Original Article

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A Study of Hematological Changes in Neonatal Sepsis

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ABSTRACT

Background: Neonatal sepsis is one of the major causes of morbidity and mortality in the newborn. Early diagnosis and treatment are crucial in the management of neonatal sepsis. Blood culture, the gold standard, takes long time and lack sensitivity. Hematological sepsis score is a simple score obtained by combining routinely available tests and is rapid, non-expensive and has shown good correlation with blood culture. With this background the study was carried out to evaluate hematological changes in neonatal sepsis and to analyse the diagnostic utility of hematological scoring system (HSS) and its correlation with C- reactive protein and blood culture in neonatal sepsis.

Methods: The study included 200 neonates admitted to Neonatal Intensive Care Unit with clinical suspicion of neonatal sepsis. CBC, Peripheral blood smear, CRP and Blood culture were obtained from all the neonate. Hematological sepsis score was calculated, and its sensitivity and specificity were obtain using blood culture as gold standard.

Result: We screened 200 neonates with suspected neonatal sepsis. Blood culture was positive in 101 (50.5%) and HSS score >2 was found in 80 (40%) neonates. CRP and HSS had sensitivity of 84.15% and 72.27% and specificity of 68.68% and 92.92% respectively.

Conclusion: Our study shows that HSS had fairly good sensitivity and specificity for neonatal sepsis screening and HHS can be utilized for neonatal sepsis screening.

Keywords: *Neonatal Sepsis, Hematological Scoring System, Blood Culture, C- Reactive Protein.*

Introduction

Neonatal sepsis is defined by systemic infections in first 28 days of life.[1] Preterm birth complications and infections are the largest contributors to the neonatal mortality. The incidence of neonatal sepsis according to the data from National Neonatal Perinatal Database (NNPD, 2002-03) is 30 per 1000 live births. Mortality due to sepsis can be prevented by early diagnosis, rational use of antibiotics and aggressive supportive care. However, early recognition is difficult as the sign and symptoms of early sepsis are non-specific. Blood culture is regarded as the gold standard test, but it takes around 48-72 hours. Further yield of blood culture is 30-70 %, so some neonates may be missed.[2] Furthermore, inability to exclude sepsis early results in the unnecessary exposure to antibiotics to the infants who do not have sepsis. Novel markers, like Interleukin-6, Interlukin-8, plasma elastase, are more sensitive early diagnosis but are not routinely available and are impractical for use in developing country like India. Leukopenia, toxic granules, immature neutrophil to total neutrophil ratio, thrombocytopenia, micro-ESR, C-reactive protein are some of the indirect markers of sepsis. These investigations are collectively referred to as sepsis screen. These investigations are rapid and available routinely. Presence of two or more of these in the background of clinical suspicion is positive sepsis screen. These can be useful for early recognition of sepsis and guide in treatment initiation. These are particularly useful in the resource limited setting where advance markers are not available. The current study is being undertaken therefore to study hematological changes in the neonatal sepsis and to analyse the diagnostic utility of hematological scoring system (HSS) and its correlation with C- reactive protein and blood culture in neonatal sepsis.

Material and Method

The study was conducted in tertiary care teaching hospital in Bhavnagar, Gujarat, India from 1st April 2018 to 1st April 2019. Ethical approval from institutional ethical committee was obtained and written and informed consent was obtained from the parents. A total of 200 neonates admitted with clinically suspected septicemia were included in the study. Neonates who received antibiotics before admission, underwent surgery, had congenital anomalies or inborn errors of metabolism and neonates of mother with pregnancy induced hypertension were excluded. A detailed history of basic demographic data like age, gender, gestational age, mode of delivery, birth weight was recorded. Clinical features at the time of admission to hospital were obtained. These included refusals to feed fever, hypothermia, respiratory distress, abdominal pain, pallor, jaundice, convulsion, lethargy and rash. Blood

counts were done using automated hematological analyser and also manually by examination of stained slides. Automated hematological analyser utilised was Nihon kohden. Slides were prepared by using the field stain. Total WBC count, absolute neutrophil count, immature to mature neutrophil ratio (I:M ratio), degenerative change in PMN, Platelet count, C-reactive protein (CRP) and blood culture were obtained. The cut-off values of above blood tests are mentioned in the table 1 and were taken from Manroe BL et al.[3] Blood culture were incubated for 48 hours before labelling them as negative. CRP estimation was done by latex agglutination method. Hematological scoring of Rodwell (HSS) was obtained as per table 1. HSS was interpreted as sepsis unlikely if <2, sepsis possible if between 3 and 4, and sepsis very likely if >4. Data was tabulated in Microsoft Office Excel worksheet and descriptive statistics were given as means, median and standard deviations at 95% confidence interval. Sensitivity, specificity, positive predictive value and negative predictive value of septic screen was calculated with culture outcome (gold standard). Comparison of neonates with blood culture positive and negative patients was done using Mann-Whitney U Test for continuous parameters (Weight, Age, Total leucocyte cunt, ANC, Platelet count). Comparison of neonates with blood culture positive and negative neonates was done using Fischer's exact test for non-continuous variables (Gender). A p value less than 0.05 was considered as significant at 95% confidence interval.

Results

A total 200 neonates with 112 (56%) male were included in the study. Mean age was 7.37 days and mean birth weight was 1.84 kg with 80% of total neonates having low birth weight. Blood cultures were positive in 101 (50.5%) of neonates. Most common organism were Klebsiella, followed by E. Coli and Acinetobacter. Frequency of identified organism is shown in the figure 1. Table 2 shows comparison of blood culture positive and negative neonates. Blood culture positive neonates were more likely to have prematurity, low birth weight, raised CRP, I:T and I:M ratio, raised total PMN and WBC counts and a higher HSS score.

A total of 80 neonates had Haematological sepsis score of Rodwell of >2. HSS >2 had sensitivity of 72.27% and specificity of 92.92%. Positive and negative predictive value were 91.25% and 76.66%. As compared to individual components combined HSS of >2 had higher sensitivity, except for I:T PMN ratio. Similarly, specificity was also high for HSS except for immature PMN counts and I:M PMN ratio. Table 3 summaries sensitivity, specificity, positive and negative predictive values of HSS and subcomponent of HSS. Taking HSS>3 as positive sepsis screen sensitivity was decreased to 55.44% however, specificity was increased to 98.98%. Similarly, HSS>4 had sensitivity 45.54% and specificity of 100%. Positive and negative predictive values of HSS>4 were 100% and 64.28% respectively. Sensitivity of CRP and HSS >2 was 73.27% and 72.27% respectively considering blood culture as gold standard. Specificity was 80.95% and 92.92% respectively.

Discussion

Neonatal sepsis is a common cause of neonatal mortality. Outcome depends on early diagnosis and initiation of

Table 1: Hematological scoring system of Rodwell.

	Abnormality	Score
I;T ratio	Increased (normal is 0.16 in first 24 hours of life and 0.12 after 24 hours of life)	1
Total PMN count	Increased or decreased (normal range: first 60 hours 7800-14500, 60-120 hours 1800-7000, 120 hours onward 1800-5400) No mature PMN seen	1 2
I:M ratio	≥ 0.3	1
Immature PMN count	Increased (Normal is <1400/mm^3 in first 24 hours, <1000/mm^3 in 24-48 hours and <600/mm^3 in 48-60, <500/mm^3 after 60 hours)	1
Total WBC count	≤5000/mm^3 or ≥25000/mm^3 30,000, and 21,000/mm^3 at birth, 12-24 h, and day 2 onward, respectively)	1
Degenerative change in PMN	≥3+ for vacuolization, toxic granulation, or Dohle bodies	1
Platelet count	≤ 150000/mm^3	1

I:T immature to total neutrophil count, I:M immature to mature neutrophil count, PMN- polymorphonuclear cell. Normal value taken from Manroe BL et al. [11]

Table 2: Comparison of blood culture positive and negative neonates.

Characteristics	Blood culture positive	Blood culture negative	P value
Age (mean ± SD)	6.83 ± 4.00	7.29 ± 4.30	0.073
Gender (male)	44	54	0.157
Birth weight (mean ± SD) Kg	1.55 ± 0.44	2.16 ± 0.59	0.00001
Total Leucocyte count cells/mm^3(mean ± SD)	12077 ± 7740	6294 ± 1896	P<0.00001
ANC (mean ± SD) cells/mm^3	7409.5 ± 5163	3168 ± 1225	p<0.00001
Degenerative changes in neutrophils (mean ± SD)	4.32 ±3.02	1.24 ±0.55	p<0.00001
I:T PMN ratio (mean ± SD)	0.18 ±0.09	0.08 ± 0.07	p<0.00001
I:M PMN ratio (mean ± SD)	0.25 ± 0.20	0.10 ± 0.17	p<0.00001
Platelet count (mean ± SD)	1.59 ± 1.01	2.38 ± 0.65	p<0.00001
HSS (mean ± SD)	3.90 ± 1.80	0.82 ± 0.95	P<0.00001

Table 3: Sensitivity, specificity, positive and negative predictive value of HSS and subcomponent of HSS.

Parameter	Sensitivity	Specificity	PPV	NPV
I:T PMN ratio	85.14%	82.82%	83.49%	84.53%
Total PMN	64.35%	74.74%	72.22%	67.27%
I:M PMN ratio	25.74%	97.97%	92.85%	56.39%
Immature PMN	55.44%	97.97%	96.55%	68.30%
Total WBC	48.51%	71.71%	63.63%	57.72%
HSS >2	72.27%	92.92%	91.25%	76.66%

Table 4: Diagnostic accuracy of HSS.

S. No	Study	Sensitivity (%)	Specificity (%)
1	Mondal SK et al [3]	84	84
2.	Sriram et al. [12]	55.30	91.7
3	Misra RN et al [13]	100	37.5
4	Bhale CP et al [8]	93.4	77.0
5	Vinay BS et al. [7]	77	41
6	Present study	72.27	92.92

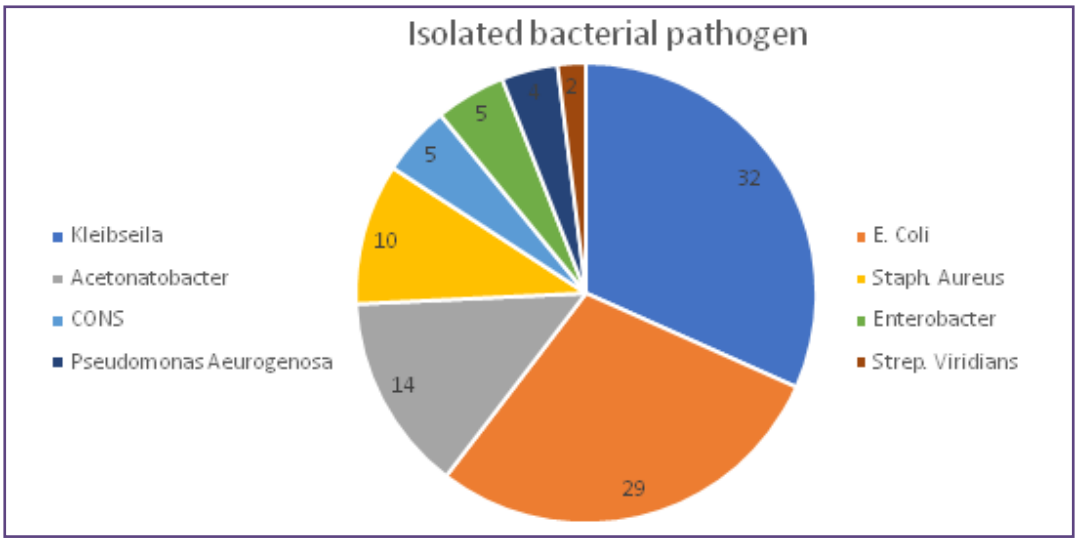

Fig. 1: Frequency of isolates in blood culture.

appropriate antibiotics. Traditional method for diagnosis of neonatal sepsis is blood culture. However, blood culture has sensitivity of 50-70% and results take at least 48 hours. Alternative methods for diagnosis include inflammatory markers, cytokines and molecular markers. Despite being sensitive and showing changes early in the course of neonatal sepsis these markers are not utilised routinely in view of cost and limited availability. Hematological changes in neonatal sepsis includes leucocytosis or leucopoenia, increase or decrease in ANC, increase in immature neutrophils and change in immature to mature PMN ratio. However, individual hematological change lack enough sensitivity or specificity. Haematological sepsis score of Rodwell combines various hematological changes and increases sensitivity and specificity. Our study included 200 neonates with suspected neonatal sepsis. Blood culture was positive in 101 (50.5%) of patients. Considering blood culture as gold standard HSS>2 had sensitivity of 72.27% and specificity of 92.92%.

Diagnostic Accuracy of CRP

CRP had sensitivity 84.15% and specificity was 68.68%. Positive and negative predictive value was 73.27% and 80.95% respectively. Other studies also have shown similar sensitivity and -specificity ranging from 81-90% and 50-84% respectively. [3][4][5][6][7][8]

Diagnostic Accuracy of Total WBC Counts

Total WBC counts had sensitivity of 64.35% and specificity of 74.74%. Previous studies also have shown variable results with sensitivity ranging from 43-75% and specificity ranging from 74-94%. Studies have shown wide variability in the sensitivity ranging from 43-75% and specificity ranging from 74-94%.[4][6][8][9][10]

Diagnostic accuracy of total ANC

In present study, ANC had sensitivity of 64.35% and specificity of 74.74%. Other studies have shown sensitivity ranging from 42-80 % and specificity ranging from 79-99%.[6][8][9][10]

Diagnostic accuracy of I:M PMN ratio

In present study I:M PMN ratio had lower sensitivity 25.74% but higher specificity of 97.97%. This contrasts with other studies wherein sensitivity was high.[6][10] However, Makkar M. et al reported low sensitivity and high specificity.[9]

Diagnostic accuracy of I:T PMN ratio

In present study I:T PMN ratio had sensitivity of 85.14% and specificity of 82.82%. Other studies have shown variable results. Makkar M et al had high sensitivity and specificity whereas Vandana G et al had low sensitivity and specificity.[5][9]

Diagnostic accuracy of HSS of Rodwell

In present study HHS>2 had sensitivity of 72.27% and specificity of 92.92%. HSS>2 has sensitivity ranging from 55-100% while specificity was 37-91%. Misra RN et al, reported very high sensitivity but low specificity, whereas Mondal SK et al, had similar sensitivity and specificity as our study. Table 4 summarise diagnostic performance of HSS in various studies.

Conclusion

Neonatal sepsis is common cause of neonatal mortality. Early diagnosis and timely initiation of antibiotics is vital for effective management. HSS is an easy, cost-effective and can be obtained by test done routinely. Our study showed HSS has a sensitivity of 72.27% and specificity of 92.92% as compared to blood culture. So HSS can be used for the screening of neonatal sepsis.

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Competing Interests

There are no conflict of interest.

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