

## Original Research Article

# Can red cell indices predict retinopathy of prematurity in preterm extreme low birth weight neonates? A single center retrospective study

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### ABSTRACT

**Background:** Improvement in neonatal health care services has led to the survival of extreme low birth weight babies over the years. This has led to increased number of retinopathy of prematurity (ROP) cases being diagnosed. Thus it becomes imperative to identify factors which can reliably predict preterm neonates at increased risk of ROP. Aims and objectives were to identify red cell indices at 4 weeks postpartum which can predict ROP in extreme low birth weight neonates.

**Methods:** Three years ROP data in extremely low birth weight neonates was retrospectively collected and analyzed.

**Results:** The mean gestational age at birth of the neonates in ROP group (n=149) and no-ROP group (n=191) was 28.25 ( $\pm 2.71$ ) weeks and 31.82 ( $\pm 2.24$ ) weeks, respectively ( $p < 0.05$ ). The mean birth weight of the neonates in ROP group and no-ROP group was 756.44 ( $\pm 95.50$ ) grams and 890 ( $\pm 109.20$ ) grams, respectively ( $p < 0.05$ ). In extremely low birth weight (ELBW) neonates, hematologic parameters such as hemoglobin, hematocrit, red blood cells, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration values were lower and white blood cell count was higher in ROP group as compared to no-ROP group ( $p < 0.05$ ).

**Conclusions:** Red cell indices may predict which extreme low birth weight neonates are at increased risk of developing retinopathy of prematurity. Being easily and widely available, red cell indices can be used as a screening test to predict ROP.

**Keywords:** Retinopathy of prematurity, Red cell indices, Preterm, Extreme low birth weight

### INTRODUCTION

Retinopathy of prematurity (ROP) is a vasoproliferative retinal disorder leading to blindness if not diagnosed and treated appropriately.<sup>1</sup> Improvement in neonatal health care services has led to the survival of extreme low birth weight babies over the years. This has led to increased number of ROP cases being detected and treated.<sup>2,3</sup> Thus it becomes imperative to identify factors which can reliably predict preterm neonates at increased risk of ROP. This would reduce the number of cases with advanced ROP and enable timely treatment. In addition, unnecessary and risky

ROP examinations in busy clinics may be avoided and the waste of time and effort prevented.

Considering the pathophysiology of ROP, the fluctuations in blood oxygen concentration alters not only angiogenic and inflammatory cytokine production in the retina but also the production of blood cells and inflammatory cytokines. Various studies have investigated platelet count, lymphocyte count, nucleated red blood cell and neutrophil-to-lymphocyte ratio (NLR).<sup>4-8</sup> In this study, we analyzed complete blood count (CBC) parameters at 4 weeks postpartum to identify whether red cell indices can predict ROP in extreme low birth weight neonates.

## METHODS

This retrospective study was conducted in the neonatology department of a tertiary care hospital between April 2015 and December 2017 and included 341 extremely low birth weight neonates (ELBW with birth weight <1000 grams). Ethical approval was obtained from the institutional ethics committee. The same ophthalmologist examined the infants according to the standards of the international committee for the classification of ROP (ICROP).<sup>9</sup>

Initial screening was done at gestational age (GA) of 31 weeks for those born before 27 weeks and at 4 weeks after birth for those born after 27 weeks' gestation. Examinations were performed 1 hour after the instillation of 1% phenylephrine and 0.5% tropicamide. Follow-up examinations and treatments were conducted according to the ICROP and ETROP criteria.<sup>9,10</sup>

The neonates were divided into 2 groups: "no-ROP" group which included babies with no signs of ROP while neonates with any stage of ROP were included in the "ROP group".

Data of complete blood count done at 4 weeks postpartum were retrieved from the medical record section of the hospital. Hemoglobin (Hb), white blood cell (WBC), neutrophil count, lymphocyte count, monocyte count, eosinophil count, basophil count, platelet count, hematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), MCH concentration (MCHC), red cell distribution width (RDW), red blood cell (RBC) were compared between the ROP and no-ROP groups. Neonates who had septicemia and those who were transfused with packed red cells before ROP examination were excluded from the study.

### Statistical analysis

Descriptive statistics of the normally distributed numeric variables were presented as mean ( $\pm$ standard deviation).

Mann-Whitney U test was used to compare non-normally distributed numeric variables between the groups. Independent-samples t test was used to compare the normally distributed numeric variables between the two groups. Factors significantly associated with ROP were determined with logistic regression analysis. P values  $\leq 0.05$  were considered statistically significant.

## RESULTS

Three hundred and forty (n=340) extreme low birth weight neonates were included in this study. The baseline characteristics of the study cohort are shown in Table 1. The mean gestational age and birth weight of the study population was 28.92 ( $\pm 2.05$ ) weeks and 894.03 ( $\pm 104.26$ ) grams, respectively. Male babies comprised of 50% (n=170) of the cohort. The no-ROP group consisted of 56% (n=191) and the ROP group consisted of 44% (n=149) of the study population. The mean gestational age at birth of the neonates in ROP group and no-ROP group was 28.25 ( $\pm 2.71$ ) weeks and 31.82 ( $\pm 2.24$ ) weeks, respectively (p<0.05). The mean birth weight of the neonates in ROP group and no-ROP group was 756.44 ( $\pm 95.50$ ) grams and 890 ( $\pm 109.20$ ) grams, respectively (p<0.05). Antenatal steroid coverage was almost equal in both the groups (p>0.05). Oxygen therapy and blood component transfusion were not significantly different in ROP group and no-ROP group (p>0.05).

In ELBW neonates, hematologic parameters such as Hb, Hct, RBCs, MCH and MCHC values were lower and WBC count was higher in ROP group as compared to no-ROP group (p<0.05). Table 2 depicts the comparison between the two groups. The risk of ROP development was positively correlated with red cell distribution width (OR=1.46, 95% CI=1.16-1.71, p<0.05) and white blood cell count (OR=1.18, 95% CI=1.67-1.45, p<0.05) and negatively correlated with Hb (OR=0.61, 95% CI=0.46-0.84, p<0.05) and platelet count (OR=0.92, 95% CI=0.89-0.97, p<0.05) when logistic regression analysis was carried out.

**Table 1: Baseline characteristics of the study cohort.**

Variable	No-ROP group (n=191)	ROP group (n=149)
Antenatal steroids, (n, %)	145 (51.4)	137 (48.6)
Male gender (n, %)	90 (53)	80 (47)
SGA, (n, %)	65 (46.8)	74 (53.2)
LSCS delivery, (n, %)	78 (47.6)	86 (52.4)
Respiratory support, (n, %)	130 (47.3)	145 (52.7)
Phototherapy, (n, %)	110 (45.8)	130 (54.2)
Oxygen therapy, (n, %)	125 (48.1)	135 (51.9)
Antibiotics, (n, %)	130 (47.3)	145 (52.7)
Parenteral nutrition, (n, %)	120 (48)	130 (52)
Formula milk, (n, %)	65 (52.8)	58 (47.2)
Blood component transfusion, (n, %)	52 (47.3)	58 (52.7)

**Table 2: Hematological parameters of ROP group and no-ROP group at 4 weeks postpartum.**

Variable	No-ROP group Mean ( $\pm$ SD)	ROP group Mean ( $\pm$ SD)	P value
Hemoglobin (g/dl)	13.1 ( $\pm$ 2.21)	11.3 ( $\pm$ 1.92)	<0.05
White blood cell count ( $\times 10^3$ /mcl)	9.16 ( $\pm$ 2.57)	11.45 ( $\pm$ 3.21)	<0.05
Neutrophil count ( $\times 10^3$ /mcl)	2.57 ( $\pm$ 0.91)	2.92 ( $\pm$ 0.74)	>0.05
Lymphocyte count ( $\times 10^3$ /mcl)	4.86 ( $\pm$ 1.95)	5.02 ( $\pm$ 1.50)	>0.05
Monocyte count ( $\times 10^3$ /mcl)	0.99 ( $\pm$ 0.56)	1.1 ( $\pm$ 0.49)	>0.05
Eosinophil count ( $\times 10^3$ /mcl)	0.29 ( $\pm$ 0.12)	0.32 ( $\pm$ 0.18)	>0.05
Basophil count ( $\times 10^3$ /mcl)	0.04 ( $\pm$ 0.01)	0.03 ( $\pm$ 0.01)	>0.05
Platelet count ( $\times 10^3$ /mcl)	312 ( $\pm$ 1.21)	334 ( $\pm$ 1.23)	>0.05
Hematocrit (%)	35.21 ( $\pm$ 5.12)	31.13 ( $\pm$ 4.87)	<0.05
Red blood cells ( $\times 10^6$ /mcl)	3.97 ( $\pm$ 0.95)	3.27 ( $\pm$ 0.65)	<0.05
Red cell distribution width (RDW) ratio	15.1 ( $\pm$ 1.4)	16.4 ( $\pm$ 1.3)	<0.05
Mean corpuscular volume (MCV) (fl)	93.2 ( $\pm$ 7.21)	90.32 ( $\pm$ 8.12)	>0.05
Mean corpuscular hemoglobin (MCH) (pg)	33.7 ( $\pm$ 6.2)	29.8 ( $\pm$ 4.1)	<0.05
Mean corpuscular hemoglobin concentration (MCHC) (g/dl)	34.7 ( $\pm$ 2.2)	32.1 ( $\pm$ 1.8)	<0.05

## DISCUSSION

In this study, CBC parameters were analyzed to determine the hematological parameters among ELBW neonates associated with development of ROP. As the process of hematopoiesis changes rapidly during initial days of life in preterm neonates, we decided to evaluate the hematological parameters at 4 weeks postpartum to avoid variations in red cell parameters.<sup>11</sup> Also, this is the time when first ROP screening is advised in extreme preterm neonates.

On logistic regression analysis, red cell distribution width and WBC count were associated with the development of ROP in ELBW neonates. Lower values of MCH in ROP group compared to no-ROP group was significantly associated with the development of ROP. This can be explained by the fact that MCH represents the mean Hb value in RBCs and Hb is critical for carrying the oxygen into the tissues. This association between MCH and ROP could originate from the nitric oxide (NO) pathways. The vasodilatation mediated by NO, is maintained in a delicate balance inside the tissues.<sup>12</sup> Excessive secretion of NO due to inflammation leads to vasodilatation, increased capillary permeability resulting in edema.<sup>13</sup>

There are some studies that have demonstrated the significance of red cell parameters in development of ROP, similar to our study. A study conducted in China in neonates born from multiple gestations, has found that anemia is an independent risk factor of ROP.<sup>14</sup> A study carried out by Banerjee et al did not find any correlation between hemoglobin levels and ROP on multiple logistic analysis which is in contrary to our study.<sup>15</sup> However, this contrast can be because of the methodological difference wherein the red cell parameters were derived on the first day of life. Studies have observed that higher nucleated RBC count on the first day of life is associated with intrauterine hypoxia; thus may be associated with development of ROP.<sup>7,8</sup>

Our study observed that WBC count along with the differential count including neutrophil, lymphocyte, monocyte and eosinophil counts were significantly higher in ROP group than No-ROP group. In the absence of any signs of infection, increased lymphocyte, neutrophil, monocyte and eosinophil counts in the ROP group could be because of the underlying inflammation. These cells induce NO release from tissues which in turn increases angiogenic factors such as VEGF.<sup>16</sup>

Our study also tried to find out if there is any association between platelet count and development of ROP. Platelets store and carry both pro and anti-angiogenic factors like platelet-derived growth factor (PDGF), epidermal growth factor (EGF) and VEGF; thus any quantitative change in platelet count may have a role in development of ROP.<sup>17,18</sup> Jensen et al observed that low platelet count is a risk factor for ROP, the finding is similar to our study.<sup>6</sup> Similar study by Yau et al concluded that thrombocytopenia is associated with ROP in babies of multiple gestation.<sup>14</sup>

## CONCLUSION

In conclusion, red cell indices may predict which ELBW neonates are at increased risk of developing ROP. Being easily and widely available, red cell indices can be used as a screening test to predict ROP. Further studies with prospective design are needed to confirm the findings.

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## REFERENCES

- O'Connor AR, Stephenson T, Johnson A, Tobin MJ, Moseley MJ, Ratib S, et al. Long-Term Ophthalmic Outcome of Low Birth Weight Children With and

- Without Retinopathy of Prematurity. *Pediatrics.* 2002;109:12-8.
2. Zin A, Gole GA. Retinopathy of prematurity-incidence today. *Clin Perinatol.* 2013;40:185-200.
  3. Bas AY, Koc E, Dilmen U. ROP Neonatal Study Group. Incidence and severity of retinopathy of prematurity in Turkey. *Br J Ophthalmol.* 2015;99:1311-4.
  4. Kurtul BE, Kabatas EU, Zenciroglu A, Ozer PA, Ertugrul GT, Beken S, et al. Serum neutrophil-to-lymphocyte ratio in retinopathy of prematurity. *JAAPOS.* 2015;19:327-31.
  5. Tao Y, Dong Y, Lu CW, Yang W, Li Q. Relationship between mean platelet volume and retinopathy of prematurity. *Graefes Arch Clin Exp Ophthalmol.* 2015;253:1791-4.
  6. Jensen AK, Ying GS, Huang J, Karp K, Quinn GE, Binenbaum G. Thrombocytopenia and retinopathy of prematurity. *JAAPOS.* 2011;15:3-4.
  7. Lubetzky R, Stolovitch C, Dollberg S, Mimouni FB, Salomon M, Mandel D. Nucleated red blood cells in preterm infants with retinopathy of prematurity. *Pediatrics.* 2005;116:619-22.
  8. Niranjan HS, Bharath Kumar Reddy KR, Benakappa N, Murthy K, Shivananda S, Veeranna V. Role of hematological parameters in predicting retinopathy of prematurity (ROP) in preterm neonates. *Indian J Pediatr.* 2013;80:726-30.
  9. An International Committee for the Classification of Retinopathy of Prematurity. The International Classification of Retinopathy of Prematurity revisited. *Arch Ophthalmol.* 2005;123:991-9.
  10. Early Treatment for Retinopathy of Prematurity Cooperative Group. Results of the early treatment for retinopathy of prematurity randomized trial: revised indications for the treatment of retinopathy of prematurity. *Arch Ophthalmol.* 2003;121:1684-96.
  11. Henry E, Christensen RD. Reference intervals in neonatal hematology. *Clin Perinatol.* 2015;42:483-97.
  12. Butcher JT, Johnson T, Beers J, Columbus L, Isakson BE. Hemoglobin alpha in the blood vessel wall. *Free Radic Biol Med.* 2014;73:136-42.
  13. Frost MT, Wang Q, Moncada S, Singer M. Hypoxia accelerates nitric oxide-dependent inhibition of mitochondrial complex I in activated macrophages. *Am J Physiol Regul Integr Comp Physiol.* 2005;288:394-400.
  14. Yau GSK, Lee JWY, Tam VTY, Yip S, Cheng E, Liu CC, et al. Incidence and risk factors for retinopathy of prematurity in multiple gestations; a Chinese population study. *Medicine.* 2015;94:867.
  15. Banerjee J, Asamoah FK, Singhvi D, Kwan AW, Morris JK, Aladangady N. Haemoglobin level at birth is associated with short term outcomes and mortality in preterm infants. *BMC Med.* 2015;27:16.
  16. Ashki N, Chan AM, Wang W, Kiyohara M, Lin L, Braun J, et al. Peroxynitrite upregulates angiogenic factors VEGF-A, BFGF, and HIF1 alpha in human corneal limbal epithelial cells. *Invest Ophthalmol Vis Sci.* 2014;55:1637-46.
  17. Italiano JE, Richardson JL, Patel-Hett S, Battinelli E, Zaslavsky A, Short S, et al. Angiogenesis is regulated by a novel mechanism: pro-and anti-angiogenic proteins are organized into separate platelet  $\alpha$  granules and differentially released. *Blood.* 2008;111:1227-33.
  18. Folkman J. Angiogenesis: an organising principle for drug discovery? *Nat Rev Drug Discov.* 2007;64:273-86.

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