International Journal of Pediatrics and Adolescent Medicine (2015) **2**, 99–106

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REVIEW ARTICLE

Prevention of the anaemia of prematurity

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Received 1 October 2015; received in revised form 26 October 2015; accepted 29 October 2015
Available online 24 November 2015

KEYWORDS
Anaemia of prematurity;
Delayed cord clamping;
Umbilical cord milking;
Placental transfusion

Abstract Despite recent advances in neonatal and perinatal medicine, extremely low birth weight infants (ELBW) are at high risk of developing anaemia of prematurity (AOP) requiring packed red blood cell (RBC) transfusions. The benefit of transfusing allogenic RBCs for AOP is a controversial issue, except for disturbances in tissue oxygenation. Although the role of erythropoietin (EPO) in the pathophysiology of AOP is well known, neither early nor late recombinant human EPO therapy alters the number or volume of RBC transfusions. It is also known that one-half of the feto-placental blood volume remains outside the newborn infant's circulation at 30 weeks of gestation if the umbilical cord is clamped immediately. Delayed cord clamping (DCC) and umbilical cord milking (UCM) are the main methods for enhancing placental transfusion. The basic principle of these approaches depends on providing high haemoglobin (Hb) levels to premature infants in the delivery room. The enhancement of placental transfusion clearly results in higher Hb levels at birth, reducing the need for RBC transfusions as well as creating a better haemodynamic status during the initial hours of life. To date, enhancement of placental transfusion in the delivery room by either DCC or UCM seems to be the best preventive measure for AOP. Yet, studies on the associated neurodevelopmental outcomes are insufficient to reach a conclusion. This review summarizes the pathophysiology, treatment and preventative strategies of anaemia of prematurity in light of the current literature.

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Peer review under responsibility of King Faisal Specialist Hospital & Research Centre (General Organization), Saudi Arabia.

http://dx.doi.org/10.1016/j.ijpam.2015.10.001

1. Introduction

Recent advances in perinatal and neonatal medicine and the broader use of antenatal steroids, exogenous surfactants, sophisticated incubators and ventilator support modalities have resulted in a remarkable increase in the survival of preterm infants. Despite these advances, extremely low birth weight (ELBW) preterm infants remain at significant risk for the most frequent life-threatening complications of prematurity, such as intraventricular haemorrhage (IVH), necrotizing enterocolitis (NEC), retinopathy of prematurity (ROP) and anaemia of prematurity (AOP) [1]. Preterm infants, particularly those with a birth weight less than 1500 g, are at high risk of requiring at least one, and often, multiple red blood cell (RBC) transfusions [2]. The mean number of transfusions in the two largest studies ranged from 3.3 to 5.7, depending on whether restrictive or liberal transfusion guidelines were used [3,4]. Nearly 50% of ELBW infants (or infants born at or before 29 weeks of gestation) receive their first transfusion during the first 2 weeks of age, and 80% receive at least one additional blood transfusion by the end of their hospitalization [5].

Although RBC transfusions are a critical part of neonatal intensive care unit (NICU) stays and can be life saving for premature infants with severe anaemia or haemorrhage, they also convey risks and are costly and not easy to utilize, especially in resource-limited settings [6]. Therefore, preventive strategies to start life with higher haemoglobin levels have become popular during the last few decades [7]. This article reviews preventive strategies for AOP, giving particular emphasis to the enhancement of placental transfusion in the delivery room.

2. Anaemia of prematurity

Anaemia of prematurity is a pathological condition unlike physiologic anaemia in newborns [8]. The pronounced decline in the haemoglobin (Hb) concentration that occurs in ELBW infants is usually associated with abnormal clinical signs and requires allogeneic RBC transfusions [8,9]. AOP is characterized by reduced endogenous erythropoietin (EPO), reduced RBC lifespan and hypo-regenerative bone marrow [7]. Non-physiologic factors related to prematurity, such as phlebotomy losses for laboratory evaluations and nosocomial infections resulting in oxidative haemolysis, also contribute to high transfusion preterm infants [10].

When tissue hypoxia occurs, the transcription and expression of EPO mRNA increases, followed by an increase in erythropoiesis. Hypoxia-induced EPO expression is controlled by an enhancer element called hypoxia inducible factor-1 (HIF-1). Induction of HIF-1 binding in hypoxic cells requires RNA and protein synthesis, as well as protein phosphorylation. Sustained hypoxia is required for increasing EPO production. EPO production occurs primarily in the liver before 30 weeks of gestation (and is produced primarily in the kidney thereafter), and hypoxia is a less effective isolated stimulus for EPO production and erythropoiesis. The switch in hypoxia responsiveness and in the site of EPO production may contribute to AOP [2]. Suboptimal erythropoiesis appears to be the result of the inadequate synthesis of EPO in response to hypoxia [11]. EPO deficiency is greater in smaller premature infants compared to less mature infants [12]. Iron, folate, vitamin B12, or vitamin E deficiencies can also contribute to inadequate erythropoiesis [11].

Traditionally, AOP has been treated with frequent packed RBC transfusions [13]. Among all age groups, the need for allogenic packed RBCs is common in newborns. Furthermore, preterm infants are among the most heavily transfused patient populations [3,4]. The goal of packed RBC transfusions in infants with AOP is to restore or maintain oxygen delivery without increasing oxygen consumption [11]. However, according to the UK Serious Hazards of Transfusion (SHOT) National Haemovigilance Scheme, an increased number of adverse events related to RBC transfusion occur in children compared to adults, more so in neonates [14]. There are recognized potential adverse associations related to RBC transfusions unique to neonates. For example, associations between RBC transfusions and NEC, IVH, and chronic lung disease (CLD) as well as mortality have also been described [15—22].

In recent years, most institutes have implemented more restrictive transfusion guidelines to reduce the number of transfusions and donor exposures [3,4]. Two larger RCTs (the Iowa and PINT trials) have examined the transfusion criteria in the ELBW population. Both trials compared restrictive with liberal transfusion criteria for clinically relevant outcomes. Both trials developed transfusion algorithms based on the need for oxygen and the level of respiratory support in conjunction with Hb or haematocrit (Hct) levels [3,4]. Both studies found that neonates in the restrictive group had fewer RBC transfusions, without an increase in mortality or morbidity. However, one critical discrepancy was present. Bell et al [3] described increases in apnoea, severe IVH and periventricular leukomalacia in infants transfused with restrictive guidelines, but the trial was not designed to study these end points. Although the rates of serious outcomes were fairly high in both groups of the Kirpalani et al [4] trial, they found no differences in the rates of serious outcomes between infants in the restrictive vs. liberal groups.

3. Preventive strategies for anaemia of prematurity

3.1. Recombinant human EPO

Prevention and treatment of AOP with recombinant human EPO (r-HuEPO) has been the subject of many randomized controlled studies for over 20 years among over 3000 infants [2]. Although the role of EPO in the pathophysiology of AOP is well known, neither early (2—14 days of life) nor late (2—3 weeks of life) r-HuEPO therapy, nor co-treatment with iron, vitamin B12 and folate, alters the number or volume of RBC transfusions [7]. The combination of early r-HuEPO and iron does not reduce the RBC transfusion requirements in infants below 1250 g of birth weight, although the reticulocyte counts and Hct values are higher in the treatment group [23]. The use of early r-HuEPO does not significantly reduce the use of one or more RBC transfusions or the number of RBC transfusions per infant compared with late r-HuEPO administration. The finding of a statistically significant increased risk of ROP (any grade) and a similar trend for ROP stage $\geq$3 with early EPO treatment is of great concern [24]. Due to the limited benefits and the possibly increased risk of ROP, the administration of EPO is not recommended [25].

The preventive strategies for AOP are summarized in Fig. 1.

3.2. Reduction of phlebotomy losses

Iatrogenic anaemia due to the repeated removal of blood for laboratory testing is common in premature infants [11]. During the first postnatal weeks, when severe neonatal cardiorespiratory illness is at its peak and frequent laboratory testing is most intense, phlebotomy loss among preterm infants is typically the most important contributor to AOP and is the reason that a transfusion is required [26].

Strategies for reducing phlebotomy losses include microsampling, batching blood labs, cord blood sampling for immediate postnatal labs (i.e., blood type and cross-match), ordering only necessary labs, careful monitoring of phlebotomy losses, and use of blood-testing devices operated at the bedside or point of care (POC) devices [27]. Madan et al [28] suggested that after the use of the POC analyzer in their NICU, the number of RBC transfusions and the mean volume of RBC transfusions decreased by 43% and 46%, respectively, in infants with birth weights less than 1 kg. Widness et al [29] found that infants who were examined with an in-line umbilical artery catheter analyser received a significantly lower amount of RBCs than infants who were examined according to regular laboratory use during the first week of life. However, these strategies, which are dependent on a sophisticated device, are limited by economic conditions, especially in developing countries.

3.3. Enhancing placental transfusion

In animal studies, it was found that immediate cord clamping after birth results in the accumulation of approximately 30%—50% of the feto-placental blood volume in the placental unit and leaves the newborn with the same amount of reduced blood volume [30,31]. Approximately one-half of the feto-placental blood volume (nearly 110 ml/kg) remains outside the newborn infant's circulation at 30 weeks of gestation if the umbilical cord is clamped immediately [31,32]. Aladangady et al [31] were the first group that demonstrated that the blood volume of ELBW preterm newborns could be increased by delayed cord clamping (DCC) for 30—90 s in both vaginal and CS deliveries. The placental transfusion via DCC was more marked after vaginal deliveries and seemed to be more apparent for preterm infants with lower gestational ages [31].

Delayed cord clamping and umbilical cord milking (UCM) are the main methods for enhancing placental transfusion. The key difference between DCC and UCM is the mechanism of cord blood transfer to the infant. In DCC, a passive transfer of additional blood volume occurs at a slow rate, mostly by uterine contractions, whereas in UCM, an active transfer of additional blood volume occurs at a rapid rate and within a short time [33].

3.4. Delayed cord clamping

A recent Cochrane meta-analysis of data from 15 randomized trials with a total of 738 infants who were born between 24 and 36 weeks' gestation found that DCC for 30—120 s, rather than immediate clamping, seems to be associated with less need for transfusion, better circulatory stability, less IVH and a lower risk for NEC [34]. Concerns about polycythaemia and hyperbilirubinemia as well as delays in transition were not included in the review due to the heterogeneity of the trials [34]. In December 2012, the American College of Obstetricians and Gynecologists (ACOG) recommended a 30—60 s delay in umbilical cord clamping for all preterm deliveries because of the associated neonatal benefits, including increased blood volume, reduced need for RBC transfusion, and decreased incidence of IVH in preterm infants [35]. The "European Consensus Guidelines on Resuscitation of the Preterm Infant" also recommended a delay of 30—60 s before clamping the umbilical cord [36]. In a clinical trial on neurodevelopmental outcomes in preterm infants by Mercer et al [37], 58 infants who were randomized to DCC (30—45 s) had similar Bayley II scores with the control group of infants at

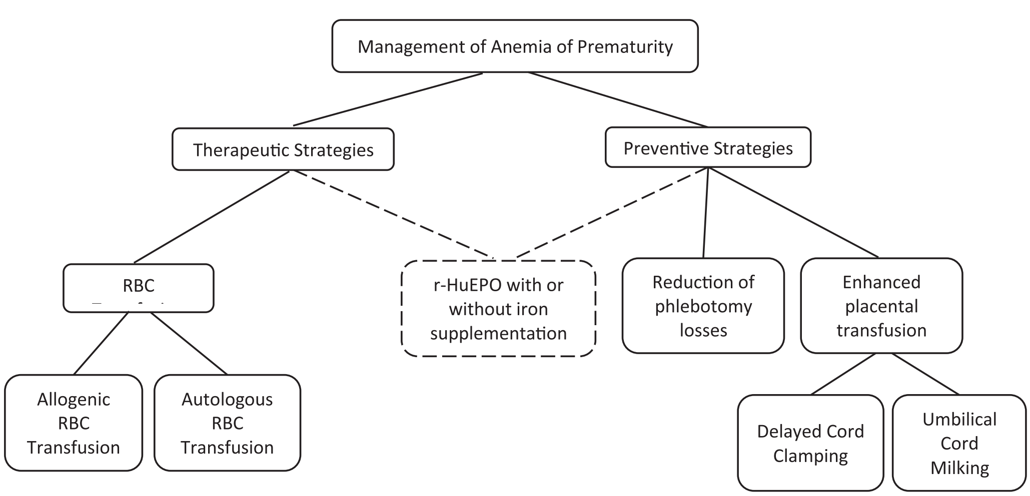

Figure 1 Therapeutic and preventive strategies for anaemia of prematurity.

seven months of age. A regression model of the effects of DCC on motor scores controlling for gestational age, IVH, bronchopulmonary dysplasia, sepsis, and male gender suggested higher motor scores in male infants with DCC [38]. More recently, Andersson et al [38] found that DCC compared with immediate cord clamping improved scores in the fine-motor and social domains at 4 years of age, especially in boys who were born at term gestation.

Although DCC has been adopted by many centers in Canada and Europe, especially in preterm populations, it has not gained wide acceptance in the United States [39]. Some researchers have noted that DCC may lead to delayed resuscitation and heat loss in infants who have a lower birth weight or a shorter gestational age [34,40]. However, it is already known that neonates who require resuscitation also need a placental transfusion as much or more so than do healthy newborns [41]. More recently, Mercer and Erickson-Owens [41] suggested that an intrapartum care provider can achieve a placental transfusion for a distressed neonate by UCM several times or can resuscitate the neonate at the perineum with an intact cord. Bringing the resuscitation to the mother's bedside is a novel concept and supports an intact cord. Adopting resuscitation with an intact umbilical cord in a hospital setting will take concentrated effort and teamwork by midwifery, obstetric, pediatric, and nursing staff [41]. They also suggested that UCM can be performed quickly by any intrapartum care provider within the current Neonatal Resuscitation Program guidelines [41]. Furthermore, a recent Cochrane review concluded that larger multi-center studies are essential and demand international collaboration to provide a scientific rationale for improving the delivery and resuscitation of the preterm infant [34]. However, more recently, the 2015 ILCOR systematic review for the neonatal resuscitation program suggested that DCC after 30 s is reasonable for both term and preterm infants who do not require resuscitation at birth. They did not recommend the routine use of UCM for ELBW infants due to concerns regarding the safety of rapid changes in blood volume by UCM for extremely preterm infants [42].

3.5. Umbilical cord milking

An alternative to DCC is UCM, in which the unclamped umbilical cord is grasped and blood is pushed toward the infant several times before it is clamped to auto-infuse blood into a preterm neonate [43]. Although Beck [44] proposed this procedure for premature babies in 1941, the first randomized controlled trial on UCM in premature babies was published by Hosono et al [40] in 2008. Hosono et al [40] were able to demonstrate reductions in both the need for RBC transfusion during the first 3 weeks of life and the total length of NICU hospitalization with UCM. They reported that the percentage of infants who had no RBC transfusions was significantly higher in the UCM group (65% vs. 30%) [40]. This study showed that UCM is a simple and cost free method that can reduce the number of RBC transfusions in preterm infants born at less than 29 weeks' gestation [40].

A second randomized controlled trial to compare UCM with ICC was published by March et al [45] in 2013. They showed that preterm infants whose cords were milked had higher Hb levels at birth and had significantly fewer transfusions in the first 14 days of life. Although the difference in the incidence of the neonatal transfusions did not reach statistical significance in their study, a 14.1% decrease was observed in the UCM group during the first 28 days of life [45]. Katheria et al [46] designed a randomized controlled trial including 60 preterm infants to investigate the effect of UCM on systemic blood flow. They found that preterm infants randomized to UCM had greater measures of superior vena cava flow and right ventricular output in the first 6 and 30 h of life. In addition, neonates receiving UCM also had greater serum Hb levels, received fewer blood transfusions, had fewer days on oxygen therapy, and had less frequent use of oxygen at 36 weeks' corrected postmenstrual age [46]. Our study group [47] previously demonstrated that UCM resulted in higher Hb levels in very low birth-weight preterm infants within the first day of life. However, the number and volume of RBC transfusions was nearly equal between the UCM and ICC groups by the end of 14 and 35 days of life and during the total length of NICU stay in our study, probably due to excessive phlebotomy losses.

More recently, Kilicdag et al [48] found that the absolute neutrophil counts (ANCs) were lower and the frequency of neutropenia was higher in the UCM group in their RCT. In contrast to former RCTs, they did not find any difference between the UCM and ICC groups in terms of the Hb and Hct levels [48].

In addition to the trials that compared UCM with ICC, there are two RCTs that aim to compare UCM with DCC [43,49]. Rabe et al [49] designed a RCT including 58 preterm infants who were randomized either to UCM or DCC for 30 s and found that the number of infants who did not need a RBC transfusion was not significantly different between the 2 intervention groups (52% for DCC and 37% for UCM). Katheria et al [43] found that UCM provides higher initial Hb levels, higher blood pressure, and improved systemic blood flow and urine output in preterm infants delivered by caesarean delivery. However, they did not find any difference in these parameters in infants delivered by vaginal delivery and speculated that more blood remained in the placenta when a neonate was delivered by caesarean section because the anaesthetic and surgical interventions interfered with the active contraction of the uterine muscles to expel the placenta [43]. Katheria et al [43] also suggested that UCM might be preferable in preterm infants delivered by caesarean delivery, particularly in newborns when immediate resuscitation is needed.

In total, umbilical cord milking has been studied in 8 RCTs and 4 controlled trials over the past two decades in preterm infants ($n = 968$) [39,40,43,45–53]. These trials are summarized in Table 1. The heterogeneity of the methodologies, primary outcomes and UCM techniques of these trials is remarkable. Furthermore, the local guidelines for RBC transfusions were not specified in most of these trials, except for the studies by Hosono et al [40], Rabe et al [49] and Alan et al [47]. Hosono et al [40] and Rabe et al [49] used more restrictive guidelines for RBC transfusion than the guideline Alan et al [47] utilized. In addition, phlebotomy losses were also not specified in

Table 1 A summary of the studies on the effect of umbilical cord milking in preterm infants.

		Population	Randomization	Blindness	UCM technique	Mode of delivery	Control condition	Phlebotomy	Guidelines for RBC transfusion	Primary outcomes
1	Katheria et al [39], 2015	154 infants, 23–32 wk	Yes	Yes	20 cm in 2 s, 4 times	CD	DCC with 45 s	Not reported	Not reported	Superior vena cava flow and right ventricular output
2	Hosono et al [48], 2015	40 infants, <29 wk	No	No	20 cm within 2 s, 1 time	CD, NSVD	UCM with 20 cm within 2 s, 2–3 times			The number of RBC transfusions and the probability of not needing a RBC transfusion
3	Kilicdag et al [46], 2015	54 infants, ≤32 wk	Yes	No	20 cm in 2 s, 4 times	CD	ICC	Not reported	Not reported	Impact of UCM on Absolute neutrophil counts
4	Patel et al [33], 2014	318 infants, <30 wk	No	No	20 cm within 2 s, 2–3 times	CD, NSVD	ICC	Not reported	Not reported	Severe IVH, NEC, death before discharge
5	Katheria et al [40], 2014	60 infants, 23 wk –31 wk 6 d	Yes	Yes	20 cm in 2 s, 3 times	CD, NSVD	ICC	Not reported	Not reported	Systemic blood flow (superior vena cava flow)
6	Alan et al [43], 2014	44 infants, ≤32 wk	Yes	No	25–30 cm, 5 cm in 1 s, 3 times	CD, NSVD	ICC	The median of 38 ml/kg in UCM and 38 ml/kg in ICC groups during the first 35 days of life	Yes	the number and volume of RBC transfusions
7	Katheria et al [49], 2014	41 infants, 23 wk –31 wk 6 d	Yes	Yes (Partial)	20 cm in 2 s, 3 times	CD, NSVD	ICC	Not reported	Not reported	HR, SpO2, MAP, and FiO2 in the delivery room
8	Christensen et al [50], 2014	First: 32 infants, 23 –40 wk. Second: 20 infants, <32 wk	No	No	In 10–15 s, 2–4 times	CD, NSVD	ICC	Not reported	Not reported	Hyperviscosity in preterm infants
9	March et al [42], 2013	75 infants, 24–28 completed wks	Yes	No	20 cm, 3 times	CD, NSVD	ICC	Not reported	Not reported	The need for RBC transfusion
10	Takami et al [51], 2012	50 infants, <29 wk	No	No	20 cm in a s, 2 –3 times	CD, NSVD	ICC	Not reported	Not reported	Cerebral and systemic perfusion

(*continued on next page*)

Table 1 *(continued)*

		Population	Randomization	Blindness	UCM technique	Mode of delivery	Control condition	Phlebotomy	Guidelines for RBC transfusion	Primary outcomes
11	Rabe et al [41], 2011	58 infants, 24 wk –32 wk 6 d	Yes	No	20 cm in 2 s, 4 times	CD, NSVD	DCC with 30 s	The median of 19 ml in UCM group 23 ml in DCC group during the first 42 d of life	Yes	To compare two strategies for enhancing placental transfusion
12	Hosono et al [38], 2008	40 infants, 24-28 wk	Yes	No	20 cm in 2 s, 2 –3 times	CD, NSVD	ICC	16 ± 3 ml/kg in the UCM group and 16.7 ± 4.8 ml/kg in the control group during first 4 wk of life	Yes	The need for RBC transfusion and morbidities

Abbreviations: CD: Caesarean delivery, HR: Heart rate, ICC: Immediate cord clamping, NSVD: Normal spontaneous vaginal delivery, RBC: Red blood cell, IVH: Intraventricular haemorrhage, MAP: Mean arterial pressure, UCM: Umbilical cord milking, wk: week.

most of these trials, except the trials performed by Hosono et al [40], Rabe et al [49] and Alan et al [47] (Table 1).

To our knowledge, there are three meta-analyses concerning UCM in preterm infants. A meta-analysis by Al-Wassia and Shah [33] evaluating the safety and efficacy of UCM at birth concluded that there was a lower risk for an oxygen requirement at 36 weeks and IVH of all grades, but no difference in the risk of mortality. A recent meta-analysis by Dang et al [54] found that UCM at preterm birth was comparatively safe and associated with a lower RBC exposure and lower incidence of IVH, NEC and death. An additional meta-analysis by Backes et al [55] concluded that enhanced placental transfusion (DCC or UCM) at birth provided better neonatal outcomes (reductions in overall mortality, lower risk of IVH, and decreased RBC transfusion incidence) in very preterm neonates.

Although the data on neurodevelopmental outcomes in premature infants are limited, some authors strongly suggest that UCM should no longer be considered experimental; rather, it is a proven intervention that ensures that premature newborns receive an adequate placental transfusion at birth [44].

4. Conclusion

Due to the increased survival of ELBW infants, AOP has become a common and serious problem for all NICUs. RBC transfusion is the only proven treatment strategy for AOP. However, there are no physiological or laboratory markers for accurate indicators of requiring RBC transfusion other than Hb values. Placental/umbilical cord blood was an untapped resource for premature infants until the last few decades. After the results and recommendations of the trials and meta-analyses failed to demonstrate that neither prevention nor treatment of AOP with r-HuEPO was effective and safe, the issue of enhanced placental transfusion has been raised. The methods for enhancing placental transfusion have many proven beneficial effects in preterm infants. This strategy clearly results in higher Hb levels at birth and a reduced need for RBC transfusions, as well as creating a better haemodynamic status during the initial hours of life. To date, the enhancement of placental transfusion in the delivery room either by DCC or UCM seems to be the best preventive measure for AOP. The effects of placental transfusion probably vary in vaginal deliveries and caesarean sections. Although the safety of rapid changes in blood volume via UCM for preterm infants remains a concern, there are no data for haemodynamic adverse effects up to now. Furthermore, studies assessing neurodevelopmental outcomes are insufficient. However, resuscitation events are not reported consistently, precluding the determination of the optimal cord clamping practice among neonates born severely depressed or requiring immediate resuscitation. Large-scale, randomized clinical trials of enhanced placental transfusion strategies to assess long-term neurodevelopmental and neurologic sequelae are needed.

One should also keep in mind that blood elements other than red blood cells are transfused via DCC or UCM and that the clinical results of the transfusion of these blood elements have not been evaluated.

5. Recommendations

- Enhancing placental transfusion by DCC (at least for 30 s) or UCM (2—4 times) during delivery seems to be the best approach for preterm infants who do not require resuscitation. Sufficient data for performing resuscitation with intact cords are still lacking.
- Cord blood should be used for initial laboratory tests.
- Phlebotomy losses should be reduced.
- Nutrition support with vitamins (folate, B12 and vitamin E) and iron should be optimized.
- Individual transfusion guidelines should be established and utilized.

Conflict of interest

The authors declare no conflict of interest.

Ethical approval

No need to obtain ethical approval for this review

References

[1] Carroll PD. Umbilical cord blood-an untapped resource: strategies to decrease early red blood cell transfusions and improve neonatal outcomes. Clin Perinatol 2015;42(3):541—56.
[2] Juul S. Erythropoietin in anemia of prematurity. J Matern Fetal Neonatal Med 2012;25(5):80—4.
[3] Bell EF, Strauss RG, Widness JA, Mahoney LT, Mock DM, Seward VJ, et al. Randomized trial of liberal versus restrictive guidelines for red blood cell transfusion in preterm infants. Pediatrics 2005;115:1685—91.
[4] Kirpalani H, Whyte RK, Andersen C, Asztalos EV, Heddle N, Blajchman MA, et al. The Premature Infants in Need of Transfusion (PINT) study: a randomized, controlled trial of a restrictive (low) versus liberal (high) transfusion threshold for extremely low birth weight infants. J Pediatr 2006;149:301—7.
[5] Martin JA, Hamilton BE, Sutton PD, Ventura SJ, Mathews TJ, Osterman MJ. Births: final data for 2008 National Vital Statistics Reports. Centers Dis Control Prev 2009;57:7.
[6] Christensen RD, Ilstrup S. Recent advances toward defining the benefits and risks of erythrocyte transfusions in neonates. Arch Dis Child Fetal Neonatal Ed 2013;98(4):F365—72.
[7] Crowley M, Kirpalani H. A rational approach to red blood cell transfusion in the neonatal ICU. Curr Opin Pediatr 2010;22(2):151—7.
[8] Wardrop CA, Holland BM, Veale KE, Jones JG, Gray OP. Nonphysiological anaemia of prematurity. Arch Dis Child 1978;53:855—60.
[9] Strauss RG. Anaemia of prematurity: pathophysiology and treatment. Blood Rev 2010;24(6):221—5.
[10] Strauss RG, Widness JA. Is there a role for autologous/placental red blood cell transfusions in the anemia of prematurity? Transfus Med Rev 2010;24(2):125—9.
[11] Aher S, Malwatkar K, Kadam S. Neonatal anemia. Semin Fetal Neonatal Med 2008;13(4):239—47.
[12] Stockman JA 3rd, Graeber JE, Clark DA, McClellan K, Garcia JF, Kavey RE. Anemia of prematurity: determinants of the erythropoietin response. J Pediatr 1984;105:786—92.
[13] Sacher RA, Luban NL, Strauss RG. Current practice and guidelines for the transfusion of cellular blood components in the newborn. Transfus Med Rev 1989;3:39—54.
[14] Stainsby D, Jones H, Wells AW, Gibson B, Cohen H. 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—9.
[15] Venkatesh V, Khan R, Curley A, Hopewell S, Doree C, Stanworth S. The safety and efficacy of red cell transfusions in neonates: a systematic review of randomized controlled trials. Br J Haematol 2012;158:370—85.
[16] Mohamed A, Shah PS. Transfusion associated necrotizing enterocolitis: a meta-analysis of observational data. Pediatrics 2012;129:529—40.
[17] Baer VL, Lambert DK, Henry E, Snow GL, Butler A, Christensen RD. Among very-low-birth-weight neonates is red blood cell transfusion an independent risk factor for subsequently developing a severe intraventricular hemorrhage? Transfusion 2011;51:1170—8.
[18] Christensen RD. Associations between "early" red blood cell transfusion and severe intraventricular hemorrhage, and between "late" red blood cell transfusion and necrotizing enterocolitis. Semin Perinatol 2012;36:283—9.
[19] Giannantonio C, Papacci P, Cota F, Vento G, Tesfagabir MG, Purcaro V, et al. Analysis of risk factors for progression to treatment-requiring ROP in a single neonatal intensive care unit: is the exposure time relevant? J Matern Fetal Neona 2012;25:471—7.
[20] Cooke RW, Drury JA, Yoxall CW, James C. Blood transfusion and chronic lung disease in preterm infants. Eur J Pediatr 1997;156:47—50.
[21] Valieva OA, Strandjord TP, Mayock DE, Juul SE. Effects of transfusions in extremely low birth weight infants: a retrospective study. J Pediatr 2009;155:331—7.
[22] Dos Santos AM, Guinsburg R, de Almeida MF, Procianov RS, Leone CR, Marba ST, et al. Red blood cell transfusions are independently associated with intra-hospital mortality in very low birth weight preterm infants. J Pediatr 2011;159:371—6.
[23] Ohls RK, Ehrenkranz RA, Wright LL, Lemons JA, Korones SB, Stoll BJ, et al. Effects of early erythropoietin therapy on the transfusion requirements of preterm infants below 1250 grams birth weight: a multicenter, randomized, controlled trial. Pediatrics 2001;108:934—42.
[24] Aher SM, Ohlsson A. Early versus late erythropoietin for preventing red blood cell transfusion in preterm and/or low birth weight infants. Cochrane Database Syst Rev 2012;17:10. CD004865.
[25] Ohlsson A, Aher SM. Early erythropoietin for preventing red blood cell transfusion in preterm and/or low birth weight infants. Cochrane Database Syst Rev 2014;26:4. CD004863.
[26] Widness JA. Pathophysiology of anemia during the neonatal period, including anemia of prematurity. Neoreviews 2008; 9(11):e520.
[27] Bishara N, Ohls RK. Current controversies in the management of the anemia of prematurity. Semin Perinatol 2009;33(1):29—34.
[28] Madan A, Kumar R, Adams MM, Benitz WE, Geaghan SM, Widness JA. Reduction in red blood cell transfusions using a bedside analyzer in extremely low birth weight infants. J Perinatol 2005;25(1):21—5.
[29] Widness J, Madan A, Grindeanu L, Zimmerman MB, Wong DK, Stevenson DK. Reduction in red blood cell transfusions among preterm infants: results of a randomized trial with an in-line blood gas and chemistry monitor. Pediatrics 2005;115:1299—306.

[30] Brace RA. Regulation of blood volume in utero. In: Hanson MA, Spencer JAD, Rodeck CH, editors. Fetus and neonate: physiology and clinical applications. Cambridge, United Kingdom: Cambridge University Press; 1993. p. 75—99.
[31] Aladangady N, McHugh S, Aitchison TC, Wardrop CA, Holland BM. Infants' blood volume in a controlled trial of placental transfusion at preterm delivery. Pediatrics 2006; 117(1):93—8.
[32] Linderkamp O. Placental transfusion: determinants and effects. Clin Perinatol 1982;9:559—92.
[33] Al-Wassia H, Shah PS. Efficacy and safety of umbilical cord milking at birth: a systematic review and meta-analysis. JAMA Pediatr 2015;169(1):18—25.
[34] Rabe H, Diaz-Rossello JL, Duley L, Dowswell T. Effect of timing of umbilical cord clamping and other strategies to influence placental transfusion at preterm birth on maternal and infant outcomes. Cochrane Database Syst Rev 2012;8. http://dx.doi.org/10.1002/14651858. CD003248.
[35] Committee on Obstetric Practice, American College of Obstetricians and Gynecologists. Committee opinion no. 543: timing of umbilical cord clamping after birth. Obstet Gynecol 2012;120(6):1522—6.
[36] Sweet DG, Carnielli V, Greisen G, Hallman M, Ozek E, Plavka R, et al., European Association of Perinatal Medicine. European consensus guidelines on the management of neonatal respiratory distress syndrome in preterm infants—2013 update. Neonatology 2013;103(4):353—68.
[37] Mercer JS, Vohr BR, Erickson-Owens DA, Padbury JF, Oh W. Seven-month developmental outcomes of very low birth weight infants enrolled in a randomized controlled trial of delayed versus immediate cord clamping. J Perinatol 2010; 30(1):11—6.
[38] Andersson O, Lindquist B, Lindgren M, Stjernqvist K, Domellöf M, Hellström-Westas L. Effect of delayed cord clamping on neurodevelopment at 4 years of age: a randomized clinical trial. JAMA Pediatr 2015;169(7):631—8.
[39] Patel S, Clark EA, Rodriguez CE, Metz TD, Abbaszadeh M, Yoder BA. Effect of umbilical cord milking on morbidity and survival in extremely low gestational age neonates. Am J Obstet Gynecol 2014;211(5):519. e1-7.
[40] Hosono S, Mugishima H, Fujita H, Hosono A, Minato M, Okada T, et al. Umbilical cord milking reduces the need for red cell transfusions and improves neonatal adaptation in infants born at less than 29 weeks' gestation: a randomised controlled trial. Arch Dis Child Fetal Neonatal Ed 2008;93(1): F14—9.
[41] Mercer JS, Erickson-Owens DA. Is it time to rethink cord management when resuscitation is needed? J Midwifery Womens Health 2014;59(6):635—44.
[42] American Heart Association. Web-based integrated guidelines for cardiopulmonary resuscitation and emergency cardiovascular care — part 13: neonatal resuscitation. ECCguidelines.heart.org.
[43] Katheria AC, Truong G, Cousins L, Oshiro B, Finer NN. Umbilical cord milking versus delayed cord clamping in preterm infants. Pediatrics 2015;136(1):61—9.
[44] Beck AC. How can the obstetrician aid in reducing the mortality of prematurely born infants? Am J Obstet Gynecol 1941; 42:355—64.
[45] March MI, Hacker MR, Parson AW, Modest AM, de Veciana M. The effects of umbilical cord milking in extremely preterm infants: a randomized controlled trial. J Perinatol 2013; 33(10):763—7.
[46] Katheria AC, Leone TA, Woelkers D, Garey DM, Rich W, Finer NN. The effects of umbilical cord milking on hemodynamics and neonatal outcomes in premature neonates. J Pediatr 2014;164(5):1045—50.
[47] Alan S, Arsan S, Okulu E, Akin IM, Kilic A, Taskin S, et al. Effects of umbilical cord milking on the need for packed red blood cell transfusions and early neonatal hemodynamic adaptation in preterm infants born $<$1500 g: a prospective, randomized, controlled trial. J Pediatr Hematol Oncol 2014; 36(8):e493—8.
[48] Kilicdag H, Gulcan H, Hanta D, Torer B, Gokmen Z, Ozdemir SI, et al. Is umbilical cord milking always an advantage? J Matern Fetal Neonatal Med 2015:1—4 [Epub ahead of print].
[49] Rabe H, Jewison A, Alvarez RF, Crook D, Stilton D, Bradley R, et al. Brighton Perinatal Study Group. Milking compared with delayed cord clamping to increase placental transfusion in preterm neonates: a randomized controlled trial. Obstet Gynecol 2011;117:205—11.
[50] Hosono S, Mugishima H, Takahashi S, Takahashi S, Masaoka N, Yamamoto T, et al. One-time umbilical cord milking after cord cutting has same effectiveness as multiple-time umbilical cord milking in infants born at $<$29 weeks of gestation: a retrospective study. J Perinatol 2015;35(8):590—4.
[51] Katheria A, Blank D, Rich W, Finer N. Umbilical cord milking improves transition in premature infants at birth. PLoS One 2014;9(4):e94085.
[52] Christensen RD, Baer VL, Gerday E, Sheffield MJ, Richards DS, Shepherd JG, et al. Whole-blood viscosity in the neonate: effects of gestational age, hematocrit, mean corpuscular volume and umbilical cord milking. J Perinatol 2014;34(1):16—21.
[53] Takami T, Suganami Y, Sunohara D, Kondo A, Mizukaki N, Fujioka T, et al. Umbilical cord milking stabilizes cerebral oxygenation and perfusion in infants born before 29 weeks of gestation. J Pediatr 2012;161(4):742—7.
[54] Dang D, Zhang C, Shi S, Mu X, Lv X, Wu H. Umbilical cord milking reduces need for red cell transfusions and improves neonatal adaptation in preterm infants: meta-analysis. J Obstet Gynaecol Res 2015;41(6):890—5.
[55] Backes CH, Rivera BK, Haque U, Bridge JA, Smith CV, Hutchon DJ, et al. Placental transfusion strategies in very preterm neonates: a systematic review and meta-analysis. Obstet Gynecol 2014;124(1):47—56.