Association between Red Blood Cell Transfusions and Necrotizing Enterocolitis

In the mid-1980s, one hospital's neonatal intensive care unit (NICU) recognized 20 cases of necrotizing enterocolitis (NEC) during a 3-month period.[1] During those 3 months, NEC developed in 31% of their very low birth weight (VLBW) neonates and in 11% of infants with birth weight >1500 g.[1] The Centers for Disease Control and Prevention was invited to assist the hospital in investigating this outbreak. Their epidemiologic investigation revealed only one significant association: red blood cell (RBC) transfusion. The association was highly significant, with an OR for NEC after transfusion of 15.1 (95% CI, 2.6-92.5). No changes in blood bank procedures or blood supply were identified as potentially culpable, and the outbreak ceased without institution of any specific changes in transfusion practice.

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Almost 10 years later, Bednarek et al[2] reported an association between transfusion practice and NEC in 6 Boston area NICUs. Using multivariate analysis, these authors found that two NICUs transfused about 70 mL/kg more RBCs over the entire NICU stay compared with the two lowest-transfusing NICUs. VLBW neonates cared for in the high-transfusing NICUs demonstrated trends toward more intraventricular hemorrhage and bronchopulmonary dysplasia, but the only statistically significant association was an increase in NEC, with a prevalence of 7% (15/232) in the high-transfusing NICUs (adjusted OR, 1.1; 95% CI, 0.5-2.2) compared with 2% (5/280) in the low-transfusing NICUs (adjusted OR, 0.3; 95% CI, 0.1-0.8; $P < .05$).

Subsequent case reports and small retrospective studies described this same association. Focusing exclusively on cases of Bell stage III NEC, in which surgical and pathological evidence confirmed that the disease entity under study was indeed NEC, we also found this association, with 40 of 112 NEC cases occurring within 48 hours (mean, 18 hours) following a RBC transfusion.[3]

At this point, it seems certain that an association indeed exists between "late" RBC transfusions (given after the first several weeks) and the development of NEC. Caution must be taken when interpreting the clinical relevance of this association, however. First, not all cases of NEC follow a RBC transfusion. In fact, as emphasized by Josephson et al[4] and Blau et al,[5] the majority of cases NEC are not temporally related to a transfusion. Second, the great majority of transfusions administered in the NICU are not followed by the development of NEC. Given these two certainties, it is clear that transfusions are not *the* cause of NEC. The critical question is whether in *some* cases of NEC, an immediately antecedent RBC transfusion is part of the pathogenesis.

Another area of caution applies to any association identified in retrospective studies. Such studies are inherently limited by their susceptibility to bias and by a failure to recognize confounding variables. In a retrospective analysis, an association between a variable and an outcome, even if statistically significant, does not prove a cause-and-effect relationship. Thus, the antecedent transfusion might have no pathogenic role in NEC development. Rather, the transfusion might be an epiphenomenologic marker of imminent NEC. Determining whether transfusions cause NEC will require a different experimental approach than has been reported to date.

Despite the remaining uncertainties, several common elements can be found among the largest studies reporting transfusion-associated NEC[3-5] that can guide the formulation of new concepts in the pathogenesis of this apparent NEC subtype. These common elements include the following: (1) Between 25% and 35% of NEC cases follow reasonably closely after a RBC transfusion (between 2 and 48 hours, but generally closer to 12 hours); (2) neonates with transfusion-associated NEC are generally born at much earlier gestation than those who develop NEC unrelated to transfusion; (3) neonates with transfusion-associated NEC have had one or more previous RBC transfusions; (4) neonates with transfusion-associated NEC are generally 3-5 weeks old, whereas those with NEC unrelated to transfusion are generally younger (1-3 weeks old); and (5) the age of the blood transfused (days since donor draw) is not different between those with transfusion-associated NEC and matched controls who underwent transfusion but did not develop NEC. Similarly, the transfused blood is not older in neonates with transfusion-associated NEC compared with matched controls who developed NEC remotely from their last transfusion (ie, more than 72 hours after transfusion).

Transfusion-associated NEC has no proven pathogenic mechanism, but at least 3 plausible explanations have been proposed. These include the hypothesis proposed by Blau et al[5] in this issue of *The Journal* that transfusion-

NEC	Necrotizing enterocolitis
NICU	Neonatal intensive care unit
RBC	Red blood cell
VLBW	Very low birth weight

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associated NEC has parallels with the transfusion-related acute lung injury reaction and thus might involve similar immunologic mechanisms, but in the intestine rather than lung.

A second possible explanation involves the reason for the transfusion. Transfusions antecedent to NEC are invariably given for a clinical purpose, in most cases a hemoglobin or hematocrit value below a "trigger" level, with a transfusion ordered according to preset guidelines. Anemia can impair blood flow to the intestine, possibly constituting an injury relevant to NEC pathogenesis.

A third possible explanation involves the well-known storage lesions that occur in banked RBC, including reduced deformability, increased RBC adhesion and aggregation, prothrombotic effects of transfusions, and nitric oxide deficiency.[6] In principle, RBC transfusions should increase oxygen delivery to tissues, but, paradoxically, the reverse can occur, as reported by Reynolds et al[7] and Gladwin and Kim-Shapiro.[8] Erythrocyte nitric oxide levels are depleted during storage, which can severely impair RBCs' hypoxic vasodilatory activity.[7-9] Transfused RBCs could act as a nitric oxide sink, predisposing to vasoconstriction and ischemic insult. Because RBCs traverse the microcirculation in line, impaired vasodilatation due to nitric oxide depletion could temporarily reduce perfusion in relevant parts of the intestinal microvasculature. This problem has been observed after RBC transfusion in adult patients, with transfusion failing to improve oxygen delivery and also being associated with ischemic events.[10] Although we have characterized the foregoing as 3 different possible pathogenic mechanisms, they might not be mutually exclusive; conceivably, elements of all 3 mechanisms could be operative.

An important clinical issue that remains incompletely addressed is whether withholding feedings during RBC infusion diminishes the risk of developing transfusion-associated NEC. In 2005, Agwu and Narchi[11] answered that question in this way: "Withholding feedings during a transfusion has theoretical benefits, but there is no published evidence to support this practice. Despite a lack of evidence, we withhold feeding during transfusions." El-Dib et al,[12] reporting from Children's National Medical Center (Washington, DC), reported a decrease in the prevalence of NEC in VLBW infants from 5.3% to 1.3% ($P < .05$) following a practice change of withholding feedings during RBC transfusions. A similar reduction in NEC prevalence was recently noted in Nashville after withholding feedings during transfusion (M. Sami Ismail and E. Scott Palmer, personal communication, Centennial Medical Center, Nashville, Tennessee, October 1, 2010). In any before–after analysis of a practice change, excluding the possibility of the Hawthorne effect is difficult. Thus, any benefit of withholding feedings during RBC transfusion remains speculative at this point. Moreover, the optimum period for withholding feedings both before and after transfusion is an important but as-yet untested variable.

Given an increasing body of evidence, it now seems proper to consider "transfusion-associated NEC" as a legitimate subtype of NEC. Although this subtype accounts for a minority of NEC cases, its pathogenesis remains unclear, and preventative measures are largely speculative, the body of work on this topic is accumulating rapidly. This high level of interest, activity, and commitment engenders hope that such efforts will eventually lead to a lower prevalence of this very troublesome disorder. ■

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