EVALUATION OF HAEMOGLOBIN LEVEL AND PLATELET COUNT IN NEONATES WITH AND WITHOUT RETINOPATHY OF PREMATURITY

Dr Lim Zi Di

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Evaluation of Haemoglobin Level and Platelet Count in Neonates With and Without Retinopathy of Prematurity

PREPARED BY:

Department of Ophthalmology and Visual Science
School of Medical Sciences
Universiti Sains Malaysia

SUPERVISOR:

PROFESSOR DR. SHATRIAH ISMAIL

Consultant Paediatric Ophthalmologist
Department of Ophthalmology and Visual Science
School of Medical Sciences
Universiti Sains Malaysia

CO-SUPERVISOR:

PROFESSOR DR. HANS VAN ROSTENBERGHE

Consultant Neonatologist
Department of Paediatrics
School of Medical Sciences
Univerisiti Sains Malaysia

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DISCLAIMER

I hereby certify that the work in this dissertation is my own except for the quotations and summaries which have been duly acknowledged. I declare that I have no financial of interest in the instruments and the computer software used in this study.

Date 19/04/2021 ……..

Dr Lim Zi Di

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TABLE OF CONTENT

CHAPTER 5: APPENDIX

ABSTRAK

Matlamat: Terdapat andaian bahawa haemoglobin dan platelet mempengaruhi perkembangan retinopati pramatang. Kajian ini bertujuan untuk membandingkan tahap min hemoglobin mingguan dan jumlah platelet mingguan antara bayi retinopati pramatang dan bayi pramatang tanpa retinopati pramatang pada enam minggu pertama kehidupan.

Kaedah: Seramai 93 bayi pramatang dengan berat lahir kurang daripada 1.5 kg dan umur gestasi kurang daripada 32 minggu direkrut di Hospital Universiti Sains Malaysia dari 2017 ke 2019. Setiap kes retinopati pramatang dipadankan secara individu (1: 2) dengan dua kes tanpa retinopati pramatang. Min tahap hemoglobin mingguan, min jumlah platelet mingguan, dan faktor risiko lain yang berkaitan didokumentasikan.

Hasil: Tiga puluh satu bayi retinopati pramatang dan 62 bayi tanpa retinopati pramatang direkrut. Dalam kumpulan retinopati pramatang, 8 berada pada tahap 1, 6 pada tahap 2 dan 16 pada tahap 3 retinopati pramatang. Min Berat lahir dan umur gestasi kumpulan retinopati pramatang adalah 962.2 g dan 27.6 minggu manakala kumpulan tanpa retinopati pramatang adalah 1056.9 g dan 28.5 minggu masing-masingTerdapat perbezaan yang signifikan dalam jumlah min platelet mingguan antara bayi retinopati pramatang dan bayi tanpa retinopati pramatang pada enam minggu pertama kehidupan (p=0.002). Tiada perbezaan min didapati setelah kovariate diambil kira (p=0.489). Perbezaan yang signifikan juga dijumpai ketika membandingkan min tahap hemoglobin mingguan pada minggu pertama kehidupan (p=0.003) dan hilang setelah menyesuaikan dengan kovariat (p=0.292).

Kesimpulan: Tiada perbezaan signifikan dalam min hamoglobin and min platelet pada enam minggu pertama kehidupan antara bayi retinopati pramatang dan bayi tanpa retinopati pramatang selepas disesuaikan dengan kovariate seperti sepsis, transfusi darah dan displasia bronkopulmonari.

ABSTRACT

Aim: Haemoglobin and platelet have been postulated to play a role in the development of retinopathy of prematurity (ROP). This study aimed to compare weekly mean haemoglobin level and platelet count between ROP and non-ROP infants in the first six weeks of life.

Method: Ninety-three premature infants with birth weight less than 1.5 kg and gestational age less than 32 weeks were recruited in Hospital Universiti Sains Malaysia from 2017 to 2019. Each ROP case was individually matched (1: 2) to two non-ROP cases. Weekly mean haemoglobin level, weekly mean platelet count, and other related risk factors were documented.

Result: Thirty-one infants with ROP and 62 infants with non-ROP were recruited. Of those with ROP, 8 had stage 1 ROP, 6 had stage 2 ROP, 16 had stage 3 ROP. The mean birth weight and gestational age of the ROP group was 962.2 g and 27.6 weeks while the non-ROP group was 1056.9 g and 28.5 weeks respectively. We found significant differences in the weekly mean platelet counts between ROP and non-ROP infants from week two to week six of life (p=0.003). A significant difference was also found when comparing weekly mean haemoglobin level at week one of life (p=0.003). However, no significant difference was found in the weekly mean platelet count (p=0.489) and weekly mean haemoglobin level (p=0.292) after adjusting to covariates.

Conclusion: There is no significant difference in mean haemoglobin level and platelet count in the first six weeks of life between ROP and non-ROP infants after adjusting to confounding factors like sepsis, bronchopulmonary dysplasia and blood transfusion.

Keywords: retinopathy of prematurity, mean platelet count, mean haemoglobin level, first six weeks of life.

CHAPTER 1:

INTRODUCTION

1.1 Retinopathy of Prematurity

Retinopathy of prematurity (ROP) is a proliferative retinopathy of infants that are born prematurely. It is estimated that 32000 children developed severe visual impairment due to ROP yearly worldwide (Blencowe *et al.,* 2016). Early detection and effective management of ROP can prevent blindness.

Prior to understanding the pathology of ROP, we first look into the physiological retinal development. Retinal angiogenesis starts around 17 weeks period of gestation (POG) in utero and develops fully just prior to a full-term birth. Angiogenesis process is stimulated by a physiological hypoxic state in the womb (Hughes *et al.,* 2000). However, if the infant is born prematurely, it will expose the infant to a non-physiological hyperoxia state in phase 1 of ROP (Madan *et al*., 2003).This subsequently causes impediment of vascular growth and obliteration of vessels. When the infant's eyes start to mature, metabolic demand increases which could now not be supplied due to poor vascular growth (Chow *et al*., 2003), this in turn, will lead to phase 2, hypoxia-revascularisation of the eye.

For retina to meet the increased metabolic demand, it upregulates VEGF and IGF-1 to trigger angiogenesis. Angiogenesis at this phase may be seen without significant pathology, or it may lead to abnormal proliferation of retinal vessels in ROP (Smith *et al*., 2003). This phase usually occurs after 32 weeks POG; however, it has a wide range of onset (Good *et al.,* 2005). Even infants born at 32 weeks POG are at risk for developing ROP (Shah *et al.*, 2012).

1.2 Risk factors for ROP

There are many risk factors for ROP with birth weight (BW) less than 1.5 kg and period of gestation (POG) less than 32 weeks being the most significant cause. Oxygen therapy including ventilation is a major risk factor for ROP as repeated hyperoxia and hypoxia episodes are implicated in the pathogenesis of ROP and strict management of oxygen therapy without fluctuations and monitoring may help in reducing the risk of developing ROP (Bateman *et al.,* 2013).

Other risk factors that have been implicated in the development of ROP include blood transfusion (Cakir *et al.,* 2018), sepsis (Stone *et al.,* 2016), hyperglycaemia (Lee *et al.,* 2016), use of surfactant (Chen *et al.,* 2011), bronchopulmonary dysplasia (Lardon *et al.,* 2016), neonatal enterocolitis (Hair *et al.,* 2016), anaemia (Banerjee *et al.,* 2015) and thrombocytopenia (Lundgren *et al*., 2018). However there is currently still insufficient evidence to determine the degree of importance of these risk factors in contributing to the pathogenesis of ROP.

1.2.1 Association of Haemoglobin with ROP

Haemoglobin is a metalloprotein that acts as an oxygen carrier that is found in red blood cells. Its function to transport oxygen throughout the body. (Cohen *at el*., 2016). A decline in haemoglobin could result in anaemia.

Neonates have a drop in RBC during the first week of life. The decline is multifactorial with both pathological and physiological factor involved. In healthy term infants, haemoglobin could fall below 10g/dL at 10-12 weeks of life but is well tolerated and does not require treatment. This is referred to as "physiological anaemia of infancy". On the other hand, premature infant has a more rapid decline of haemoglobin which usually occurs during first 4-6 weeks of life to approximately 8g/dL for birth weight 1.0-1.5 kg and 7 g/dL for birth weight less than 1 kg. (Strauss *et al*., 2010). The common cause of such occurrence includes immaturity of the haematopoietic system (Crowley *et al*., 2010), iatrogenic blood loss from surgery or blood taking and insufficient production of erythropoietin (Strauss *et al*., 2010). Almost all extremely preterm infants had receive blood transfusions during hospitalisation (Crowley *et al.*, 2010).

To our knowledge, there is still no clear explanation of the relationship between anaemia and the development of ROP. Few studies have been done to determine whether anaemia is a risk factor for ROP; however, the results remain controversial (Table 1). In 2001, a study was done in America, suggesting that anaemia is protective of ROP development (Englert *et al*., 2001). More recent studies suggested otherwise. It was found in a study that low haemoglobin level at birth is associated with ROP development (Banerjee *et al*., 2015) while the other two studies suggested that anaemia in the first week of life is associated with ROP (Lundgren *et al*.,

2018,2019). Our study thus aimed to investigate further by comparing mean weekly mean haemoglobin between infants with and without ROP.

Table 1: Summary of literature associating ROP with Haemoglobin

Name	Country	Year	Sample size	Result	Conclusion
Englert *et al*	USA	2001	107	p = 0.17	Anaemia did not affect severe ROP as independent risk factor
Banerjee *et al*	UK	2015	920	OR 3.2 95%CI (1.5-6.4)	Low haemoglobin at birth is associated with ROP in premature infant
Lundgren *et al*	Sweden	2018	227	OR 1.46 95%CI (1.16-1.83)	Anaemia during the first week postnatal was an independent risk factor for ROP
Lundgren *et al*	Sweden	2019	78	P = 0.003	Anaemia during first week postnatal is a significant risk factor for severe ROP

1.2.2 Association of Platelet with ROP

The main function of platelets is for haemostasis. The normal range of platelet in a neonate is 150- 500 × 10^3/ microL and some author suggest premature infant have a lower limit of normal. (Clakravorty *et al*., 2012). Like haemoglobin, platelet count falls after birth for the first few days but then increases after the first week of life. (Bolat *et al*., 2012). Low platelet count (< 150 × 10^3/ microL) is called thrombocytopenia while high platelet count (> 500 × 10^3/ microL) is called thrombocytosis. Both of which could be associated with ROP.

Platelets have been implicated in the pathogenesis of ROP, as they contain both pro-angiogenic and anti-angiogenic cytokines like VEGF (Vascular-Endothelial Growth Factor), PDGF (Platelet-Derived Growth Factor) and BDNF (Brain-Derived Neurotrophic Factor) (Battinelli *et al*., 2019). Platelets are a significant source of VEGF, and platelet counts are related to systemic VEGF levels (Aksoy *et al.*, 2014). A recent study concluded that there is a strong correlation between platelet count and systemic VEGF-A, PDGF-BB and BDNF in ROP infants (Hellgren *et al.,* 2021). However, whether ocular VEGF levels are reflective of platelet levels remains controversial (Aksoy *et al.*, 2014).

To the current date, the relationship between platelet counts and ROP remains poorly defined, with most studies observing an association between thrombocytopenia and ROP development or severity (Cakir *et al.*, 2018; Jensen *et al.*, 2018; Sancak *et al.*, 2019), others found no association (Bourla *et al.*, 2008; Korkmaz *et al.*, 2018), and one recent study even documenting that thrombocytosis is associated with ROP (Del Rey *et al.*, 2019). Our study thus aimed to

investigate further by comparing mean weekly platelet counts between infants with and without ROP.

Table 2: Summary of literature associating ROP with Platelet Count and thrombocytopenia

Name	Country	Year	Sample size	Result	Conclusion
Bourla *et al*	USA	2008	178	p = 0.689	No association between thrombocytopenia and ROP
Rastogi *et al*	USA	2011	286	p < 0.001	A >30% drop in platelet counts associated with ROP is independent of thrombocytopenia
Jensen *et al*	USA	2011	161	OR 6.69 95%CI (2.82-15.9)	Thrombocytopenia is associated with severe ROP, primarily in zone 1 ROP
Lundgren *et al*	USA	2017	18	p < 0.001	Thrombocytopenia at time of ROP diagnosis is associated with APROP development
Korkmaz *et al*	Turkey	2017	146	p > 0.05	Platelet count do not differ between premature infant with and without ROP
Cakir *et al*	USA	2018	202	OR 2.97 95%CI (1.37–6.46)	Thrombocytopenia is independently associated with severe ROP
Sancak *et al*	Turkey	2018	182	OR 59.0 95%CI (51.14-71.0)	There is a significant association between thrombocytopenia and Type I ROP
Jensen *et al*	USA	2018	100	OR 2.8 95%CI (1.4-5.6)	Thrombocytopenia from birth to 34 weeks POG is associated with subsequent severe ROP
Del Rey *et al*	Spain	2019	193	p < 0.001	Late thrombocytosis at one month postnatal may be associated with ROP

Retinopathy of Prematurity, ROP

Aggressive Posterior Retinopathy of Prematurity, APROP

Period of Gestation, POG

1.3 Rationale of the study

To the current date, the relationship between platelet counts and haemoglobin level with ROP remains poorly defined with related studies showing conflicting result. There is currently insufficient evidence to determine the degree of importance of low haemoglobin level and platelet count in contributing to the pathogenesis of ROP. This study aims to provide clarity on this subject and impart better understanding of the relationship between ROP, haemoglobin and platelet.

As an infant's life is vulnerable, more so their eyes. Once an infant develops severe ROP, it will be an uphill battle for the ophthalmologist to restore their vision. Therefore it is known that prevention is better than treatment in ROP. With understanding the full aspect of the disease, neonatologists will have a better chance of preventing the occurrence of ROP. This study will help to identify whether platelet count and haemoglobin level play a role in the development of ROP.

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CHAPTER 2:

RESEARCH OBJECTIVE

2.1 General Objective

To determine the mean level of haemoglobin level and platelet count in neonates with and without ROP

2.2 Specific Objectives

1. To compare the mean haemoglobin level at birth, week 1, 2, 3, 4, 5 and 6 between neonates with and without ROP.
2. To compare the mean platelet count at birth, week 1, 2, 3, 4, 5 and 6 between neonates with and without ROP.

CHAPTER 3:

MANUSCRIPT

Evaluation of Haemoglobin Level and Platelet Count in Neonates With and Without Retinopathy of Prematurity

Zi Di Lim [1,3], Hans Van Rostenberghe[2,3], Ismail Shatriah [1,3]

1. Department of Ophthalmology and Visual Science, School of Medical Sciences, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia

2. Department of Paediatrics, School of Medical Sciences, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia

3. Hospital Universiti Sains Malaysia, Jalan Raja Perempuan Zainab 2, 16150 Kota Bharu Kelantan, Malaysia.

Correspondence: Ismail Shatriah

Department of Ophthalmology and Visual Science, School of Medical Sciences, Universiti Sains Malaysia, Kubang Kerian 16150, Kelantan, Malaysia

Email shatriah@usm.my

3.1 Abstract

Aim: Haemoglobin and platelet have been postulated to play a role in the development of retinopathy of prematurity (ROP). This study aimed to compare weekly mean haemoglobin level and platelet count between ROP and non-ROP infants in the first six weeks of life.

Method: Ninety-three premature infants with birth weight less than 1.5 kg and gestational age less than 32 weeks were recruited in Hospital Universiti Sains Malaysia from 2017 to 2019. Each ROP case was individually matched (1: 2) to two non-ROP cases. Weekly mean haemoglobin level, weekly mean platelet count, and other related risk factors were documented.

Result: Thirty-one infants with ROP and 62 infants with non-ROP were recruited. Of those with ROP, 8 had stage 1 ROP, 6 had stage 2 ROP, 16 had stage 3 ROP. The mean birth weight and gestational age of the ROP group was 962.2 g and 27.6 weeks while the non-ROP group was 1056.9 g and 28.5 weeks respectively. We found significant differences in the weekly mean platelet counts between ROP and non-ROP infants from week two to week six of life (p=0.003). A significant difference was also found when comparing weekly mean haemoglobin level at week one of life (p=0.003). However, no significant difference was found in the weekly mean platelet count (p=0.489) and weekly mean haemoglobin level (p=0.292) after adjusting to covariates.

Conclusion: There is no significant difference in mean haemoglobin level and platelet count in the first six weeks of life between ROP and non-ROP infants after adjusting to confounding factors like sepsis, bronchopulmonary dysplasia and blood transfusion.

Keywords: retinopathy of prematurity, mean platelet count, mean haemoglobin level, first six weeks of life.

3.2 Introduction

Retinopathy of prematurity (ROP) is a proliferative disorder of the retinal vasculature in premature infants. It is estimated that every year, 32,000 premature infants worldwide developed severe visual impairment due to ROP (1). The pathogenesis of ROP is multifactorial, with low birth weight, low gestational age and supplemental oxygenation being some of the implicated risk factors (2-6).

To the current date, the relationship between haemoglobin level and platelet counts with ROP remains poorly defined, with most studies observing an association between thrombocytopenia (7, 8) or anaemia (9, 10) with the development of ROP, others found no association (11, 12). These differences may be attributed to differences in study methodology and statistical analysis, as the majority of these studies evaluated platelet levels and haemoglobin level as a qualitative variable, i.e. even a single episode of low platelet counts or low haemoglobin level as thrombocytopenia or anaemia respectively. We believe that such an approach may overestimate the level of pathology. This study aims to compare weekly mean platelet count and haemoglobin level in ROP and non-ROP infants from birth till week six of life to determine whether it affects the development of ROP.

3.3 Materials and Methods

Study population

This was a cohort study among preterm infants admitted to Hospital Universiti Sains Malaysia from September 2017 to December 2020. The study was approved by the Human Research Ethics Committee of Universiti Sains Malaysia (USM/JEPeM/18090441). The conduct of the study followed the tenets of the declaration of Helsinki.

Inclusion criteria included all premature infants with gestational age less than 32 weeks and birth weight less than 1.5 kg. Infants with significant congenital abnormalities, ocular defects, and infants who died before their ROP status was known were excluded. Each ROP case was individually matched (1: 2) to two non-ROP cases with a birth weight within 100 grams different and a gestational age within one week of the study subjects. Subjects with extremely small gestational age or birth weight who could not be matched were excluded. Subjects with individual or parental history of platelet or haemoglobin related disease (e.g. idiopathic thrombocytopenic purpura, hemangioma and thalassemia) were excluded.

Data collection

Data collected included birth weight, gestational age, gender, ROP stage, treatment, weekly mean haemoglobin and weekly mean platelet cell count from birth. Associated risk factors for ROP including necrotising enterocolitis, intraventricular haemorrhage, culture-proven sepsis, bronchopulmonary dysplasia, duration of supplemental oxygenation, and total volume of blood transfusion were also documented. Data included were from birth until week six of life.

3.4 Statistical analysis

Statistical analyses were performed using IBM SPSS Statistics for Windows, Version 26.0 (IBM Corp, Armonk, NY, USA). Forward stepwise multiple logistic regression was used to screen for confounding factors (i.e., other factors associated with ROP which may have influenced platelet counts). Repeated measure analysis of variance (ANOVA) was performed to determine the mean differences in weekly haemoglobin level and platelet counts from birth to week six of life between infants with and without ROP. The mean haemoglobin level and platelet count was then adjusted for confounding factors identified during logistic regression using the repeated measure analysis of covariance (ANCOVA). The repeated measure ANCOVA between groups was used to compare intergroup differences in mean haemoglobin level and platelet counts at specific times, and repeated measure ANCOVA within–between groups was used to compare changes in mean haemoglobin level and platelet count across time between groups.

3.5 Results

A total of 93 infants were included in this study. Five infants who died before six weeks of life were excluded. The ROP group consisted of 31 infants and the group without ROP consisted of 62 infants. Of those study subject, 8 (8.6%) had stage 1 ROP, 6 (6.4%) had stage 2 ROP, 16 (17.2%) had stage 3 ROP, 1 (1.1%) had stage 4 ROP and 19 infants (20.4%) required ROP treatment. Among the treatment group, three had zone II stage 2 with plus disease, 10 had zone II stage 3 with plus disease, three had zone I stage 3 without plus disease, two had zone I stage 3 with plus disease and one had stage 4b ROP. All infants received laser treatment. Two infants with zone 1 stage 3 with plus disease received adjuvant intravitreal ranibizumab and one infant

with stage 4b ROP required surgical intervention. The mean birth weight and gestational age of the ROP group was 962.2 gram and 27.6 weeks while the non-ROP group was 1056.9 gram and 28.5 weeks respectively. Other clinical characteristics of our study subjects are shown in Table 1. 51.6% of infants of the ROP group and 6.4% of the non-ROP group had culture-proven sepsis and 38.7% of infants with ROP and 6.4% of the non-ROP group had bronchopulmonary dysplasia. Other co-morbidities of the infants are shown in Figure 1.

There was a significant difference in mean haemoglobin between the ROP and non-ROP group during the first week of life (p=0.003) (Table 2). However, this difference disappeared after adjustment for confounders. The unadjusted and adjusted mean haemoglobin level in each group is illustrated in Figure 2.

We found significant differences in the weekly mean platelet count between infants with and without ROP from week two to week six of life. However, repeated analysis with adjustments for covariates found no significant differences (Table 3). Overall, although there were significant intergroup differences in the change of mean platelet counts over the first six weeks (p=0.002), this significance was lost after adjusting for confounders (p=0.489). The unadjusted and adjusted mean platelet count in each group is illustrated in Figure 3.

In univariate analysis, mean birth weight (OR 0.95, 95% CI 0.91–0.99), mean gestational age (OR 0.997, 95% CI 0.994–0.999), amount of blood transfusion (OR 1.04, 95% CI 1.02–1.05), duration of supplemental oxygenation (OR 1.16, 95% CI 1.05–1.29), culture-proven sepsis (OR 15.46, 95% CI 4.50–53.12) and bronchopulmonary dysplasia (OR 12.42, 95% CI 3.16–48.72)

were statistically significant risk factors for the development of ROP. However, multivariable analysis identified only blood transfusion (OR 1.03, 95% CI 1.010–1.056), bronchopulmonary dysplasia (OR 6.41, 95% CI 1.28–31.93) and culture-proven sepsis (OR 8.79, 95% CI 2.09–36.93) as independent risk factors for ROP. Table 4 summarizes these results.

3.6 Discussion

Advances in neonatal care have contributed to improved survival of preterm infants, particularly in technologically developed regions (13, 14). Paradoxically, the incidence of ROP in such areas is decreasing, while the converse is true of lower-income countries (15, 16). ROP demographics and risk factors appear to differ between regions, with severe ROP occurring even in more mature infants in low and middle-income countries (17, 18). The importance of identifying risk factors could enhance our screening programs thus cannot be overemphasised. To our best knowledge, this is the first study to document that adjusted weekly mean haemoglobin level and platelet counts over the first six weeks of life have no direct relationship with ROP.

Infants with ROP had lower mean haemoglobin level than matched infants without ROP. This difference was statistically significant at week one of life. These findings are consistent with the results of Lungren et al, who found that infants with greater duration of anaemia during the first week of life were at higher risk of ROP (9, 10). Similarly, Banerjee et al observed that low haemoglobin at birth is associated with ROP development (19). However, haemoglobin is affected by a variety of factors, and failure to account for these may have accounted for the conclusions in the aforementioned studies. After adjustment for confounders we found no

significant difference in haemoglobin between the two groups. This suggests that lower haemoglobin level is not directly related to the development of ROP. Rather, it may reflect the physiological processes taking place in these premature infants, such as inhibition of erythropoietin production by sepsis. This finding is in line with Englert et al, that found anaemia did not affect ROP as an independent risk factor (20).

Similarly, we observed that infants with ROP had significantly lower weekly unadjusted mean platelet counts from week two to week six compared to their counterparts without ROP. This finding is consistent with the literature, in which significant association between thrombocytopenia and ROP was observed when the former was treated as a categorical variable (i.e. thrombocytopenia was present in any subject with a single episode of platelet count less than 150,000/microL) (7-10, 21-23). However, platelet counts may be affected by a variety of factors, including sepsis (24), blood transfusion (25) and bronchopulmonary dysplasia (26). Our finding of a lack of significance between the mean platelet counts in both groups after adjustment for confounders demonstrates that platelet counts are not independently related to the development of ROP. This is in keeping with the results of Korkmaz et al., who found no difference in mean platelet counts between ROP and control groups during the hypothetical first and second phases of ROP (12). Since the transition of phase 1 to phase 2 has a wide range of onset (27), it is difficult to pinpoint a single time point at which platelet counts influence ROP development. Thus, we believe that an approach utilizing weekly mean platelet counts from birth is more reflective of the overall clinical picture, and that the observed differences in platelet counts between infants with and without ROP are the result of other co-morbidities in these patients.

Platelets have been implicated in the pathogenesis of ROP, as they contain both pro-angiogenic cytokines like VEGF (Vascular-Endothelial Growth Factor), PDGF (Platelet-Derived Growth Factor) and BDNF (Brain-Derived Neurotrophic Factor) (28). A recent study concluded that there is a strong correlation between postnatal platelet count and systemic VEGF-A, PDGF-BB and BDNF in ROP infants in which low platelet during the second phase of ROP was associated with a reduced level of these angiogenic factors (29). Although there should be an increased retinal VEGF level during phase 2 of ROP, the author found decreased systemic VEGF during this same period. Further research is needed to address whether systemic VEGF level affects retinal VEGF level (30).

Sepsis, presence of bronchopulmonary dysplasia and volume of blood transfusion are well-known risk factors for ROP (26, 31, 32). However, the mechanisms by which they affect platelet and haemoglobin physiology in ROP have not been thoroughly discussed. We hypothesise that reduction in platelet counts and haemoglobin level indirectly reflects the ongoing septic process which compounds the underlying susceptibility of these infants to ROP. Sepsis not only causes endothelial damage with resultant increased platelet activation, initiating a vicious cycle of subsequent platelet-mediated cytotoxic endothelium damage (33), but also causes bone marrow suppression, with consequent decreased platelet production (34). In bronchopulmonary dysplasia, anomalous lung development with ventilation-perfusion abnormalities causes hypoxemia (35) which condition could be aggravated by anaemia. Platelet deficiency may subsequently compound the disease severity, as platelet-derived-growth factors (PDGF) are key components of normal alveolarization (36). The need for blood transfusions due to anaemia may trigger platelet activation and aggregation (37), causing micro-occlusion of retinal vessels and resulting in platelet consumption from the bloodstream. However, ROP severity has not