

## Diagnosis of Neonatal Sepsis by Blood Culture and Thrombocytopenia



### Microbiology

**KEYWORDS :** Neonatal Sepsis, Thrombocytopenia, Blood Culture

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### ABSTRACT

*Introduction: The early diagnosis for neonatal sepsis is important because the infection may present a very serious threat to the baby. In situations in which the clinical picture leaves the physician in doubt about the infection status that a diagnostic test is likely to be most useful. The presence of thrombocytopenia is seen frequently in early sepsis with or without evidence of overt DIC. Aims & objective: To compare the results of blood culture and platelet count for diagnosis of neonatal sepsis. Material & methods: This study was conducted on 170 neonates admitted in NICU with clinical diagnosis of septicemia. We have taken 100 age and weight matched neonates as control group. Blood from the patient was collected for culture and complete blood count. Blood culture and platelet count of the patients were compared. Results: Thrombocytopenia was seen in 85.88% cases where as bacterial culture was positive in only 45.29% cases. Further it was noted that, in Gram negative septicemia, thrombocytopenia was more severe as compared to gram positive septicemia. Conclusion: It is concluded that thrombocytopenia is early predictor of septicemia but other causes of neonatal thrombocytopenia should be ruled out.*

### INTRODUCTION

The early diagnosis for neonatal sepsis is important because the infection may present a very serious threat to the baby. In situations in which the clinical picture leaves the physician in doubt about the infection status that a diagnostic test is likely to be most useful. Most of ill and premature infants have low platelet count. [1]

Apart from infection due to any of the above agents there are many other causes of thrombocytopenia such as immune mediated, chromosomal anomalies and genetic disorders. [2]

The presence of thrombocytopenia is seen frequently in early sepsis with or without laboratory evidence of overt DIC [3, 4]

Various studies are carried out in developed countries and in India show vast difference in growth of isolates. It is noted that in western worlds, Gram positive bacteria such as Group B Streptococci and coagulase negative Staphylococci (CONS) are usual isolates whereas in India Gram negative bacteria form majority of isolates. Klebsiella species is noted to be the commonest organism along with E. coli, Staph. aureus, Staph. epidermidis, Group B Streptococci, Enterococcus faecalis, Enterobacter sp., Acinetobacter sp., Pseudomonas sp., Proteus sp. seen in developing countries [2, 5-8] Culture is not free from error because it can be falsely sterile, or because of the low yield caused by insufficient sample volumes, intermittent or low-density bacteremia, or suppression of bacterial growth by earlier (i.e., intrapartum) antibiotic administration. [9] Thrombocytopenia is usually seen with Gram positive septicemia as compared to Gram negative septicemia [10] and low platelet is usually seen even before the pathogens are cultured from the blood. Therefore, thrombocytopenia may be considered as an important and early tool in diagnosis of septicemia in neonates [11].

### Mechanism of thrombocytopenia in septicemia

The mechanism of thrombocytopenia in patients with septicemia, with no evidence of disseminated intravascular coagulation (DIC) is uncertain. On the other hand thrombocytopenia in DIC is considered mainly due to consumption of certain coagulation factors and circulating platelet. There is evidence of

an activated fibrinolytic mechanism by the presence of FDP in serum and low levels of factor II, V, VIII and fibrinogen. All these observations indicate consumption of circulating platelet as a usual cause of thrombocytopenia [3, 12]

Bacteria or bacterial products may cause endothelial damage leading to platelet adhesion and aggregation or may bind directly to platelets leading to accelerated clearance from circulation [13, 14]. There may be a possibility of immune mechanism for development of thrombocytopenia in septicemia as there is presence of circulating immune complex in septicemic patients and reduced number of complement complex in patients with septicemic shock [15, 16]

### MATERIAL AND METHODS

This study was done from January 2013 to June 2013 in the Department of Microbiology, N.H.L. Medical College, Paldi, Ahmedabad.

**Inclusion criteria:** A total 270 full term neonates of >2.5 kg weight were included in the study, Among them 170 were clinically suspected cases of septicemia and admitted to NICU, the other 100 neonates were considered as control.

**Exclusion criteria:** Full term neonates <2.5 kg or preterm neonates were not included in the study. Full term neonates without clinical diagnosis of septicemia were excluded.

Blood was collected with all aseptic precaution in EDTA tube. Platelet count was performed by automatic hematological cell counter as a part of complete blood count.

Blood was collected with all aseptic precaution in blood culture bottle containing glucose phosphate broth, it was incubated at 37°C for 5 days. If turbidity or hemolysis was seen, subculture was done on that day. Subculture was done on Nutrient agar, blood agar, Sabourad's dextrose agar and MacConkey's agar media after 24 hour and 5 days of incubation. Blood culture was reported sterile if no growth was seen on subculture after 5 days of incubation.

## IDENTIFICATION OF THE ISOLATION

Cases							Control			
	Blood culture positive	%	Blood culture negative	%	Total	%		Blood culture positive	Blood culture negative	Total
Thrombocytopenia	58	39.73	88	60.27	146	85.88	Thrombocytopenia	Nil	Nil	Nil
Normal Platelet count	19	79.17	5	20.83	24	14.12	Normal Platelet count	Nil	100	100
Total	77	45.29	93	54.71	170		Total	Nil	100	100

After over night incubation, isolated organisms were identified with conventional methods. Colony characteristics on Nutrient agar, blood agar, Sabourad's dextrose agar and MacConkey's agar were studied and organism was identified by examination of Gram stain smear, motility and using the various biochemical tests.

Bacteriological profile of septicemia with and without thrombocytopenia					
Causative organism		Total	Thrombocytopenia	Normal Platelet count	Platelet count range in cases of thrombocytopenia ( $\mu$ l)
Gram negative bacteria	Klebsiella pneumoniae	28	28	-	$33 \times 10^3 - 94 \times 10^3$
	E. coli	11	11	-	$35 \times 10^3 - 99 \times 10^3$
	Acinetobacter	7	7	-	$49 \times 10^3 - 58 \times 10^3$
	Pseudomonas	1	1	-	$35 \times 10^3$
Total		47	47	-	
Gram positive bacteria	Enterococcus faecalis	15	6	9	$95 \times 10^3 - 110 \times 10^3$
	Coagulase negative Staphylococcus	10	1	9	$95 \times 10^3$
	Staphylococcus aureus	5	4	1	$110 \times 10^3 - 115 \times 10^3$
Total		30	11		

## OBSERVATIONS

This study was done on 270 neonates. 170 had clinical diagnosis of septicemia and 100 were controls who were full term neonates. Out of 100 neonates 60 were male and 40 were female. Average platelet count  $185 \times 10^3 \mu$ l-1.

Table 1 shows that thrombocytopenia was recorded in 85.88% (146 cases), among them blood culture was positive in 39.73% (58 cases) and in 60.27% (88 cases) blood culture were negative. 24 cases had normal platelet count, out of which 79.17% (19 cases) had positive blood culture and 20.83% (5 cases) had negative blood culture. Controls had normal platelet count and negative blood culture.

In similar study by S. H. Arif showed that 71 (83.5%) neonates had low platelet count ( $<150 \times 10^3 \mu$ l), indicating that thrombocytopenia is an important finding in sick neonates admitted in NICU. Out of these 71 cases only 24 (33.8%) cases showed positive blood culture.

In present study P value was  $0.00073 < 0.05\%$  and significant at 5% level of significance. The study showed that there is significant relation in platelet count and septicemia, indicating that clinically suspected cases of septicemia platelet count was low in 85.88% (146 cases), out of which 39.73% (58) had positive blood culture.

Neonatal septicemia was commonly due to Gram negative bacteria (n=47, 61.03%), mainly because of Klebsiella pneumoniae (n=28) and Escherichia coli (n=11) and only 38.96% were due to Gram positive bacteria. Enterococcus faecalis was most common cause of Gram positive septicemia. Severe Thrombocytopenia was observed in Gram negative sepsis. In study by S. H. Arif It was observed from the study that Gm +ve bacteria is commonly isolated as compared to Gm -ve in neonates with septicemia.

## DISCUSSION

Neonatal sepsis or sepsis neonatorum refers to systemic infection of the newborn. It is characterized by a constellation of a nonspecific symptomatology in association with bacteremia. Prompt recognition, appropriate antimicrobial therapy and judicious supportive care are the key determinants of positive outcome in this serious pediatric emergency. In developing countries, sepsis including meningitis, respiratory infections, diarrhea, and neonatal tetanus is the commonest cause of mortality responsible for 30-50 per cent of 5 million total neonatal

deaths each year. It is estimated that almost 20 per cent of all neonates develop infection and approximately 1 per cent die of the serious systemic infections. Not surprisingly, sepsis is the commonest admitting diagnosis among neonates at referral facilities. Several risk factors are identified in the mothers and in the neonates who make them susceptible to infections. Neonatal sepsis requires rapid diagnosis. Thrombocytopenia occurs early in the course of septicemia, therefore platelet count can be considered as early predictor for diagnosis of septicemia. Among neonates with thrombocytopenia 39.73% had positive blood culture suggesting that low platelet count is important finding in sepsis. Many cases with low platelet count had negative blood culture. Thus platelet count is important but nonspecific indicator of neonatal sepsis.

This study indicate that Gram negative bacteria is common than Gram positive bacteria as a cause of septicemia in neonate. Study by K. Chugh on "Bacteriological profile of neonatal septicemia" showed similar finding. [17] While other study by G Kartikeyan on "Neonatal Sepsis : Staphylococcus aureus as the predominant pathogen" showed isolation of Gram positive bacteria. [18] Among Gram negative sepsis Klebsiella was the commonest organism. Thrombocytopenia was more severe in Gram negative sepsis.

Klebsiella has ability to cause outbreak of serious infection in NICU because of its ability to colonise neonates, to survive in inanimate environment and ability to acquire antibiotic resistance. [19]

## CONCLUSION

It is concluded from the study that septic neonates had common finding of thrombocytopenia. Klebsiella pneumoniae is the most common isolate and it was accompanied by thrombocytopenia. Many cases with thrombocytopenia had no growth in blood culture. This indicates that thrombocytopenia may be considered as early but nonspecific indicator of neonatal sepsis and other causes should be ruled out.

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