RESEARCH PAPER

Relationship between Packed Red Blood Cell Transfusion and Severe Form of Necrotizing Enterocolitis: A Case Control Study

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Objective: To determine if packed red blood cell transfusion is associated with onset of necrotizing enterocolitis, and whether withholding feed has any association with it.

Methods: Case records of 100 preterm neonates, (<34 weeks gestation) who developed necrotizing enterocolitis and 99 random age-and gestation-matched controls were evaluated for any blood transfusion 48 h before onset of necrotizing enterocolitis.

Results: During the study period 26% infants received packed red blood cell transfusion within 48-hours prior to onset of disease and 84% of these infants were not fed around the time of transfusion. Infants who developed necrotizing enterocolitis after transfusion were older, of lower gestational age, birth weight and more likely to develop stage 3 disease. They had a lower hematocrit at birth and before onset of disease and withholding feeds around transfusion did not prevent necrotizing enterocolitis. Odds of mortality in these infants was 2.83 (95% CI 0.97-8.9) and survivors had no significant difference in incidence of periventricular leukomalacia and length of hospital stay.

Conclusion: Blood Transfusion associated necrotizing enterocolitis is a severe, mainly surgical form of disease.

Keywords: *Blood transfusion, Feeding, Periventricular leukomalacia, Prematurity.*

Necrotizing enterocolitis (NEC) afflicts 6-10% of very low birth weight (<1500 g) infants, and leads to higher morbidity, mortality and increased length of hospital stay [1]. Several risk factors such as intestinal immaturity, a genetic predisposition and abnormal microbial colonization of the intestines predispose premature infants to develop NEC [2]. Recently an association between receiving packed red blood cell (PRBC) transfusions and the onset of NEC has been observed [3-5]. Majority of very low birth weight (VLBW) infants will require one or more PRBC transfusions during the course of their neonatal intensive care unit (NICU) stay. It is unclear whether withholding enteral feedings around the time of transfusion is effective in reducing or preventing transfusion-associated NEC. We undertook this study to observe association of PRBC transfusion and NEC at our institution, and whether the practice of withholding feeds around the time of PRBC transfusion is helpful in prevention of this type of NEC.

Methods

Our study was conducted in the Level 3b NICU at Vidant Medical Center that serves as the regional referral center for 29 counties in Eastern North Carolina, caring for both inborn and outborn infants with over 1000 admissions to the NICU annually. It is affiliated teaching hospital for the Brody School of Medicine at East Carolina University. This study was Health Insurance Portability and Accountability Act (HIPPA) compliant and approved by the combined Institutional Review Board of East Carolina University and Vidant Medical Center.

Our institutional database was searched for all NICU admissions the time period from January 1, 2002 through December 31, 2011 to identify infants with a gestational age at birth ≤34 weeks who developed stage 2a or greater NEC using modified Bell's Criteria [6]. Infants with known chromosomal anomalies, congenital heart disease, or a diagnosis of an isolated spontaneous intestinal perforation were excluded. Eligible matched controls were identified by matching 1 to 1 with each case patient for gestational age at birth (±10 days), admission date (±4 weeks) and birth weight (±150g). The electronic medical records for each of these cases and controls were then reviewed and data elements collected for further analysis.

Maternal and infant characteristics historically associated with an increased risk of developing NEC were recorded. Maternal factors included the following: pregnancy induced hypertension, chorioamnionitis,

administration of antenatal steroids, preterm premature rupture of membranes (PPROM), and prolonged rupture of membranes (PROM). Infant data elements collected included; gestational age at birth, birth weight, gender, mode of delivery, Apgar scores at 1 and 5 minutes, pre-transfusion hematocrit, corrected gestational age at onset of NEC, relationship of NEC onset to a transfusion in the preceding 48 hours, and severity of NEC along with any therapy provided. Additional infant data elements included the presence of a hemodynamically significant patent ductus arteriosus (PDA) and treatment, if any, hypotension and therapy provided, type of feeding (breast milk or formula feedings), administration of postnatal steroids for prevention or treatment of chronic lung disease, and enteral feeding status at the time of any transfusion. Data on outcome variables included: diagnosis of bronchopulmonary dysplasia (BPD) as defined by oxygen requirement at 36 weeks corrected gestational age, periventricular leucomalacia (PVL) as defined by ultrasonography scan at 36 weeks corrected gestational age, retinopathy of prematurity (ROP), length of stay, and death prior to hospital discharge. The decision to provide a PRBC transfusion was at the discretion of attending physician. Our hospital policy throughout the study period was that infants who required a PRBC transfusion were transfused with PRBC that had been stored for less than five days. We did not divide an adult unit of blood into aliquots to limit donor exposure. All transfused PRBC units were cytomegalovirus (CMV) negative, irradiated and leuko-reduced. Donor blood was collected and stored in citrate phosphate dextrose (CPD) solution.

Statistical methods: Statistical analyses were performed with SPSS 20. Chi-square tests and Fisher's exact tests were done to measure degree of association between categorical variables. Analysis of variance (ANOVA) was used for comparisons involving continuous variables between NEC and non-NEC. A simple logistic regression was performed to calculate odds of having major outcomes variables like mortality and PVL. Statistical significance was set at a $P < 0.05$.

RESULTS

Over the ten year period from January 1, 2002 to December 31, 2011, there were 9022 admissions to the NICU, and 131 neonates were identified as having had NEC. Thirty-one infants did not meet the predetermined inclusion criteria leaving a total of 100 patients with NEC for evaluation (***Fig.* 1**).

The overall incidence of NEC during the study period was 1.45% of all admissions to the NICU, and 3.67% in infants ≤34 weeks gestation. The incidence of NEC did not vary significantly at our center during the 10 year study period. The NEC and matched control groups were similar in gestational age at birth and most other demographic variables but transfusion-associated NEC infants were significantly smaller in birth weight compared to other infants with NEC and non NEC controls ($P<0.05$) (***Table* I**). There were more female and Hispanic subjects in the transfusion-associated NEC group, and a larger proportion of them had hemodynamically significant PDA which required surgical ligation ($P<0.05$). A significantly greater number of mothers of transfusion-associated NEC infants were diagnosed with pregnancy-induced hypertension as compared to controls (***Table* I**).

Infants with transfusion-associated NEC had a significantly lower hematocrit at birth, and also a lower

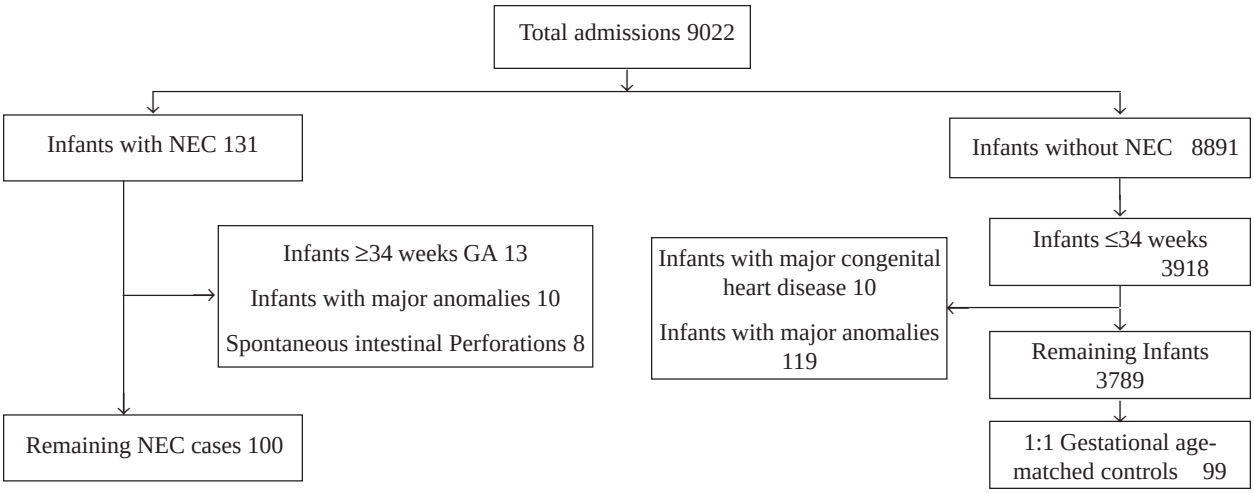

FIG. 1 *Patient and control selection flow chart with inclusion and exclusion criteria.*

hematocrit just before onset of NEC (***Table* II**). Infants with transfusion-associated NEC were of lower gestational age at birth, had lower birth weight, and were more likely to have had a 5 minute Apgar score of less than 6. Infants in the transfusion-associated group were more likely to be of blood type B+ and less likely to be blood type A+. The rates of premature rupture of membranes, maternal PIH and absent end diastolic umbilical flow were similar between the two groups of NEC.

Infants with transfusion-associated NEC were older at the time when NEC developed, and had a higher rate of surgical (stage 3a+3b) NEC. They were also more likely to have had their feedings held around the time of blood transfusion.

DISCUSSION

Our study demonstrated an association between PRBC transfusion and onset of NEC in about a quarter of our infants over the 10-year study period. These infants were smaller in their birth weight, were born at lower gestational age, and were more likely to have had a hemodynamically significant PDA. Infants in our study did not demonstrate a protective effect of withholding feedings before blood transfusion.

This main limitations of the study are: a retrospective observational design; predominantly African-American population, and a small sample size. Absence of a standard feeding protocol or a policy on withholding feedings at the time of transfusion during the study period precluded demonstration of a strong relationship between PRBC transfusion and NEC.

Some recent studies have identified a similar association between transfusion and transfusion-associated NEC [5, 7-15]. A meta-analysis of several of these studies concluded that there is a strong association

TABLE I DEMOGRAPHIC CHARACTERISTICS OF NEC INFANTS AND CONTROLS

	Transfusion-associated NEC (n=26)	*Other NEC (n=73)*	*Controls (n=99)*
*Gestational age (Wks)	27.3 (2.5)	29.2 (3.0)	28.7 (2.9)
*Birth weight (g)	992.8 (377.6)	1287.7 (457.2)	1260.7 (785)
#Male infants	5 (19.2)	46 (63.0)	55 (55.6)
Race: White	4 (15.4)	19 (26.0)	30 (30.3)
Race: African American	18 (69.2)	51 (69.9)	67 (67.7)
Caesarian delivery	15 (57.7)	35 (48.6)	65 (65.7)
Apgar score >6 at 5 min	11 (42.3)	55 (75.3)	63 (64.9)
#PDA	15 (57.7)	28 (38.4)	46 (46.5)
Feeds: Breast Milk	10 (40)	27 (39.1)	39 (50.6)
*Hematocrit at Birth	42.6 (6.9)	47.7 (8.1)	45.5 (7.7)
Pregnancy induced hypertension	3 (11.5)	12 (16.4)	3 (3.0)

*NEC: Necrotizing enterocolitis; PDA:patent ductus arteriosus Values in No. (%) or *mean (SD); #Significantly (P<0.05) different between transfusion-associated and other NEC group.*

TABLE II CLINICAL STATUS AT ONSET OF NEC AND OUTCOMES IN TWO GROUPS

	Transfusion-associated NEC (n=26)	*Other NEC (n=73)*	*P value*
*Hematocrit at birth (%)	42.6 (6.9)	47.7 (8.1)	0.006
*Pre-transfusion hematocrit (%)	27.4 (4.5)	34.5 (8.5)	<0.001
*Age at onset of NEC (d)	28.4 (13)	19.0 (14.9)	0.005
Feeding withheld around transfusion	22 (84.6)	25 (36.8%)	<0.001
Deaths	8 (30.8)	10 (13.7)	0.08
ROP	8/20 (40.0)	21/54 (38.9)	0.93
PVL	4/24 (16.7)	5/63 (7.9)	0.25
*Length of stay (d)	73.5 (48.6)	54.3 (44.6)	0.07

*NEC: Necrotizing enterocolitis; ROP: Retinopathy of prematurity; PVL: Perventricular leukomalacia; Values in No. (%) or * mean (SD).*

WHAT IS ALREADY KNOWN?

- Onset of Necrotizing enterocolitis is preceded by blood transfusion in some cases and it leads to a severe form of disease with high morbidity and mortality.

WHAT THIS STUDY ADDS?

- Blood transfusion-associated Necrotizing enterocolitis seems to be a severe form of disease and withholding feedings around the time of transfusion does not seem to prevent this entity.

between PRBC transfusion and odds of developing NEC [16]. A more recent meta-analysis has been more cautious in reaching the same conclusion [17]. On the other hand, two recent reports did not find PRBC transfusion as a risk factor for NEC [18,19]. Some investigators have reported a significant decrease in the overall NEC rate following the institution of a policy of withholding enteral feedings during and after PRBC transfusion [20]. However, in our study, 84.6% of transfusion-associated NEC cases had their enteral feedings withheld during the time of transfusion as well as for several hours afterwards, suggesting that withholding enteral feedings around the time of a transfusion may not prevent NEC.

In conclusion, our study suggests a temporal association of a PRBC transfusion with the development of NEC that has also been reported by others. Our data also suggest that infants with transfusion-associated NEC represent a subset that is more likely to be seen in smaller VLBW infants. Withholding enteral feedings around the time of a PRBC transfusion also does not seem to be an effective strategy to decrease the occurrence of transfusion-associated NEC.

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