Thrombocytopenia, bleeding, and use of platelet transfusions in sick neonates

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Survival rates for infants born prematurely have improved significantly, in part due to better supportive care such as RBC transfusion. The role of platelet transfusions in neonates is more controversial. Neonatal thrombocytopenia is common in premature infants. The primary causal factors are intrauterine growth restriction/maternal hypertension, in which the infant presents with thrombocytopenia soon after birth, and sepsis/necrotizing enterocolitis, which are the common morbidities associated with thrombocytopenia in neonates > 72 hours of age. There is no evidence of a relationship between platelet count and occurrence of major hemorrhage, and cardiorespiratory problems are considered the main etiological factors in the development of intraventricular and periventricular hemorrhage in the neonatal period. Platelet transfusions are used commonly as prophylaxis in premature neonates with thrombocytopenia. However, there is widespread variation in the pretransfusion thresholds for platelet count and evidence of marked disparities in platelet transfusion practice between hospitals and countries. Platelet transfusions are biological agents and as such are associated with risks. Unlike other patient groups, specifically patients with hematological malignancies, there have been no recent clinical trials undertaken comparing different thresholds for platelet transfusion in premature neonates. Therefore, there is no evidence base with which to inform safe and effective practice for prophylactic platelet transfusions. There is a need for randomized controlled trials to define the optimal use of platelet transfusions in premature neonates, who at present are transfused heavily with platelets.

Background

The routine practice of supportive use of RBC transfusions has undoubtedly been one of the factors responsible for improving outcomes in very premature neonates. In contrast, the optimal role of platelet transfusions, the second most commonly transfused blood component in neonates, remains controversial. Neonatal thrombocytopenia and all types of bleeding are frequent and are important clinical problems for very preterm neonates, with approximately 25% of neonates admitted to neonatal intensive care units developing thrombocytopenia. However, the close temporal association between low platelet counts and the occurrence of clinical bleeding that is often seen in sick babies does not in itself establish cause and effect, nor does it provide justification for the assumption that there are benefits for giving platelet transfusions to attempt correction of low platelet counts. Thrombocytopenia is a risk factor for poorer neonatal outcomes, but it is unclear whether it is a marker of severity of illness and comorbidity or an indicator for the use of platelet transfusions. As discussed herein, our lack of understanding of the nature of these relationships is a main reason why the role of platelet transfusions in neonates remains poorly defined.[1,2]

Definitions of thrombocytopenia

Peripheral blood platelet counts can be measured as early as at 18 weeks of gestation. Fetal platelet counts are often more than 150×10^9/L and therefore similar to the results found in mature neonates. Neonatal thrombocytopenia is generally defined as a platelet count below 100×10^9/L, and is defined as severe when it is below 50×10^9/L. However, these definitions are pragmatic thresholds, largely drawn by analogy with wider hematology practice. The use of specific counts in the peripheral blood to define different thresholds of severity may convey into the mind of an attending physician that there are stepwise changes in bleeding risk or outcomes at different platelet count levels, which is not known. Defining thresholds of clinically significant thrombocytopenia by levels of platelet count in the peripheral blood does not take into account changes in platelet function,[3,4] which differ between neonates and older children and adults, nor does it consider the capacity of BM to increase platelet production or produce more metabolically active platelets.

Prevalence and severity of thrombocytopenia

Estimates of the prevalence of neonatal thrombocytopenia vary at between 1% and 5% of all neonates.[1,5-8] Thrombocytopenia affects approximately 25% (range, 22%-35%) of all neonates admitted to neonatal intensive care units, and the risk of developing thrombocytopenia is inversely proportional to gestational age (GA).

Main causes of thrombocytopenia

Hematologists are familiar with the more predictable profiles of thrombocytopenia in the setting of children with hematological malignancies receiving myeloablative chemotherapy. In contrast, both the main etiologies and patterns of thrombocytopenia in neonates are highly variable and may be more difficult to diagnose and predict at their onset.[8] In some cases, the likely cause of thrombocytopenia can be postulated from events surrounding birth and the postnatal age.

In a premature neonate, including those small for gestational age (typically defined as less than 10th percentile for weight), thrombocytopenia in the first 72 hours after birth is often considered to be due to antenatal and maternal factors (eg, placental "insufficiency," perinatal hypoxia, and antenatal/perinatal infection), with reduced

megakaryopoiesis.[8] After 72 hours from birth, severe thrombocytopenia in premature neonates is most likely due to postnatally acquired infection, often bacterial, or necrotizing enterocolitis (NEC). The pathophysiology involves both the suppression of platelet production due to cytokine release (with evidence of decreased numbers of clonogenic megakaryocyte progenitors and low levels of thrombopoietin), and increased consumption, sometimes as part of disseminated intravascular coagulation (with evidence of shortened platelet survival and increased platelet-associated IgG). Profound thrombocytopenia can develop quickly when it develops late; for example, the pattern and duration of thrombocytopenia in neonates with NEC can be severe, of rapid onset, and then protracted with multiple episodes. The sick preterm neonate may also have comorbidities such as liver disease, which will also affect hemostasis (ie, platelet dysfunction and coagulopathies), in addition to inflammatory changes in sepsis that affect endothelial dysfunction.[9]

Overall, by far the most common causal factors for neonatal thrombocytopenia are intrauterine growth restriction/maternal hypertension, sepsis, and NEC. Algorithms to assist in the diagnosis of the different causes of thrombocytopenia have been published, and may be helpful in directing specific investigations (eg, neonatal alloimmune thrombocytopenia [NAIT]).[10]

Immune thrombocytopenia

Early thrombocytopenia can be caused by immune destruction of platelets by maternally derived Abs. Ab-mediated platelet dysfunction may also contribute to the bleeding tendency. When an otherwise healthy term neonate presents with severe thrombocytopenia within 48 hours of birth, NAIT should be suspected. A typical presentation is petechiae/bruising or bleeding alongside severe thrombocytopenia, often well less than 50×10^9/L.[8,11] NAIT, in many proven cases, results from maternally derived anti-human platelet antigen 1a (anti-HPA1a) or anti-HPA5b platelet Abs. Serological testing of mother and neonate through mAb-specific immobilization of platelet antigen testing can detect and confirm maternal HPA Abs, and parents and infants can be genotyped for all common HPA antigens. However, results from these tests may not be available immediately, and severely thrombocytopenic neonates ($< 30 \times 10^9$/L) should receive platelet transfusions, ideally with platelets previously typed negative for the common HPA1a/5b antigens, if available. IV Igs may reduce the need for platelet transfusions until spontaneous recovery in platelet count occurs 1-6 weeks after birth. All infants with NAIT (or suspected NAIT) and thrombocytopenia after birth (because bleeding may occur at higher thresholds) should be monitored for intracranial hemorrhage by cranial ultrasound. There is a very strong association with HLA: the ability of an HPA-1a$^-$ woman to form anti-HPA1a is strongly determined by the HLA DRB3*0101 allele, such that HLA DRB3*0101$^+$ women are 140 times more likely to form anti-HPA1a than HLA DRB3*0101$^-$ women.[12] It should also not be forgotten that pregnant women and mothers with HPA Abs are at risk of posttransfusion purpura, and transfusion with HPA-compatible RBCs and platelets, if available, should be considered, unless in an emergency.

Maternal (auto-) immune thrombocytopenia (maternal ITP) is another cause of neonatal thrombocytopenia, which may be suspected in an otherwise healthy (typically term) neonate. The often nonspecific platelet Ab transfers across the placenta to the fetal circulation. In the majority of cases, there is a known history of maternal ITP or lupus. If thrombocytopenia is unexpected in a neonate, the maternal platelet count should be checked.[13] Normally, the neonatal platelet count will increase within a week as the Ab levels decrease, but in some cases, the count remains low for up to 8 weeks. Sometimes prolonged thrombocytopenia develops, which may be difficult to separate from other causes of thrombocytopenia such as sepsis. Siblings of neonates with a history of prolonged thrombocytopenia due to maternal ITP usually exhibit the same pattern. Intracranial hemorrhage is less commonly seen compared with infants with NAIT. The treatment of choice for severe thrombocytopenia ($< 20 \times 10^9$/L) or bleeding is IV Igs, although data on their effectiveness are very limited.[14] Platelet transfusions should only be considered for life-threatening bleeding.

Consequences of thrombocytopenia

The clinical consequences of thrombocytopenia can be followed both clinically and with laboratory monitoring, by assessing risks of bleeding and by specialist tests of hemostatic function.[15] The main clinical concern for neonates with severe thrombocytopenia is major bleeding, including intracranial hemorrhage, and specifically intraventricular and periventricular hemorrhage (IVH-PVH).[16] The incidence of major IVH-PVH may exceed 30% in the most premature neonates, and approximately 75% of cases of IVH-PVH develop in the first 48 hours after birth. Because the premature neonatal brain is relatively fragile with a poorly developed subependymal matrix and weak endothelial supporting structures, it is not surprising that rupture of small blood vessels for any reason is a significant risk. Relevant factors in the etiology of IVH-PVH include cardiovascular and respiratory instability and changes in vascular perfusion pressures, both perinatally and postnatally. These causes have been raised when considering possible associations between RBC transfusion and occurrence of IVH.[17]

One of the first studies to assess the impact of thrombocytopenia prospectively in neonates was reported by Andrew et al.[15] Nearly 100 neonates with platelet counts $< 100 \times 10^9$/L were compared with an age-, weight-, and disease-matched control group. The scores for overall hemorrhage, based on measures of skin and organ bleeding, were greater in thrombocytopenic neonates compared with sick infants with normal platelet counts. Another study looking at a modified bleeding time indicated an inverse relationship with platelet count, but the strength of the relationship was weak.[4]

In a more recent prospective study, across 7 neonatal units, 169 neonates with platelet counts $< 60 \times 10^9$/L were enrolled.[18] A bleeding assessment tool was used to attempt standardized recording of different types of predefined minor, moderate, and major bleeding across neonatal units. Major hemorrhage occurred in 13% of severely thrombocytopenic neonates (IVH, 53%; pulmonary, 26%; renal, 11%). The majority (84%) of neonates who had a major hemorrhage were born at < 30 weeks of gestation. Neonates with thrombocytopenia secondary to intrauterine growth restriction or pregnancy-induced hypertension tended not to have major hemorrhage, despite platelet counts $< 60 \times 10^9$/L in some cases. There was no evidence of a relationship between platelet count and occurrence of major hemorrhage,[18] and 91% of neonates with platelet counts $< 20 \times 10^9$/L did not develop major hemorrhage. In that study, the common forms of clinically evident minor and moderate bleeding included bleeding from the endotracheal tube, oozing from the skin at puncture sites, and bleeding through the nasogastric secretions. In a follow-up analysis of all bleeding outcomes, gestational age < 34 weeks, early postnatal age for development of severe thrombocytopenia (within 10 days of birth), and NEC were found to be the strongest predictors for an increased

number of bleeding events; in contrast, a lower platelet count in itself was not a strong predictor of increased bleeding risk.[19]

Other studies have addressed the pattern and burden of bleeding in neonates with thrombocytopenia, including a retrospective study of patterns of minor types of bleeding that reported a causal relationship between thrombocytopenia and cutaneous bleeding (defined as heavy bruising or oozing from previous venipuncture sites).[20,21] Associations between bleeding and GA and postnatal age may relate to the laboratory studies by Del Vecchio et al,[22] who reported that, irrespective of the platelet count, the bleeding time was prolonged 2-fold on day 1 for preterm babies of < 33 weeks of gestation compared with those > 38 weeks, but by day 10, the difference in the bleeding time for any GA was insignificant.

Platelet transfusions

The only readily available specific treatment for thrombocytopenia in neonates remains platelet transfusions; treatments with thrombopoietic growth factors are investigational and research studies have not been undertaken in neonates and therefore will not be considered further in this review.[23] Two types of platelet products are available: pooled platelets, which are derived from whole-blood donations from donors, and apheresis platelets, which are collected by cell-separation techniques from donors. Splits of units of platelet concentrates may be prepared for neonatal use. After production, platelet concentrates are stored in plasma in incubators at 20°-24°C for 5 days or more (depending on use of bacterial screening methods); platelets must be agitated during storage before transfusion. During storage, platelets undergo several changes affecting platelet function (eg, aggregation and release) and platelet structure (eg, microvesiculation). However, the clinical consequences of this "platelet storage lesion" in neonates are unclear. In several countries, additional (microbiological) safety measures apply for blood components collected for transfusions to neonates under several conditions[24] such as repeat donors (1 donation in the last 2 years) or CMV-seronegative donors.[25] Despite improvements in donor microbiological screening and testing, a small risk of viral, bacterial, or protozoal contamination of platelets remains. There is interest in applying new pathogen-reduction technologies to platelet concentrates to inactivate and further minimize the risks of transfusion transmission of known and future pathogens.

Evidence of efficacy

Few would argue against the presumed beneficial effect that (therapeutic) platelet transfusions have in treating neonates with severe thrombocytopenia who are actively bleeding, such as with massive pulmonary hemorrhage. However, many neonatologists consider that the additional, if not primary, goal for platelet transfusions is to prevent severe and life-threatening bleeding in neonates with severe thrombocytopenia (prophylaxis). This leads to 2 main evidence-based questions surrounding the use of prophylactic platelet transfusions: (1) are prophylactic platelet transfusions superior to a strategy of therapeutic-only platelet transfusions for the prevention of severe thrombocytopenic bleeding, and (2) if prophylactic platelet transfusions are recommended, what is the optimal platelet dose and platelet count threshold for neonates with thrombocytopenia and/or before invasive procedures/surgery?

The effectiveness of platelet transfusions in clinical trials has been assessed traditionally by the measurement of a surrogate marker, the platelet count increment (at 1 hour and/or 24 hours after transfusion). However, the assessment of bleeding is really the more clinically relevant measure, in addition to mortality and neurodevelopmental outcome. The way that bleeding has been recorded and assessed in many platelet trials has varied markedly.[26] Application of the lessons from the design of platelet transfusion trials in other areas, such as for adult patients with hemophilia or hematological malignancies, to neonatal practice is beginning, including the development of standardized assessment tools to record bleeding.[27]

There has been only one randomized controlled trial in neonates to assess a threshold level for the effectiveness of prophylactic platelet transfusions, which reported that moderate thrombocytopenia (defined as 50-150 $\times 10^9$/L) was not detrimental to short-term neonatal outcome, specifically to prevent risk of progression of IVH-PVH.[28] However, neonates with severe thrombocytopenia ($< 50 \times 10^9$/L) were excluded from this study because of their perceived high risk of hemorrhage.

When considering the selection of different thresholds for prophylactic platelet transfusion, it is interesting that no studies in any patient group have clearly identified "stepwise" changes in rates of bleeding at different platelet count levels, which might provide baseline information to guide the selection of platelet counts for prophylactic platelet transfusion. In a large randomized controlled trial of different platelet doses in adult and children with hematological malignancies, bleeding rates were broadly similar across all platelet counts and occurred on 25% of study days when the platelet count was 5×10^9/L or lower, compared with 17% of study days when the platelet count was between 6×10^9/L and 80×10^9/L.[29] No comparable platelet dose trials exist to inform neonatal practice.

Use of platelet transfusions

Despite the limited evidence, platelet transfusions are used frequently in modern neonatal clinical practice in thrombocytopenic neonates, often as prophylaxis.[30-33] Early studies estimated that 25% of neonates whose platelet counts decreased to below 150×10^9/L received one or more transfusions, increasing to 50% in extremely low birth weight (< 1000 g) neonates. In a prospective study,[18] 69% of all neonates with a platelet count less than 60×10^9/L were given a platelet transfusion. Most transfusions were given at platelet counts between 10 and 50×10^9/L. The 50th and 90th percentile pretransfusion platelet count thresholds observed were 27×10^9/L and 48×10^9/L, respectively. In that study, 173 of 415 (42%) transfusions were administered at a minimum platelet count of $< 25 \times 10^9$/L and 383 of 415 (92%) transfusions were administered at a minimum platelet count of $< 50 \times 10^9$/L. Other hospital studies describing neonatal platelet transfusion practice have found variable rates of transfusion, some as high as 9% of all admissions. Murray et al[32] demonstrated no increased rates of hemorrhage irrespective of whether platelets were administered to neonates with severe thrombocytopenia.

In general, policies and protocols for neonatal platelet transfusion therapy vary widely between clinicians and institutions, reflecting the generally broad nature of recommendations in local and national guidelines, which themselves are based on consensus rather than evidence.[33] Several recent surveys have highlighted the marked disparities in practice, both for routine prophylaxis and before invasive procedures. A large web-based survey of neonatologists in Canada and the United States reported variations and that platelet transfusions were administered frequently to nonbleeding neonates with platelet counts $> 50 \times 10^9$/L.[34] These practices appear more liberal than for countries in Europe.[35] In the United Kingdom, a telephone survey of all tertiary level neonatal units has shown

variations in platelet transfusion practice, and the most common thresholds for transfusion in healthy or stable term and preterm infants were 25×10^9/L and 30×10^9/L, respectively.[36]

Risks of platelet transfusions

Any discussion of use of platelets needs to acknowledge risks. Platelet transfusions are biological agents and are a costly and scarce resource.[37-39] Some potential risks related to platelet transfusion are well described, including errors in administration, cases of incorrect blood component transfused, or special requirements not met (eg, irradiated components). Data over nearly 10 years from the United Kingdom's Serious Hazards of Transfusion (SHOT) national hemovigilance scheme against a population-based epidemiological study of transfused patients have indicated that a disproportionate number of overall adverse events occurred in neonatal compared with pediatric and adult transfusion practice. In that study, there were no cases of transfusion-transmitted infection in neonates (and there was 1 case of transfusion-associated GVHD in a 13-day-old infant who received nonirradiated RBCs).[40]

It is highly likely that there is under-recognition of adverse events in infants already sick from other causes and who are characterized by immunological immaturity. Platelets are the blood component most likely to be contaminated by bacteria because they are stored at room temperature. Concerns of possible transfusion-transmitted variant Creutzfeldt-Jakob disease is particularly relevant for neonatal transfusion practice in some countries, given the long life expectancy of many of these recipients, and this illustrates the importance of considering transfusion risks to neonates from future pathogens. Other studies have suggested an association between repeated platelet transfusions and adverse events such as hepatic dysfunction after NEC.[41,42] Finally, side effects related to transfusions do not just reflect the adverse events or reactions related to the blood components themselves, but also the problems of maintaining vascular access in infants with risks of extravasated/infected IV cannulae.

Future research

Despite the recognized high incidence and prevalence of thrombocytopenia in neonates, there is still a dearth of prospective information on the types and incidence of bleeding and the clinical consequences of bleeding in this vulnerable group of patients. Recent randomized trials addressing RBC transfusion triggers for anemic neonates have contributed to the debate about appropriate and safe thresholds for neonatal RBC transfusions. Comparable platelet trigger trials assessing clinically relevant outcomes have not been undertaken in preterm neonates with thrombocytopenia,[43] although one is now under way (http://www.planet-2.com). Establishing more evidence-based thresholds for prophylactic platelet transfusion may be complicated by the clinical diversity in this patient group (eg, gestational age, postnatal age, presence of coagulopathy, and mechanism of thrombocytopenia, including consumption vs production), and therefore will require clinical trials with larger sample sizes. Without such trials, recommendations and guidelines for neonatal platelet transfusion practice will continue to be based largely on consensus and will inevitably show differences between hospitals and countries. All hospitals should develop locally agreed upon guidelines for platelet transfusions, supported by strategies for implementation, while awaiting results from such clinical trials.[44,45]

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References

1. Sallmon H, Sola-Visner M. Clinical and research issues in neonatal anemia and thrombocytopenia. *Curr Opin Pediatr*. 2012;24(1):16-22.
2. Strauss RG. Platelet transfusions in neonates: questions and answers. *Expert Rev Hematol*. 2010;3(1):7-9.
3. Bednarek FJ, Bean S, Barnard MR, et al. The platelet hyporeactivity of extremely low birth weight neonates is age dependent. *Thrombosis Res*. 2009;124:42-45.
4. Del Vecchio A. Use of the bleeding time in the neonatal intensive care unit. *Acta Paediatr Suppl*. 2002;91(438):82-86.
5. Burrows RF, Kelton JG. Thrombocytopenia at delivery: A prospective survey of 6715 deliveries. *Am J Obstet Gynecol*. 1990;162:731-734.
6. Sainio S, Jarvenpaa AS, Renlund M, et al. Thrombocytopenia in term infants: A population-based study. *Obstet Gynecol*. 2000;95:441-446.
7. Christensen RD, Henry E, Wiedmeier SE, et al. Thrombocytopenia among extremely low birth weight neonates: data from a multihospital healthcare system. *J Perinatol*. 2006;26:348-353.
8. Roberts I, Stanworth S, Murray N A. Thrombocytopenia in the Neonate. *Blood Rev*. 2008;22(4):173-186.
9. Goerge T, Ho-Tin-Noe B, Carbo C, et al. Inflammation induces hemorrhage in thrombocytopenia. *Blood.* 2008;111:4958-4964.
10. Chakravorty S and Roberts I. How I manage neonatal thrombocytopenia. *Br J Haematol*. 2012;156:155-162.
11. Egbor M, Knott P, Bhide A. Red-cell and platelet allommunisation in pregnancy. *Best Pract Res Clin Obstet Gynaecol*. 2012;26:119-132.
12. Williamson LM, Hackett G, Rennie J, et al. The natural history of fetomaternal allimmunization to the platelet-specific antigen HPA-1a as determined by antenatal screening. *Blood.* 1998;92: 2280-2277.
13. Burrows RF, Kelton JG. Incidentally detected thrombocytopenia in healthy mothers and their infants. *N Engl J Med*. 1988;319:142-145.
14. Ballin A, Andrew M, Ling E, Perlaman M, Blanchette V. High dose intravenous gammaglobulin therapy for neonatal idiopathic autoimuune thrombocytopenia. *J Pediatr*. 1988;112:789-792
15. Andrew M, Castle V, Saigal S, et al. Clinical impact of neonatal thrombocytopenia. *J Pediatr*. 1987;110:457-464.
16. de Vries LS. Hemorrhagic lesions of the central nervous system. In: Stevenson DK, Benitz WE, Sunshine P, et al, eds. *Fetal and Neonatal Brain Injury*. Cambridge, United Kingdom: Cambridge University Press; 2012:285-295.
17. Baer VL, Lambert DK, Henry E, Snow GL, Christensen RD. Red blood cell transfusion of preterm neonates with a Grade 1 intraventricular hemorrhage is associated with extension to a Grade 3 or 4 hemorrhage. *Transfusion.* 2011;51(9):1933-1939.
18. Stanworth SJ, Clarke P, Watts T, et al. Prospective, observational study of outcomes in neonates with severe thrombocytopenia. *Pediatrics.* 2009;124:e826-e834.

19. Muthukumar P, Venkatesh V, Curley A, et al. Severe thrombocytopenia and patterns of bleeding in neonates: Results from a prospective observational study and implications for use of platelet transfusions [published online ahead of print June 27, 2012]. *Transfus Med*. doi:10.1111/j.1365-3148.2012.01171.x.
20. von Lindern JS, van den Bruele T, Lopriore E, Walther FJ. Thrombocytopenia in neonates and the risk of intraventricular hemorrhage: a retrospective cohort study. *BMC Pediatr*. 2011; 11:16.
21. Baer VL, Lamber DK, Christensen RD. Severe thrombocytopenia in the NICU. *Pediatrics.* 2009;124:e1095-e1100.
22. Del Vecchio A, Latini G, Henry E, Christensen RD. Template bleeding times of 240 neonates born at 24 to 41 weeks gestation. *J Perinatol*. 2008;28:427-431.
23. Sallmon H, Gutti RK, Ferrer-Marin F, Liu ZJ, Sola-Visner MC. Increasing platelets without transfusion: is it time to introduce novel thrombopoietic agents in neonatal care? *J Perinatol*. 2010;30(12):765-769.
24. New HV, Stanworth SJ, Engelfriet CP, et al. Neonatal transfusions. *Vox Sanguinis*. 2009;96:62-85.
25. Josephson CD, Castillejo MI, Caliendo AM, et al. Prevention of transfusion-transmitted cytomegalovirus in low-birth weight infants (≤1500 g) using cytomegalovirus-seronegative and leukoreduced transfusions. *Transfus Med Rev*. 2011;25:125-132.
26. Heddle NM, Cook RJ, Webert KE, Sigouin C, Rebulla P. Biomedical Excellence for Safer Transfusion Working Party of the International Society for Blood Transfusion. Methodologic issues in the use of bleeding as an outcome in transfusion medicine studies. *Transfusion.* 2003;43:742-752.
27. Tosetto A, Castman G, Rodeghiero F. Bleeding scores in inherited bleeding disorders: clinical or research tools? *Haemophilia.* 2008;14:415-422.
28. Andrew M, Vegh P, Caco C, et al. A randomized controlled trial of platelet transfusions in thrombocytopenic premature infants. *J Pediatr*. 1993;123:285-291.
29. Slichter SJ, Kaufman RM, Assmann SF, et al. Dose of prophylactic platelet transfusions and prevention of haemorrhage. *N Engl J Med*. 2010;362:600-613.
30. Dohner ML, Wiedmeier SE, Stoddard RA, et al. Very high users of platelet transfusions in the neonatal intensive care unit. *Transfusion.* 2009;49:869-872.
31. Del Vecchio A, Sola MC, Theriaque DW, et.al. Platelet transfusions in the neonatal intensive care unit: factors predicting which neonates will require multiple transfusions. *Transfusion.* 2001;41:803-808.
32. Murray NA, Howarth LJ, McCloy MP, Letsky EA, Roberts IA. Platelet transfusion in the management of severe thrombocytopenia in neonatal intensive care unit neonates. *Transfus Med*. 2002;12(1):35-41.
33. Saxonhouse M, Slayton W, Sola MC. Platelet transfusions in the infant and child. In: Hillyer CD, Luban NLC, Strauss RG, eds. *Pediatric Handbook of Transfusion Medicine*. San Diego, CA: Elsevier; 2004:253-269.
34. Josephson CD, Su LL, Christensen RD, Hillyer CD, et al. Platelet transfusion practices among neonatologists in the United States and Canada: results of a survey. *Pediatrics.* 2009;123:278-285.
35. Cremer M, Sola-Visner M, Roll S, et al. Platelet transfusions in neonates: practices in the United States vary significantly from those in Austria, Germany, and Switzerland. *Transfusion.* 2011;51(12):2634-2641.
36. Chaudhary R, Clarke P. Current transfusion practices for platelets and fresh, frozen plasma in UK tertiary level neonatal units. *Acta Paediatr.* 2008;97:135.
37. Kleinman S, Chan P and Robillard P. Risks associated with transfusion of cellular blood components in Canada. *Transfus Med Rev*. 2003;17:120-162.
38. Baer VL, Lambert DK, Henry E, Snow GL, Sola-Visner MC, Christensen RD. Do platelet transfusions in the NICU adversely affect survival? Analysis of 1600 thrombocytopenic neonates in a multihospital healthcare system. *J Perinatol*. 2007;27(12):790-796.
39. Slonim AD, Joseph JG, Turenne WM, Sharangpani A and Luban NLC. Blood transfusions in children: a multi-institutional analysis of practices and complications. *Transfusion.* 2008;48:73-80
40. Stainsby D, Jones H, Wells AW, Gibson B, Cohen H. SHOT Steering Group. Adverse outcomes of blood transfusion in children: analysis of UK reports to the serious hazards of transfusion scheme 1996-2005. *Br J Haematol*. 2008;141:73-79.
41. Kenton AB, Hegemier S, Smith EO, et al. Platelet transfusions in infants with necrotizing enterocolitis do not lower mortality but may increase morbidity. *J Perinatol*. 2005;25:173-177.
42. Fernandes CJ, O'Donovan DJ. Platelet transfusions in infants with necrotizing enterocolitis. *Curr Hematol Rep*. 2006;5:76-81.
43. Blajchman MA, Glynn SA, Josephson CD, Kleinman SH. State-of-the Science Symposium Transfusion Medicine Committee. Clinical trial opportunities in Transfusion Medicine: proceedings of a National Heart, Lung, and Blood Institute State-of-the-Science Symposium. *Transfus Med Rev*. 2010; 24(4):259-285.
44. Motta M, Testa M, Tripodi G, Radicioni M. Changes in neonatal transfusion practice after dissemination of neonatal recommendations. *Pediatrics.* 2010;125(4):e810-e817.
45. Baer VL, Henry E, Lambert DK, et al. Implementing a program to improve compliance with neonatal intensive care unit transfusion guidelines was accompanied by a reduction in transfusion rate: a pre-post analysis within a multihospital health care system. *Transfusion.* 2011;51(2):264-269.