained, sampling site and antibiotic used prior to the sample [11,12].At 18 hr of membrane rupture, the incidence of early-onset disease with group B streptococcus increases significantly; 18 hr is the appropriate cutoff for increased risk of neonatal infection. [13] In this study, 27 neonates (31.3%) had prolonged rupture of membrane in EONS, and 11neonates (17.2%) in LONS. This result differs from Goldenberg RL. et al study who reported (50.6%) for EONS & (25.85%) for LONS [14]. Brodie SB, et al reported (42.3%) for EONS and (18.8%), for LONS [15]. This difference may be explained by possible higher rate of direct exposure of neonate to vertical transmission of microorganism from genital tract during the delivery process especially for EONS. The attack rate of neonatal infection increase in presence of maternal fever. Intrapartum maternal temperature ≥38ºC (100.4ºF) is a risk factor [16]. In this study, 20 neonates had a history of maternal fever (23.7%) in EONS &16 neonates (25.85%) in LONS. Goldenberg showed (88.2%) association with maternal fever in EONS and (10.4%) in LONS [14]. While Brodie [15] reported that only (44.1%) in EONS and (19.7%) in LONS. This difference in the results of different studies may depends on the type of microorganism, duration of fever, gestational age and duration of membrane rupture.In our study, antenatal care also had an effect on the health of the neonates. There were 36 neonates (41.9%) of mothers with poor antenatal care, and 35 neonates (54.7%) of those with no antenatal care for EONS group, VS. 30 neonates (34.9%) of mothers with poor antenatal care, and 21 neonates (32.8%) of those with no antenatal care for LONS group. Gensen HB, et al reported that there was (40.7%) association with poor antenatal care and (66.4%) of those with no antenatal care [17]. This may depend on the education of the individual and community and regularity of antenatal care and the methods applicable for good antenatal care. The patterns of pathogens associated with neonatal sepsis have changed over time. The incidence of early-onset sepsis mainly caused by Group B streptococcus decreased. In contrast, the incidence of late-onset sepsis, predominantly caused by coagulase-negative staphylococci increased significantly [18]. In this study the organisms isolated from blood cultures of EONS group were: *enterobacter; klebsiella; staphylo-coccus aureus; staphylococcus epidermedis and pseudo-monas aeruginosa*; and these are surprisingly not the common known pathogens causing EONS (*Group B strepto-coccus, Escherichia coli, Listeria monocytogenes*). These yeilds may be explained by acquisition of microorganisms either from the community or from previous admission to the neonatal care units. The organisms isolated from blood cultures of LONS group were:*; staphylococcus epidermedis, staphy-lococcusaureus, klebsiella and Escher-ichia coli* .In this study, the mortality rate reported was 16 neonates (18.6%) in EONS, and 2 (3.1%) for LONS. For EONS group, 13 neonates (81.2%) were died after 3 days of admission (with positive blood culture, and the organisms were: enterobacter; pseudo-monas eaeruginosa and klebsiella). In LONS group the two patients who were died had positive blood culture and the microor-ganismwas pseudo-monas aeruginosa. Adams-Chapman I, et al reported (10%) mortality rate of neonatal sepsis because all bactereamic infections are included in the definition [19].In this study, CRP had no significant value for the diagnosis of neonatal sepsis, positive in

54 neonates (62.7%) of neonates in EONS and 39 neonates (60.9%) in LONS. These results differ from that of Tariq Ghafoor, et al, who observed (35.5%) positive CRP for proven sepsis by blood culture [20]. Shabbir et al found positive CRP in (74%) [21].The discrepancy in the result of CRP may be due to different methods of estimation and/or variation in criteria of positivity of the test. CRP could be a key parameter for individually guiding the duration of antibiotic treatment in a major subgroup of newborns with suspected bacterial infection. This approach would allow considerably shorter courses of antibiotic therapy [22]. Platelet count has a high sensitivity (p=0.01) for those with EONS in comparison to the results of WBC and ANC (p=0.152) and (0.429) respectively. These findings differ from Tariq Ghafoor et al, who reported ANC, platelet and WBC with EONS of (71.4%), (64.3%) and (39.3%) respectively for proven sepsis [20]. This may depend on severity of infection, age of neonate and criteria of studied group. The number of neonates with I:T neutrophils count was 87 (58%), and this was significant number, and was significantly higher in LONS group, 61 neonates (70%) than EONS group 26 neonates (30%) and may be useful in patients with false negative blood culture, and in proceeding toward successful management, and in decreasing the morbidity and mortality of EONS and LONS

Conclusion

Premature neonates were more susceptible for early neonatal sepsis than the full term baby, and prolonged rupture of membranes was more frequently affecting the early neonatal sepsis in comparison to the late neonatal sepsis. Some risk factors has no significant difference between EONS and LONS such as gender, maternal fever, and antenatal care. Incidence of positive blood culture was higher in EONS than LONS, with significant difference in the causative organism between the two groups. Antibiotics used before blood culture highly affect the culture results. CRP, platelet counte specially thrombocytopenia and I:T neutrophils count had high sensitivity to the diagnosis of neonatal sepsis. Death was more frequent in EONS than LONS. This study shows the limitation in getting good blood culture yields in our local laboratories, other studies are needed to evaluate the significance of other investigations like a variety of diagnostic markers of infection. It is unclear which surrogate markers for infection are most helpful.

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