ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

ORIGINAL RESEARCH

Nucleated Red Blood Cells: Predictor of Prognosis & Mortality Risk in Critically Ill Patients

Veeresh Kumar Dhanni[1], Sandhya Chauhan[1], Anmol Gera[1]

[1]Assistant Professor, Department of General Medicine, TMMC & RC, Moradabad, Uttar Pradesh, India

Corresponding Author:
Dr. Veeresh Kumar Dhanni,
Assistant Professor, Department of General Medicine, TMMC & RC, Moradabad, Uttar Pradesh, India.
Email: veeresh.dhanni@gmail.com

Received Date: ? Acceptance Date: ?

Abstract
Background: To study Nucleated Red Blood Cells as predictor of prognosis & mortality risk in critically ill patients.
Material and Methods: Critically ill patients with A.P.A.C.H.E II score > 25 were enrolled in the study. NRBC positive & NRBC negative groups were made. Prognosis & Mortality of both groups were compared and analyzed.
Results: N.R.B.C was absent in 144 (72%) patients whereas present among 56 (28%) patients including 41 (28.87%) male patients whereas 15 (25.86%) female patients. The patients in whom N.R.B.C was present reported mean age as 58.97 years whereas not having N.R.B.C was 57.29 years. The patients with N.R.B.C reported mean BMI as 26.33Kg/m2 whereas, the patients without N.R.B.C was 25.95Kg/m2. N.R.B.C was present among patients having pulmonary diseases (28.57%) followed by gastrointestinal diseases (21.43%) and acute coronary syndrome (16.07%). Patients with N.R.B.C was present reported mean intensive care treatment (days) as 8.22 days whereas patients without N.R.B.C was 3.41 days with $p<0.01$. The patients with N.R.B.C reported mean A.P.A.C.H.E. II score to be 31.67 whereas not having N.R.B.C was 25.49. N.R.B.C was present in 27(48.21%) patients who deceased whereas among 29 (51.79%) patients who being discharged. Area under the R.O.C curve was 0.894. N.R.B.C.s in blood showed sensitivity and specificity of 72.3% and 90.6%, respectively.
Conclusion: When N.R.B.C concentration increased, the predictive value for mortality is increased. Because N.R.B.C.s are found in blood relatively early before death, screening for N.R.B.C.s could aid in the early detection of high-risk patients.
Keywords:

Introduction

In critically and chronically unwell patients, N.R.B.C.s, also known as Normoblasts or erythroblasts, have been explored as a predictor of higher in-hospital mortality and poor clinical outcomes. N.R.B.C.s has been employed as a biomarker to identify substantial hypoxia and inflammatory damage.[1] N.R.B.C.s are rarely found in the peripheral blood; nonetheless, their presence in bloodstream is linked to serious illnesses and suggests a poor prognosis.[2,3] N.R.B.C in the blood was linked to higher levels of erythropoietin, IL-3, and IL-6. As a result, the detection of N.R.B.C could think of as metric for calculating the severity of life-threatening hypoxia and inflammatory damage.[4-6] Nucleated RBCs (N.R.B.C.s) in blood sample are normally seen in the neonates up to the fifth day of life.[7] 3-10N.R.B.C.s/100WBC is normal count at that stage, which might be increased in conditions like Premature Birth & Fetal Hypoxia.[8-9] But the presence of N.R.B.C.s in adult or after infant stage, without any myelophthisic condition, means that bone marrow barrier has been disrupted or extra-medullary hematopoiesis has been activated. Furthermore, the existence of N.R.B.C.s with in venous blood of critically sick patients - without a hematological cause - has been linked to a bad prognosis, with mortality being proportional to the number of N.R.B.C.s in the blood.[10] As erythropoiesis intensifies in an attempt to compensate for acute anaemia (A.A), fast hemolysis (F.H) or blood loss can cause N.R.B.C.s to be discharged into the bloodstream. N.R.B.C.s is also discharged into the peripheral circulation when the bone marrow is damaged or stressed, as is typically the case with hematologic disorders.[11] For adults and children, a normal N.R.B.C.s reference range is 0 N.R.B.C.s/100WBCs.[12] The "gold standard" for laboratory identification and counting of N.R.B.C.s in peripheral blood is still direct light microscopy (L.M) of May-Grünwald-Giemsa (M.G.G) stained blood smears.[13] In our daily intensive care practise, finding a good marker for mortality and morbidity will always be a challenge. Intensivists are constantly concerned about whether to terminate therapy and when to transfer patients to wards. Finding the significance of N.R.B.C.s in critically ill patients will not only help in prognosticating the disease, but will also help in tailoring the treatment, making it early and aggressive in N.R.B.C positive patients.

ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

Material and Methods

Study design and place: This study is carried out at TMU Hospital (TMMC&RC) as a Prospective Observational Hospital Based research after obtaining consent as well as permission from the respective authorities.

Study population: Our target/study population was 200 critically ill patients admitted to Medicine Department of TMU Hospital under ICU care The period of study was 12 months duration from December 2019 to December 2020

Inclusion criteria:

Patients with age more than or equal to 19 years admitted to Medicine Department ICU care. With A.P.A.C.H.E.II score more than or equal to 25; i.e., Critically Ill.[14,15]

Exclusion criteria:

Any myelophthisic, haemolytic or post hemorrhagic condition. Patient on haematinics or erythropoietin.

N.R.B.C Measurement

N.R.B.C counts were measured using a XN350 Sysmex multi-parameter automated hematology Analyzer.

Peripheral blood smears were prepared with reagent and stains; then leukocytes, basophils and nucleated erythrocytes were counted by the help of light microscope.

Results

Table 1: Incidence of N.R.B.C in blood of critically ill patients

N.R.B.C	No. of cases	%
Present	56	28
Absent	144	72
Total	200	100

This prospective observational study was carried out at TMU Hospital (TMMC&RC) in 200 critically ill patients admitted to Medicine Department of TMU Hospital within ICU care. [Table 1] shows incidence of N.R.B.C in blood of critically ill patients. Out of total 200 patients, N.R.B.C was absent in 144(72%) patients whereas present in 56 (28%) patients.

Table 2: Gender distribution according to N.R.B.C

Gender	N.R.B.C				Total	
	Present		Absent			
	N=56	%	N=144	%	N=200	%
Male	41	28.87	101	71.13	142	71
Female	15	25.86	43	74.14	58	29
Chi Square	2.93					
p value	0.82					

[Table 2] shows gender wise distribution according to presence of N.R.B.C in the patients. N.R.B.C was present in 41(28.87%) male patients whereas in 15(25.86%) female patients. On applying the chi square test to compare the gender wise distribution according to presence of N.R.B.C in the patients, insignificant difference was found (p=0.82).

Table 3: Mean age according to N.R.B.C

N.R.B.C	Age		t test	p value
	Mean	SD		
Present	58.97	7.26	0.78	0.54
Absent	57.39	8.05		
Total	58.11	7.48		

[Table 3] shows mean age of patients according to presence of N.R.B.C. The patients in which N.R.B.C was present reported mean age as 58.97 years whereas the mean age of patients not having N.R.B.C was 57.29 years. During the comparison of mean age of patients divided on the basis of presence of N.R.B.C, insignificant difference was found (p=0.54).

Table 4: Mean BMI according to N.R.B.C

N.R.B.C	BMI (kg/m^2)		t test	p value
	Mean	SD		
Present	26.33	3.29	0.56	0.71
Absent	25.95	3.18		
Total	26.04	3.24		

ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

[Table 4] shows mean BMI of patients according to presence of N.R.B.C. The patients in which N.R.B.C was present reported mean BMI as 26.33 Kg/m2 whereas the mean BMI of patients not having N.R.B.C was 25.95 Kg/m2. During the comparison of mean BMI of patients divided on the basis of presence of N.R.B.C, insignificant difference was found (p=0.71).

Table 5: Diagnosis according to N.R.B.C

Diagnosis	**N.R.B.C**				**p value**
	Present		**Absent**		
	N=56	**%**	**N=144**	**%**	
Acute coronary syndrome/AMI	9	16.07	62	43.06	0.021*
Cardiac arrhythmia	4	7.14	18	12.50	0.42
Cardiac insufficiency	6	10.71	8	5.56	0.09
Pulmonary diseases	16	28.57	13	9.03	0.003*
Gastrointestinal diseases	12	21.43	10	6.94	0.006*
Cerebral diseases	4	7.14	18	12.50	0.18
Toxication	3	5.36	7	4.86	0.69
Metabolic diseases	1	1.79	6	4.17	0.36
Other diagnosis	1	1.79	2	1.39	0.89

*: statistically significant

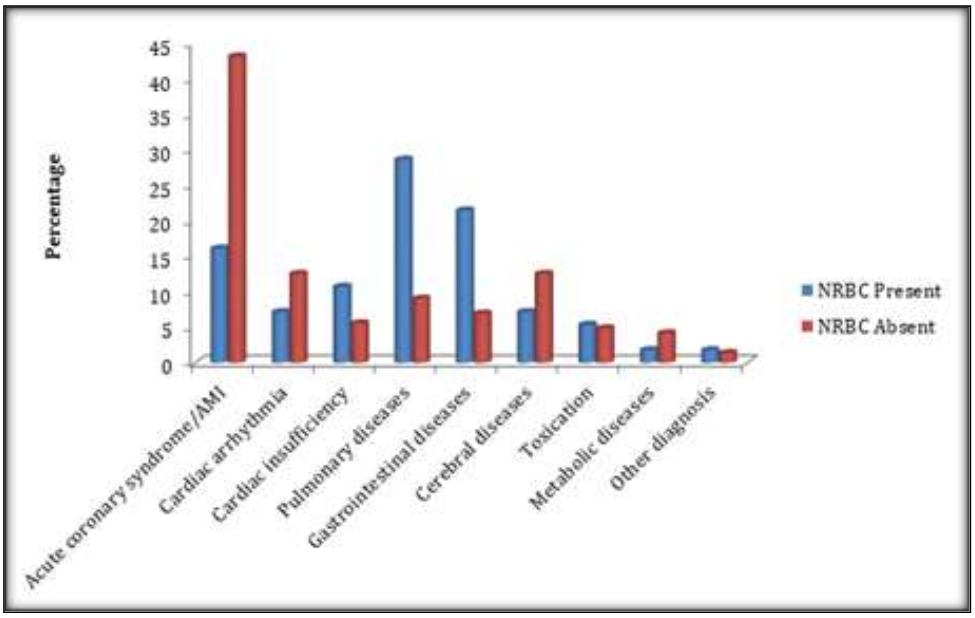

Figure 1: Diagnosis according to N.R.B.C

[Table 5, Figure 1] shows diagnosis according to N.R.B.C in the patients. N.R.B.C was present among patients having pulmonary diseases(28.57%) followed by gastrointestinal diseases(21.43%) and acute coronay syndrome(16.07%). On applying the chi square test to compare the presence of N.R.B.C in relation to specific diagnosis in the patients, the difference in proportion was found to be statistically significant for Acute coronary syndrome/AMI(p=0.021), pulmonary diseases(p=0.003) and Gastrointestinal diseases(p=0.006).

Table 6: Mean intensive care treatment (days) according to N.R.B.C

N.R.B.C	**ICU (days)**		**t test**	**p value**
	Mean	**SD**		
Present	8.22	2.08	24.51	<0.01*
Absent	3.41	0.84		

*: statistically significant

[Table 6] shows mean intensive care treatment (days) of patients according to presence of N.R.B.C. The patients in which N.R.B.C was present reported mean intensive care treatment (days) as 8.22 days whereas the mean intensive care treatment (days) of patients not having N.R.B.C was 3.41 days. During the comparison of mean intensive care treatment (days) of patients divided on the basis of presence of N.R.B.C, significant difference was found (p<0.01).

ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

Table 7: Comparison of mean A.P.A.C.H.E.II according to N.R.B.C

N.R.B.C	A.P.A.C.H.E.II		t test	p value
	Mean	SD		
Present	31.67	4.79	21.08	<0.01*
Absent	25.49	3.91		

*: statistically significant

[Table 7] shows A.P.A.C.H.E.II of patients according to presence of N.R.B.C. The patients in which N.R.B.C was present reported mean A.P.A.C.H.E.II score of 31.67 whereas the mean intensive care treatment (days) of patients not having N.R.B.C was 25.49. During the comparison of mean A.P.A.C.H.E.II score divided on the basis of presence of N.R.B.C, significant difference was found (p<0.01).

Table 8: Mortality according to N.R.B.C

Mortality	N.R.B.C				Total	
	Present		Absent			
	N=56	%	N=144	%	N=200	%
Present	27	48.21	13	9.03	40	20
Absent	29	51.79	131	90.97	160	80
Chi Square	16.74					
p value	<0.01*					

*: statistically significant

[Table 8, Figure 2] shows patients who deceased, their distribution according to presence of N.R.B.C in them. N.R.B.C was present in 27(48.21%) patients who deceased whereas among 29(51.79%) patients who being discharged. On applying the chi square test to compare the mortality presence wise distribution according to presence of N.R.B.C in the patients, significant difference was found (p<0.01). Mortality increased with increasing N.R.B.C concentration.

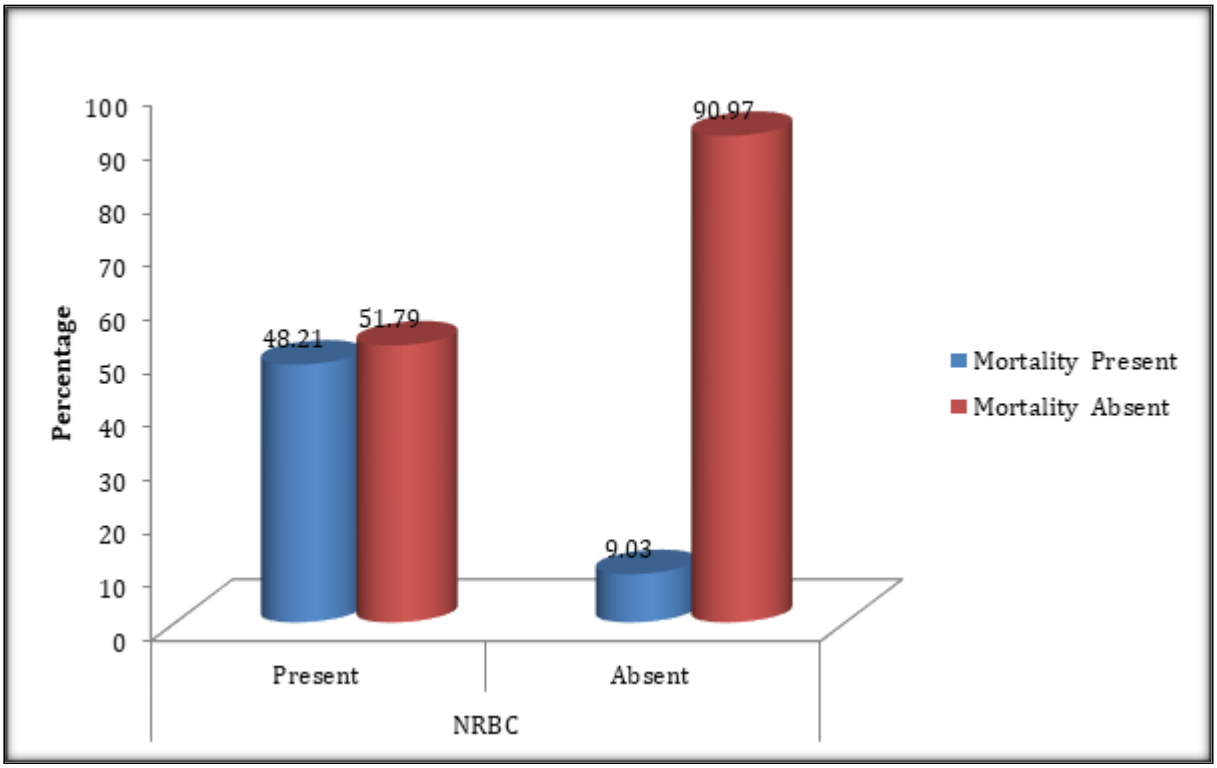

Figure 2: Mortality according to N.R.B.C

Discussion

This prospective observational study was carried out at TMU Hospital (TMMC&RC) on 200 consecutive critically ill patients admitted to Medicine Department of TMU Hospital under ICU care. The aim of the study N.R.B.C.s as predictor of prognosis & mortality risk in critically ill patients. Out of total 200 patients, N.R.B.C was absent in 144 (72%) patients whereas present among 56(28%) patients. A study conducted by Stachon A et

ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

al,[2] reported incidence of N.R.B.C.s 17.5% among critically ill patients, and in another study conducted by same author reported incidence of N.R.B.C.s in the blood as 19.2% among critically ill patients at least once. N.R.B.C was present in 41(28.87%) male patients whereas in 15(25.86%) female patients with statistically insignificant difference (p=0.82). The patients in whom N.R.B.C was present reported mean age as 58.97 years whereas not having N.R.B.C was 57.29 years. Bolulu et al.[16] (2005) found a significant increase in the incidence of N.R.B.C.s with age (P=0.01), with N.R.B.C-positive patients averaging 61 years of age compared to 56 years for N.R.B.C-negative patients. The patients in whom N.R.B.C was present reported mean BMI as 26.33Kg/m^2 whereas the mean BMI of patients not having N.R.B.C was 25.95Kg/m^2 with statistically insignificant difference. A study conducted by Stachon A et al,[2] reported mean BMI as 27.08Kg/m^2 among N.R.B.C positive patients whereas 26.5Kg/m^2 among N.R.B.C negative patients. N.R.B.C was present among patients having pulmonary diseases (28.57%) followed by gastrointestinal diseases (21.43%) and acute coronary syndrome (16.07%). On applying the chi square test to compare the presence of N.R.B.C in relation to specific diagnosis in the patients, the difference in proportion was found to be statistically significant for Acute coronary syndrome/AMI (p 0.021), pulmonary diseases (p 0.003) and G.I.T (p 0.006).The patients in which N.R.B.C was present reported mean intensive care treatment (days) as 8.22 days whereas the mean intensive care treatment (days) of patients not having N.R.B.C was 3.41 days with p<0.01. Madalina D,[17] reported that the length of stay in the ICU was on average 7.1 days for critically ill patients. The patients with N.R.B.C reported mean A.P.A.C.H.E.II score to be 31.67 whereas not having N.R.B.C was 25.49, with statistically significant difference (p<0.01). Stachon A et al,[2] in their study revealed similar results too i.e. N.R.B.C level increases with A.P.A.C.H.E.II score too. N.R.B.C was present in 27(48.21%) patients who deceased whereas among 29(51.79%) patients who being discharged. Another study conducted by same author reported the mortality of N.R.B.C-positive patients was 42.0%;[3] this was significantly higher (p-0.001) than the mortality of N.R.B.C-negative patients (5.9%). On applying the chi square test to compare the mortality presence wise distribution according to presence of N.R.B.C in the patients. Area under the R.O.C curve was 0.894. N.R.B.C.s in blood showed sensitivity and specificity of 72.3% and 90.6%, respectively. (R.O.C A.U.C 0.89; 95% CI 0.827–0.961; $p < 0.001$).

Conclusion

When N.R.B.C concentration increased, the predictive value for death increased as well, but it appears to decline after the N.R.B.C.s are no longer in circulation. Other laboratory and clinical risk indicators had little effect on the prognostic relevance of N.R.B.C.s. It appears that existing risk models, such as A.P.A.C.H.E II, can be improved. Moreover, because N.R.B.C.s are found in blood relatively early before death, screening for N.R.B.C.s could aid in the early detection of high-risk patients. Although more research is needed to see if identifying N.R.B.C.s might help doctors decide whether or not to adjust a patient's therapy, our current findings suggest that N.R.B.C-positive patients should continue to get intensive care.

References

1. Schreier S, Triampo W. The Blood Circulating Rare Cell Population. What is it and What is it Good For?. Cells. 2020 ;9(4):790.
2. Stachon A, Segbers E, Holland-Letz T, Kempf R, Hering S, Krieg M. Nucleated red blood cells in the blood of medical intensive care patients indicate increased mortality risk: a prospective cohort study. Crit Care. 2007;11(3):1-8.
3. Stachon A, Holland-Letz T, Kempf R, Becker A, Friese J, Krieg M. Poor prognosis indicated by nucleated red blood cells in peripheral blood is not associated with organ failure of the liver or kidney. Clin Chem Lab Med (CCLM). 2006;44(8):955-61.
4. Latimer KS, editor. Duncan and Prasse's veterinary laboratory medicine: clinical pathology. John Wiley & Sons; 2011 Jul 26.
5. Hemoglobinometry A, Red C, Principle EI, Principle OD, Histogram E, Width RC. Principles and Practice of Clinical Hematology. Linne & Ringsrud's Clinical Laboratory Science-E-Book: The Basics and Routine Techniques. 2015;2:291.
6. Lokeshwar MR, Shah NK, Agarwal BR, Manglani MV, Sachdeva A, editors. Textbook of pediatric hematology and hemato-oncology. Jaypee Brothers Medical Publishers (P) Limited; 2016.
7. Ridgway D, Miller DR, Baehner RL. Blood disease of infancy and childhood: VIIth edition. JAMA.1995;274(8):660-666.
8. Crosby WH. Hyposplenism: an inquiry into the normal function of the spleen. Annual Rev Med. 1963;14:349-370.
9. Hanlon-Lundberg KM, Kirby RS. Nucleated red blood cells as a marker of acidemia in neonates. Am J Obstet Gynecol. 1999;181:196-201.
10. Stachon A, Sondermann N, Imohl M, Krieg M. Nucleated red blood cells indicate high risk of in-hospital mortality. J Lab Clin Med. 2002;140:407-412.
11. LE BEAU MM. Cytogenetics and gene rearrangement. Williams Hematology. 6th ed. New York, NY: McGraw-Hill. 2001:115-23.
12. Green DW, Mimouni F. Nucleated erythrocytes in healthy infants and in infants of diabetic mothers. J Pediatr. 1990;116:129-131.
13. Buoro S, Manenti B, Seghezzi M. Which clinical significance has automatic detection of very low levels of nucleated red blood cells in the peripheral blood?. Ann Translational Med. 2016;4(11).

ISSN: 0975-3583,0976-2833 VOL15, ISSUE 04, 2024

14. World Health Organizations: Publications on HIV Guidelines: Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: Chapters 1-4: Overview of the introductory chapters: Definition of key terms: Consolidated ARV guidelines: Jun2013: Chapters1-4.
15. Wagner DP, Draper EA. Acute physiology and chronic health evaluation (APACHE II) and Medicare reimbursement. Health Care Financing Review: Nov1984: Annual Supplement: 91-105.
16. Stachon A, Bolulu O, Holland-Letz T, Krieg M. Association between nucleated red blood cells in blood and the levels of erythropoietin, interleukin 3, interleukin 6, and interleukin 12p70. Shock. 2005;24(1):34-9.
17. Mădălina D, Negoiță S, Rely M, Calu V, Corneci D, Anca G, Cornelia T, Azamfirei L. The Presence of Nucleated Red Cells in the Blood of Critical Care Patients is Associated with an Increased Mortality Risk. Acta Medica Marisiensis. 2011;57(5).