



## Significance of Nucleated RBC Count in Term Neonates with Asphyxia Neonatorum

Authors

\***S.Sonia Rosalind Martina**<sup>1</sup>, **S.Saravanan**<sup>2</sup>, **M.Venkatadesikalu**<sup>3</sup>

<sup>1</sup>Post Graduate, Department of Paediatrics, RMMCH, Chidambaram-608002, Tamilnadu, India

<sup>2</sup>Associate Professor, Department of Paediatrics, RMMCH, Chidambaram-608002, Tamilnadu, India

<sup>3</sup>Professor, Department of Paediatrics, RMMCH, Chidambaram-608002, Tamilnadu, India

### ABSTRACT

**Objective:** To study the correlation of nucleated red blood cell (NRBC)/100 white blood cell (WBC) count with perinatal asphyxia in terms of severity and short term outcome.

**Material and Methods:** A prospective, comparative case-control study was undertaken at RMMCH Chidambaram from January 2015 to July 2016, which included a total of 100 neonates with 50 asphyxiated babies (case group) and 50 normal babies (control group). The venous blood was collected immediately after delivery for measurement of NRBC/100 WBC. Early neonatal outcome of both groups was also evaluated in relation to the NRBC/100 WBC count. Statistical analysis was performed.

**Results:** The mean NRBC/100 WBC count was  $16.76 \pm 11.46$  in case group and in control group  $3.78 \pm 2.79$ . Also a statistically significant correlation existed between severity of asphyxia (stage of hypoxic-ischemic encephalopathy [HIE]), poor outcome and higher number of NRBC/100 WBC count.

**Conclusion:** It is an inexpensive and easily available procedure to evaluate perinatal asphyxia, especially in a resource poor country, where blood gas analysis facilities are not available in majority of places. Also, it is a good predictor of short-term outcome of asphyxiated babies.

**Keywords:** Cord blood, nucleated red blood cells, perinatal asphyxia, hypoxic-ischemic encephalopathy.

### Introduction

Asphyxia neonatorum, also called birth or newborn asphyxia, is defined as failure to start regular respiration within a minute of birth. Asphyxia neonatorum is a neonatal emergency as it may lead to hypoxia (lowering of oxygen supply to the brain and tissues) and possible brain damage or death if not correctly managed. Newborn infants normally start to breathe without assistance and usually cry after delivery. By one minute after birth most infants breathe well. If an infant fails to establish sustained respiration after birth, the infant is diagnosed with asphyxia neonatorum.

Perinatal asphyxia is a major cause of acute mortality and chronic neurologic disability amongst survivors, and is a complication that occurs between 2-10% of deliveries.<sup>1</sup> No single parameter can define perinatal asphyxia, rather a combination of parameters like fetal distress, meconium-stained liquor, low Apgar score, umbilical cord blood pH and clinical features of hypoxic ischemic encephalopathy (HIE) can predict it. The 1996 guideline from the American Academy of Pediatrics (AAP) and American College of Obstetricians and Gynecologists (ACOG)<sup>2</sup> for HIE indicate that all of the following

must be present for designation of perinatal asphyxia severe enough to result in acute neurological injury:

- ❖ Profound metabolic or mixed acidemia (pH <7) in an umbilical blood sample and persistence of an Apgar score of 0-3 for longer than 5 minutes.
- ❖ Neonatal neurologic sequelae (e.g-seizure, coma, hypotonia)
- ❖ Multiple organ involvement (e.g- kidney, lungs, liver, heart and intestines).

But, as per Cloherty et al<sup>3</sup> perinatal asphyxia is defined as a condition of fetal hypoxia and hypercarbia, identified by fetal acidosis as umbilical artery pH <7. The condition can be diagnosed by fetal distress, meconium aspiration, low Apgar score <6 by 5 minutes, umbilical cord blood pH <7. Recent studies on hematological variations in asphyxiated neonates as a predictor of neonatal asphyxia have suggested that number of nucleated red blood cells (NRBCs) in cord blood of asphyxiated neonates help in identifying birth asphyxia. The number of NRBC/100 white blood cells (WBC) is quite variable but is rarely >10.<sup>4,7</sup> The instances, where number of NRBCs exceed 10/100 WBC are asphyxia, prematurity, Rh-sensitization, maternal diabetes mellitus and intrauterine growth retardation.

Considering the hematopoietic response to hypoxia in utero the elevated NRBC/100 WBC count is being hailed as the marker for not only perinatal asphyxia but also the chances of the neonates to develop neurological sequelae.<sup>4,7-10</sup>

**Materials and Methods**

A prospective case control study was conducted in NICU, RMMCH. A total of 100 newborns were enrolled after consent from parents, 50 were cases with birth asphyxia who met the inclusion criteria of Apgar score <3/10 at 1 minute and controls were with Apgar score >7/10 at 1 minute. Preterm babies, babies with downs syndrome, TORCH, sepsis & Rh incompatibility with hemolysis were excluded. 2ml of venous blood collected within 6 hours of birth and used for making peripheral

smears with Leishman stain and complete blood counts. Number of NRBC were counted per 100 WBC in peripheral smear and were reported.

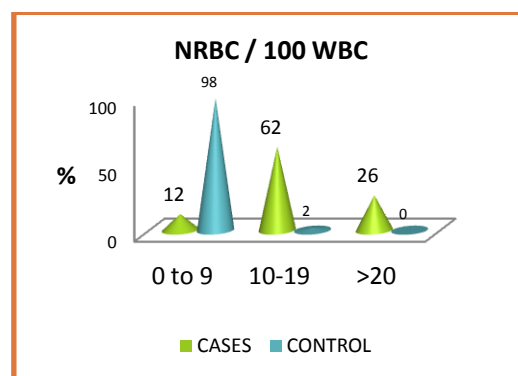
**Results**

There is no statistical significant difference in the age, parity status, gestational age of mother, birth weight and mode of delivery between cases and control newborns. The Apgar score at different intervals are comparatively less in cases than in controls. The monocytes and NRBC count are significantly higher in cases than in controls. The majority of cases have NRBC count between 10-19 per 100 WBC whereas 98% of controls have NRBC count of 0-9 per 100 WBC. There is negative correlation between Apgar score and NRBC values. There is statistically significant positive correlation is observed between HIE stage and NRBC values.

That is, when HIE stage is higher, NRBC values is also higher and vice versa. There is statistically positive con-elation between NRBC with time of onset of direct breastfeeds, NICU stay, reflex recovery, cry/sucking/activity and hospital stay.

**NRBC in Cases and Controls**

NRBC Count (Per 100 WBC)	Cases		Control		'P' value
	N	%	N	%	
0-9	6	12	49	98	0.001
10-19	31	62	1	2	
≥ 20	13	26	-	-	
Total	50	100	50	100	



Relation between NRBC count and Apgar Score

APGAR Score	APGAR 1 Minute				APGAR 5 Minute			
	Cases		Control		Cases		Control	
	N	NRBC Count	N	NRBC Count	N	NRBC Count	N	NRBC Count
0-3	44	17.05 ± 12.08	-	-	6	23 ± 9.84	-	-
4-6	6	14.67 ± 5.01	19	4.26 ± 3.97	38	16.03 ± 12.16	-	-
7-10	-	-	31	3.48 ± 1.75	6	15.17 ± 6.40	50	3.78 ± 2.79

There is negative correlation between Apgar score and NRBC values. That is when Apgar score is more, the NRBC value is less and vice-versa. But the 'p' value is insignificant ( $P > 0.05$ ). Pearson's correlation coefficient is used to study the relationship of two variables.

Difference in NRBC Count with Reference to Stages of HIE

HIE Stage	N	NRBC Count
		Mean ± S.D.
HIE Stage 1	19	10.00 ± 3.40
HIE Stage 2	24	20.83 ± 14.01
HIE Stage 3	7	21.14 ± 7.20
	50	'F' value 'P' value 6.55 0.003

The mean count of NRBC is highest ( $M = 21.14 \pm 7.20$ ) for HIE stage 3 when compared to HIE stage 2 ( $M = 20.83 \pm 14.01$ ) and HIE stage 1 ( $10.00 \pm 3.40$ ).

NRBC count and various immediate outcomes in cases

NRBC	Time taken for DBF in days	NICU Stay in days	Neonatal Reflex recovery in days	C/S/A recovery in days	Hospital Stay (days)
0-9	3.00 ± 0.63	3.17 ± 0.98	2.17 ± 0.41	2.00 ± 0.00	4.67 ± 0.82
10-19	4.74 ± 2.49	5.45 ± 2.84	4.10 ± 2.88	3.87 ± 2.90	7.84 ± 3.30
≥ 20	4.46 ± 3.57	6.23 ± 4.95	4.38 ± 3.55	3.85 ± 3.05	9.46 ± 4.59

All the parameters are delayed if NRBC count is more as shown in the above Table. The delay is maximum for cases with  $\geq 20$  NRBC count.

Discussion

In the present study, attempt has been made to evaluate the relation of venous blood NRBC/100WBC in predicting perinatal asphyxia and its immediate outcome. The NRBC/100WBC count in control and case group was  $3.78 \pm 2.79$  and  $16.76 \pm 11.46$  respectively this correlates with the study done by Gupta et al<sup>11</sup> in cord blood. They found NRBC of  $10.34 \pm 3.87$  in case group &  $5.7 \pm 2.33$  in control group many different studies all over the world have revealed increased

NRBC/100WBC in umbilical cord blood following perinatal asphyxia.

Also, in the present study, we found higher NRBC/100WBC with higher degree of HIE,  $10 \pm 3.40$  in HIE stage 1,  $20.83 \pm 14.01$  in stage 2, and  $21.14 \pm 7.20$  in stage 3 with p value of 0.003 which is significant. This correlates with the study done by Hermansen,<sup>9</sup> Sikarwar and Gupta,<sup>11</sup> Phelan et al;<sup>5</sup> Philip and Tito,<sup>6</sup> who found that NRBC increases with increase in stages of HIE. Our study also revealed increased NRBC/100WBC count with low Apgar scores many authors like Hanlon-Lundberg and Kirby,<sup>12</sup> Boskabadi et al,<sup>13</sup> Saracoglu et al,<sup>10</sup> had reported similar findings in their studies.

**Conclusion**

Thus in this study venous blood NRBC count has shown to be a good predictor of perinatal asphyxia. It is cost-effective and does not require any special expertise or any high-tech facilities, it may be a useful, reliable, inexpensive and easily available marker to evaluate perinatal asphyxia, specially in a resource poor country, where blood analysis facilities are not available in majority of place.

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