Original Article

OPEN ACCESS

Relationship of Nucleated Red Blood Cells with Severity of Perinatal Asphyxia and its Short-Term Outcome

Mehwish Iqbal[1], Athar Razzaq[2], Fiaz Ahmad Malik[3], Muhammad Ali[4], Muhammad Imran[5], Maham Shahzadi[6]

[1,6]Resident, Pediatrics department, [2]Consultant neonatology ,[3]Consultant pediatrician
[4]Consultant pediatrician/ fellow neonatology,[5]Consultant ophthalmologist
(Recep Tayyip Erdogan Hospital Muzaffargarh)

Author`s Contribution

[1,6]Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work, [2]Active participation in active methodology, [3,4]Drafting the work or revising it critically for important intellectual content, Final approval of the version to be published, [5]statistical analysis

Funding Source: None
Conflict of Interest: None

Received: Mar 06, 2022
Accepted: Aug 15, 2022

Address of Correspondent
Dr. Mehwish Iqbal
Resident, Pediatrics department,
Recep Tayyip Erdogan Hospital Muzaffargarh
Iqbal.mehwish@yahoo.com

ABSTRACT

Objective: To compare the nucleated red blood cell (NRBC) counts of healthy and asphyxiated newborns as a key clinical indicator of the severity of birth asphyxia and short-term outcome.

Methodology: This case control study was conducted at the department of neonatology of Recep Tayyip Erdogan Hospital Muzaffargarh from June 2020 to November 2020. Study A total of 112 term newborns, including 56 cases (with birth asphyxia) and 56 controls (normal babies) aged <24 hours, were included. After obtaining informed consent, blood samples were taken immediately after birth for the measurement of the NRBC count. All the enrolled cases were admitted to the NICU as per indication and observed for the severity of birth asphyxia. The relationship between the cord blood NRBC count and the severity of birth asphyxia was assessed. All the information was recorded using a self-structured study proforma, and SPSS version 26 was used for the purpose of data analysis.

Results: The overall average age of the neonates was 6.18±2.66 hours. Mean age, birth weight, and gestational age were statistically insignificant in accordance to cases and controls. Males were in the majority (75.0%), and females were 25.0% of the case group. The mean NRBCs in the cases were 16.62 ± 17.10, which was significantly higher than the controls at 1.69 ± 2.42 (p-0.0001). The neonatal NRBs average was statistically insignificant according to gender in both cases and controls, while it was significantly associated with the severity of perinatal hypoxia (p 0.0001). The average NRBC count was also significantly higher in cases of cardiomegaly, those having ventilation needs and respiratory distress.

Conclusion: The mean NRBC was observed to be significantly higher in cases compared to controls. The NRBC was considered to be a non-invasive, simple prognostic marker for neonatal asphyxia, may indicate substantial severity of perinatal hypoxia and its related complications.

Keywords: Birth asphyxia, NRBCs, correlation, mortality.

Cite this article as: Iqbal M, Razzaq A, Malik FA, Ali M, Imran M, Shahzadi M. Relationship of Nucleated Red Blood Cells with Severity of Perinatal Asphyxia and its Short-Term Outcome. Ann Pak Inst Med Sci. 2022; 18(3):246-251. doi. 10.48036/apims.v18i3.674

Introduction

Perinatal asphyxia happens when newborns can't start and keep breathing right after they are born. This can cause severe illness and death in the first week after birth. [1] It is a disorder that develops when there is an impairment of blood gaseous exchange, which leads to hypoxemia and hypercapnia.[1] The maternal hemodynamic impairment (amniotic fluid embolus), rupture of the uterus, abruption of placenta, compression or knot of the umbilical cord, or infection could all causes of the perinatal asphyxia.[2] Asphyxia might happen before the birth, or it can happen right after the birth in a patient who is already impaired and needs resuscitation.[2,3] Perinatal asphyxia can result in hypercarbia, severe metabolic acidosis, increasing hypoxemia, newborn encephalopathy, and failure of several organ systems, and it can even cause the perinatal

mortality.[4-6] Each year, out of the world's total population of 140 million newborns, 10 to 15 million may not cry or breathe after birth.[7] It is also a major cause of neurological morbidity and mortality in neonates, especially in developing countries like Pakistan.[8]

Pakistan is one of the countries with the highest neonatal mortality rate (NMR) in the world. According to the Pakistan demographic and health survey (PDHS) 2017–2018, the NMR of Pakistan is 42 per 1000 live births. Among the causes of neonatal deaths in Pakistan, asphyxia accounts for 25% of total neonatal mortality. In the developed nations, the rate of perinatal asphyxia is approximately 2 per 1000 live neonatal births; however, the proportion is 10 times higher compared to the developing nations, where no adequate access to newborn and maternal care.[9] Although the World Health Organization identifies perinatal asphyxia as being one of the 3 most prevalent causes of 11% of deaths in children under the age of five, followed by 15% pneumonia and 17% preterm delivery.[9] The diagnosis of asphyxia is carried out using the numerous approaches nowadays.[10]

Although, still needed, an accurate marker in order to estimate the length of hospital stay and the overall prognosis of newborns with asphyxia.[10] It has been proposed that the number of nucleated red blood cells (NRBC) found in the umbilical cord of neonates is an indicator of the birth asphyxia.[10] NRBCs, also known as erythroblasts, seem to be the premature erythrocytes that are typically discovered in the blood of newborns, and the presence of these cells in peripheral circulation is linked to the nature of hypoxia of fetal growth.[11] However, until now in Pakistan, no study has been done on nucleated red blood cells (NRBs) and its relationship to the severity of perinatal asphyxia and its short-term outcome. As there are no laboratory tests available in Pakistan for the direct evidence of asphyxia, it's a useful, cost-effective tool that provides indirect evidence of perinatal asphyxia. This study will help us determine the magnitude of the problem and will help in diagnosis, treatment, and management protocols.

Methodology

This case-control study was conducted at the neonatology department of Recep Tayyip Erdogan Hospital Muzaffargarh during 6 months from June 2020 to November 2020 after receiving ethical approval from the hospital ethical review committee. Non probability consecutive sampling technique was used. All the neonates with birth asphyxia admitted within 24 hours of life, gestation age 34 to 42 weeks, and neonates without birth asphyxia (controls) were included in the study. The sample size was calculated using a 95% confidence interval, an 80% power, and a sample size ratio of 1.

Open epi sample size calculator of mean difference. For a comparison of NRBs between moderate and severe asphyxia, I will take 56 cases, which include 28 patients with grade 2 asphyxia and 28 patients with grade 3 asphyxia. And for my third objective, I will take an equal number of non-asphyxiated otherwise normal, 56 neonates. All the babies admitted after 24 hours of delivery, those having any other risk factor (Cyanotic heart disease, intrauterine growth restriction, Hemolytic disease of new born, Post term babies) or patients having any dysmorphism assessed on history and medical records were excluded. The severity of birth asphyxia was categorized as mild (1–10), moderate (11–14), and severe (15) according to the Thompson scoring of hypoxic ischemic encephalopathy.[12] A detailed history was taken, and important aspects were noted, like the mode of delivery, place of delivery, history of crying at the time of delivery, history of prolong or obstructed labor, history of antenatal steroids, gestational age, history of leaking, vaginal discharge, and fever in the perinatal period. Then a general physical and systemic examination of the neonate was carried out for activity, breathing, grimace, skin color, pulse, and respiration.

Blood samples were taken from all patients with perinatal asphyxia and the control group. Blood sample will be sent for NRBCs, renal function test, liver function test, coagulation profile, on daily basis and observe the number of nucleated red blood cells and other parameters for multi organ involvement in case and control group. Investigations were carried out in the Hospital laboratory of Recep Tiyyap Erdogan Hospital Muzaffargarh. All the information was recorded by self-structured study proforma and Statistical package for social science (SPSS) version 26 was used for the purpose of data analysis.

Results

In this study, a total of 112 newborns were studied, particularly as 56 cases (perinatal hypoxia) and 56 controls (normal), to assess the association of NRBC with perinatal hypoxia. The mean age of the cases was 6.60$\pm$1.10 hours, and the mean age of control newborns was 4.0$\pm$2.0 hours. Mean birth of the cases was

2.48+0.45 kg and birth weight of controls was 2.90±0.37 kg. The average length of the cases' newborns was 46.35±4.22 cm, while the average length of the controls was 47.78±3.97 cm. mean gestation age of the of the cases group was 36.33±1.23 weeks and mean gestational age of control group was 37.54±2.98 weeks. Although mean age and birth weight were statistically significant (p= <0.05), while mean length and mean gestational age were statistically insignificant according to both groups (p=>0.05) as shown in table I.

In this study, males were in the majority in the case group (75.0%) and females were 25.0%, while in the control group both male and female newborns were equal. Findings were statistically significant according to both groups (p= 0.006). Rate of c-section was significantly higher 80.4% in control group, while 64.3% c-section rate was in cases group out of 56 cases in each group (p= 0.057). Table II

In this study average of NRBCs was seen significantly high in cases 16.62±17.10 as compared to control group as 1.69±2.42 (p=0.0001). Table III

As shown in table III, there was a mean creatinine level, AST and ALT were significantly higher, and platelets averaged significantly lower in the case group compared to the control group (p = 0.05), whereas Hb, leukocytes, and CRP were statistically insignificant in both study groups (p = 0.05).

Table I: Descriptive statistics of neonatal age, weight, length and gestational age (n=112)

Variables	Study groups	N	Mean±SD	p-value
Age (hours)	Control	56	4.00±2.00	
	Cases	56	6.60±1.10	0.0001
Birth weight (kg)	Control	56	2.90±0.37	
	Cases	56	2.48±0.45	0.0001
Length (cm)	Control	56	47.78±3.97	
	Cases	56	46.35±4.22	0.068
Gestational (weeks)	Control	56	37.54±2.98	
	Cases	56	36.33±1.23	0.320

Cases = Newborns with perinatal hypoxia, **Control**= Newborns without perinatal hypoxia, **Std. D** = Standard deviation

Table II: Cases distribution according to neonatal gender and MOD (n=112)

Variables			Perinatal hypoxia			p-value
			No	Yes	Total	
Gender	**Females**	N. of cases	28	14	42	
		%	50.0%	25.0%	37.5%	0.006
	Males	N. of cases	28	42	70	
		%	50.0%	75.0%	62.5%	
Mode of delivery (MOD)	**NVD**	N. of cases	11	20	31	
		%	19.6%	35.7%	27.7%	0.057
	C-section	N. of cases	45	36	81	
		%	80.4%	64.3%	72.3%	

Table III: Descriptive statistics of neonatal NRBs, Hb, leukocytes and CRP age (n=112)

Variables	Study groups	N	Mean±SD	p-value
NRBCs	Control	56	1.69±2.42	
	Cases	56	16.62±17.10	0.0001
HB (g/dl)	Control	56	13.85±4.44	
	Cases	56	14.46±4.22	0.781
Leukocytes	Control	56	15.22±4.64	
	Cases	56	21.14±14.28	0.417
CRP	Control	56	2.00±0.00	
	Cases	56	1.91±0.28	0.544
Platelets	Control	56	279.50±81.40	
	Cases	56	198.31±62.18	0.018
Creatinine	Control	56	0.500±0.115	
	Cases	56	3.0365±2.051	0.001
AST	Control	56	13.50±0.57	
	Cases	56	135.76±85.02	0.001
ALT	Control	56	6.60±0.33	
	Cases	56	46.92±15.10	0.011

Table IV: Average of neonatal NRBs according to gender in cases and controls (n=112)

Study groups	Gender	N	Mean±SD	p-value
Controls	Female	28	2.28±2.96	0.069
	Male	28	1.10±1.57	
Cases	Female	14	16.42±7.27	0.961
	Male	42	16.69±19.38	
Severity of perinatal hypoxia (n=56)	Mild	26	8.38±5.52	0.0001
	Moderate	19	18.15±14.25	
	Severe	11	33.45±25.96	

Average of neonatal NRBs were statistically insignificant according to gender and MOD in both cases, and the control p-values were quite insignificant. Average NRBC count was significantly high in cases of cardiomegaly, those having ventilation need and respiratory distress (*p-<0.05*). Although the average of neonatal NRBs was significantly associated with the severity of perinatal hypoxia (p-0.0001), Table IV

Discussion

Perinatal asphyxia is still a clinical major problem through the world, considering the potentially adverse outcomes. The severe neonatal asphyxia can damage various organs, such as the brain, the heart, the lungs, necrotizing enterocolitis, the kidneys and the hepatic incompetence; it can even put the survival of a newborn at risk.[13] It has been observed that the raised NRBCs in the blood of umbilical cord could be a helpful indicator of birth asphyxia.[14] In this study 112 newborns were studied, to assess the association of NRBC with perinatal hypoxia and we found average of NRBCs was seen significantly high in cases 16.62±17.10 as compared to control group as 1.69±2.42 (p=0.0001) along with the average of neonatal NRBs was significantly associated to severity of perinatal hypoxia (p-0.0001). Consistently Meena P et al[15] reported that the average of NRCs count was significantly higher 17.82±19.55 in the with birth asphyxia, in contrast to controls 1.42±3.26, and furthermore they found a strong negative correlation of NRBCs with the severity of birth asphyxia. Kumar A et al[16] also found almost similar findings as the average nRBCs was markedly higher 20.97±8.17 in the cases of Perinatal asphyxia, compared to the controls (5.57±1.98) (p-0.0001) On the other hand, Jyoti Biswas et al[17] reported that the average nRBCs was significantly higher (54.06±22.42) in case group as compared to controls (10.32±5.86) (p-0.001). In the line of this study, in few previous publications, it has been revealed that NRBC is a haematological marker in newborns and that it has a connection to intrauterine hypoxia.[10,18,19] Like a result, in hypoxic situations like prenatal hypoxia, enhanced erythropoiesis and the subsequent release of NRBCs into the infant's blood circulation serve as a compensating response. Additionally, it has been proposed that an elevated NRBC count not only indicates prenatal hypoxia but also indicate the possibility of development of risk of the neurological complications.[10,20,21]

In this study mean age and birth weight were statistically significant (p= <0.05), while mean length and mean gestational age were statistically insignificant according to both groups (p=>0.05). In this study males were in majority in cases group (75.0%) and females were 25.0%, while in control group both male and female newborns were equal, findings were statistically significant according to both groups (p= 0.006). Rate of C-section was significantly higher 80.4% in control group, while 64.3% C-section rate was in cases group out of 56 cases in each group (p= 0.057). There was mean of creatinine level, AST and ALT were significantly high and platelets average significantly lower in case group in contrast to control group (p= <0.05), while Hb, Leukocytes and CRP were statistically insignificant according to both study groups (p=>0.05). Furthermore, the average of neonatal NRBs were statistically insignificant according to gender and MOD in both cases and controls (p >0.05), while NRBC count average was significantly high in cases of cardiomegaly, those having ventilation need and respiratory distress (p-<0.05). Consistently Kumar A et al[16] reported that there was no significant difference in the cases and controls according to average age of the mothers, mean age of gestation, average weight of birth, and gender of the newborns. On the other hand, Meena P et al[15] also found similar findings as there was no significant difference in both case and control groups in accordance to maternal age, birth weight, gestational age, maternal hemoglobin and gender of the newborns. Considering the need of locating laboratory and clinical indicators that offer quick prognostic data on stability of the patients, the increased nRBC counts could be helpful in improving predictive model for newborns.[22]

Conclusion

The mean NRBC was observed to be significantly associated (higher) in cases compared to controls. The NRBC was considered to be a non-invasive, simple prognostic marker for neonatal asphyxia, may indicate substantial severity of perinatal hypoxia and its related complications. This was a limited sample size and single-center study with several other limitations. However, to evaluate the more considerable clearance rate of the NRBCs in terms of the predictive value, the finding must be further explored by large scale studies specially at local level.

References

1. Sendeku FW, Azeze GG, Fenta SL. Perinatal asphyxia and its associated factors in Ethiopia: a systematic review and meta-analysis. BMC pediatrics. 2020 ;20(1):1-1. https://doi.org/10.1186/s12887-020-02039-3
2. Gillam-Krakauer M, Gowen Jr CW. Birth asphyxia. August 22, 2022. https://www.ncbi.nlm.nih.gov/books/NBK430782/
3. Viaroli F, Cheung PY, O'Reilly M, Polglase GR, Pichler G, Schmölzer GM. Reducing Brain Injury of Preterm Infants in the Delivery Room. Front Pediatr. 2018;6:290. https://doi.org/10.3389/fped.2018.00290
4. Mamo SA, Teshome GS, Tesfaye T, Goshu AT. Perinatal asphyxia and associated factors among neonates admitted to a specialized public hospital in South Central Ethiopia: A retrospective cross-sectional study. PLoS ONE 2022;17(1): e0262619. https://doi.org/10.1371/journal.pone.0262619
5. Simiyu IN, McHaile DN, Katsongeri K, Philemon RN, Msuya SE. Prevalence, severity and early outcomes of hypoxic ischemic encephalopathy among newborns at a tertiary hospital, in northern Tanzania. BMC pediatrics. 2017;17(1):131. https://doi.org/10.1186/s12887-017-0876-y
6. Wosenu L, Worku AG, Teshome DF, Gelagay AA. Determinants of birth asphyxia among live birth newborns in University of Gondar referral hospital, northwest Ethiopia: A case-control study. PloS one. 2018;13(9):e0203763. https://doi.org/10.1371/journal.pone.0203763
7. Sunny AK, Paudel P, Tiwari J, Bagale BB, Kukka A, Hong Z, Ewald U, Berkelhamer S, Ashish KC. A multicenter study of incidence, risk factors and outcomes of babies with birth asphyxia in Nepal. BMC pediatrics. 2021;21(1):1-8. https://doi.org/10.1186/s12887-021-02858-y
8. Ghosh B, Mittal S, Kumar S, Dadhwal V. Prediction of perinatal asphyxia with nucleated red blood cells in cord blood of newborns. International Journal of Gynecology & Obstetrics. 2003;81(3):267-71. https://doi.org/10.1016/S0020-7292(03)00124-3
9. Gebregziabher GT, Hadgu FB, Abebe HT. Prevalence and associated factors of perinatal asphyxia in neonates admitted to ayder comprehensive specialized hospital, Northern Ethiopia: a cross-sectional study. International journal of pediatrics. 2020;2020. https://doi.org/10.1155/2020/4367248
10. Boskabadi H, Zakerihamidi M, Sadeghian MH, Avan A, Ghayour-Mobarhan M, Ferns GA. Nucleated red blood cells count as a prognostic biomarker in predicting the complications of asphyxia in neonates. The Journal of Maternal-Fetal & Neonatal Medicine. 2017;30(21):2551-6. https://doi.org/10.1080/14767058.2016.1256988
11. Colaco SM, Ahmed M, Kshirsagar VY, Bajpai R. Study of nucleated red blood cell counts in asphyxiated newborns and the fetal outcome. International Journal of Clinical Pediatrics. 2014; 3(3):79-85. https://doi.org/10.14740/ijcp169w
12. Robertson NJ GF. Hypoxic ischemic brain injury. In: JM IR, editor. china: Elsevier. 2012; 1136-37
13. Yu Y, Gao J, Liu J, Tang Y, Zhong M, He J, Liao S, et al. Perinatal maternal characteristics predict a high risk of neonatal asphyxia: A multi-center retrospective cohort study in China. Frontiers in medicine. 2022;9. https://doi.org/10.3389/fmed.2022.944272
14. Goel M, Dwivedi R, Gohiya P, Hegde D. Nucleated red blood cell in cord blood as a marker of perinatal asphyxia. Journal of Clinical Neonatology. 2013 Oct 1;2(4):179. https://doi.org/10.4103/2249-4847.123097
15. Meena P, Gunawat M, Suman RL. Correlation of nucleated red blood cells with severity of birth asphyxia and its immediate outcome in term newborns. Int J Contemp Pediatr 2016;3: 841-6 https://doi.org/10.18203/2349-3291.ijcp20161430
16. Kumar A, Singh RD, Suryavanshi S, Mohan K. Cord blood nucleated red blood cell count as a predictor of long term sequelae in cases of perinatal asphyxia: a one-year follow-up study. Int J Contemp Pediatr 2018;5:1805-10 https://doi.org/10.18203/2349-3291.ijcp20183510
17. Jyoti Biswas, Alka Bhambri, Vedika A. Bhat. A clinical study for prediction of perinatal asphyxia using nucleated red blood cells as an indicator in umbilical cord blood of newborns. International Journal of Contemporary Medical Research

2020;7(1):A12-A15.
https://doi.org/10.21276/ijcmr.2020.7.1.25

18. Antonucci R, Porcella A, Pilloni MD. Perinatal asphyxia in the term newborn. JPNIM 2014;3:e030269
19. Buonocore G, Perrone S, Gioiaa D, et al. Nucleated red blood cell count at birth as an index of perinatal brain damage. Am J Obstet Gynecol. 1999;181:1500-5
https://doi.org/10.1016/S0002-9378(99)70396-0
20. McCarthy J, Capullari T, Thompson Z, et al. Umbilical cord nucleated red blood cell counts: normal values and the effect of labor. J Perinatol 2006;26:89-92
https://doi.org/10.1038/sj.jp.7211437
21. Hermansen M. Nucleated red blood cells in the fetus and newborn. Arch Dis Child Fetal Neonatal Ed 2001;84:F211-F5
https://doi.org/10.1136/fn.84.3.F211
22. Morton SU, Brettin K, Feldman HA, Leeman KT. Association of nucleated red blood cell count with mortality among neonatal intensive care unit patients. Pediatrics & Neonatology. 2020;61(6):592-7.
https://doi.org/10.1016/j.pedneo.2020.07.009