ORIGINAL ARTICLE ›››

Beneficial Erythropoietic Effects of Recombinant Human Erythropoietin in Very Low-Birth Weight Infants: A Single-Center Randomized Double-Blinded Placebo-Controlled Trial

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─── ABSTRACT ───

Purpose: Due to appearing serious a complication following repeated blood products transfusion in premature neonates, beneficial protective effects of recombinant human erythropoietin (rhEpo) is being widely investigated. The present study evaluated the effectiveness of rhEpo in preventing anemia of prematurity by assessment of the changes in serum erythropoiesis parameters in very low birth weight (VLBW) neonates. **Materials and Methods:** In a single-center randomized, double-blinded, placebo-controlled trial, 60 premature infants with birth weight <1500g who were born between 24 and 31 weeks of gestation admitted to the neonatal intensive care unit of Fatemieh Hospital in Hamadan were enrolled. The study medication (rhEpo or placebo) was randomly assigned to each patient. The rhEpo (250 U rhEpo/kg body weight at birth, equal to 1 mL solution/kg birth weight) and placebo drug solutions were indistinguishable. The study drug was administered 3 times weekly for 3 weeks. **Results:** Monitoring hemoglobin, hematocrit and reticulocyte count showed no relevant deviation from baseline to the 2nd week of the study. However, there was relevant difference for these parameters between investigated patients who received rhEpo and the patients who received placebo during the 3rd week of the medication. 16 (53.3%) in the intervention group and 20 (66.7%) in the control group required blood transfusion with an insignificant difference. No serious side-effects were observed in either the rhEpo or placebo group. **Conclusion:** Our study suggested that rhEpo is an effective drug for improving erythropoiesis parameters after 3 weeks of administration in VLBW infants regardless its effect on the need for blood transfusions.

Key words:
Anemia, clinical trial, erythropoietin, prematurity, neonate

INTRODUCTION

Because of the expected declining hemoglobin level in premature infants during the 1st weeks of life, therapeutic and curative approaches such as repeated blood transfusion in these infants are potentially considered.[1] Various investigations have linked this phenomenon in very low birth weight (VLBW) infants to reduced fetoplacental transfusion, decreased mean life of neonatal erythrocytes, excessive blood sampling for diagnostic tests, as well as erythropoietin releasing interruption.[2] This type of infantile anemia is associated with considerable low reticulocyte count and deficient erythropoietin production.[3] Despite important advances in the safety of blood transfusion and blood derivatives, concerns have been raised because of the appearance of serious complications such as infections after the use of these blood products. Recent efforts have focused on reducing use of these products and replacing with other appropriate and effective treatment regimens including recombinant human erythropoietin (rhEpo).[4-8] The beneficial protective effects of rhEpo have been most intensively investigated in experimental animal models

and also in different clinical trials of adult humans. Furthermore, some studies on premature infants could show that the beginning course of rhEpo in VLBW infants within the 1st weeks of life might has significant effects on preventing need to erythrocyte transfusions and its-related complications.[9-12]

The present study aimed to evaluate the effectiveness of rhEpo therapy in preventing anemia of prematurity and its progression by assessment of the changes in erythropoiesis

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parameters such as hemoglobin, hematocrit and reticulocyte count in VLBW infants.

MATERIALS AND METHODS

This was a single-center phase II clinical trial designed as a randomized, double-blinded, placebo-controlled trial. Premature infants with birth weight <1500 g who were born between 24 and 31 weeks of gestation admitted to the neonatal intensive care unit of Fatemieh Hospital in Hamadan were eligible for enrollment. All included infants were more than 6 days old and their daily calorie consumption was >50 kcal/kg; at least half of this energy was received orally. The exclusion criteria were: Severe congenital malformations, dysmorphic syndromes, hemolytic anemia and acute severe infection. The trial was approved by the ethical committee of the Hamadan University of Medical Sciences. Written informed consent was obtained from the parents of eligible infants. The infants were randomly assigned in a 1:1 allocation in favor of rhEpo, using a computer-based random-number generator. The study medication (rhEpo or placebo) was randomly assigned to each patient. The rhEpo and placebo drug solutions were indistinguishable. PDpoietin (250 U rhEpo/kg body weight at birth, equal to 0.1 mL solution/kg birth weight; Pooyesh Darou Inc., Iran) or an equivalent volume of normal saline placebo was given subcutaneously 3 days a week every other day on Monday, Wednesday and Friday. Treatment in both groups continued until 36 weeks corrected gestation or discontinued if the hematocrit exceeded 0.45 at any time. All injections were performed subcutaneously. All patients in both groups were also given oral iron as ferrous sulfate, 4 mg/kg/day during the entire treatment period. All infants were assessed for hemoglobin, hematocrit and reticulocyte count at 1, 2 and 3 weeks using the Sysmex KX21N hematology analyzer. The infants were weighed weekly on a Kubota Baby scale. The numbers of blood transfusions were recorded.

A specific transfusion protocol used to administer transfusions for all subjects. Transfusion guidelines for premature infants included: (1) Asymptomatic infants with Hct <21% and reticulocytes <2%; (2) infants with hematocrit <31% and hood $O_2 < 36\%$ or mean airway pressure <6 cm H_2O by continuous positive airway pressure (CPAP) or intermittent mandatory ventilation (IMV) or >9 apneic and bradycardic episodes per 12 h or 2/24 h requiring bag-and-mask ventilation while on adequate methylxanthine therapy or heart rate >18/min or respiratory rate >80/min sustained for 24 h or weight gain of <10 g/d for 4 days on 100 kcal/kg/d or having surgery; (3) infants with hematocrit <36% and requiring >35% O_2 or mean airway pressure of 6-8 cm H_2O by CPAP or IMV.

Results were presented as absolute frequencies and percentages for categorical variables. Categorical variables were compared using a Chi-square test or Fisher's exact test when more than 20% of cells with expected count of <5 were observed. Continuous variables were also compared using a *t*-test. Statistical significance was determined as a $P \geq 0.05$. All statistical analyses were performed using SPSS software (version 19.0, SPSS Inc., Chicago, Illinois).

RESULTS

A total of 60 VLBW infants were eligible for the current study that were allocated to the rhEpo group ($n = 30$) or to the placebo group ($n = 30$). Table 1 summarizes the baseline demographic data. Importantly, the rhEpo and placebo groups were comparable with regard to gestational age, birth weight, age at the time of study, length of stay in hospital and gender. 16 (53.3%) in the rhEpo group and 20 (66.7%) in the placebo group required blood transfusion ($P = 0.289$). The mean levels of hemoglobin, hematocrit and reticulocyte were also similar across the two groups at baseline. Monitoring of the parameters of hemoglobin, hematocrit and reticulocyte count showed no any relevant deviation from baseline to the 2nd week of the study [Table 2]. However, there was relevant difference for these parameters between infants who received rhEpo and the infants who received placebo during the 3rd week of the study. Changes in the levels of hemoglobin, hematocrit and reticulocyte were similar between the two groups during the first 2 weeks of evaluation, but were significantly different between groups after 2 weeks [Figure 1].

DISCUSSION

The increasing trend of blood transfusions in premature neonates prompted the design and implementation of a large number of trials to evaluate the effectiveness of

Table 1: Baseline characteristics in the intervention and control groups

Characteristics	$n=30$		P value
	Intervention group	Control group	
Gestational age (wk)	28.41±1.40	28.33±2.20	0.822
Birth weight (kg)	1.23±0.27	1.16±0.17	0.234
Age at the beginning (day)	13.40±5.71	10.96±5.50	0.097
Male gender	6 (20.0)	8 (26.7)	0.881
Transfusion requirement	16 (53.3)	20 (66.7)	0.289
Times of transfusion	1.90±0.38	2.20±1.20	0.197
Length of hospital stay (day)	31.5±13.2	31.9±14.9	0.913
Hemoglobin (gr/dl)	11.41±1.18	11.36±1.65	0.893
Hematocrit (%)	32.80±3.30	34.00±4.11	0.217
Reticulocyte (%)	2.11±1.50	1.99±0.61	0.501

Data are presented as mean±SD or number (%)

Table 2: Measures of laboratory parameters at the three study time points

Characteristics	*n* = 30		*P* value
	Intervention group	**Control group**	
Hemoglobin			
Week 1	10.70 ± v0.92	10.33 ± 1.22	0.153
Week 2	9.63 ± 1.38	9.57 ± 1.29	0.863
Week 3	10.90 ± 1.60	8.17 ± 0.37	<0.001
Hematocrit			
Week 1	31.36 ± 3.60	30.70 ± 3.85	0.533
Week 2	29.70 ± 3.65	29.25 ± 3.83	0.603
Week 3	32.61 ± 5.03	29.22 ± 3.55	0.004
Reticulocyte			
Week 1	1.62 ± 0.91	1.24 ± 1.00	0.129
Week 2	1.74 ± 1.18	1.44 ± 0.71	0.238
Week 3	3.50 ± 0.86	0.90 ± 0.59	<0.001

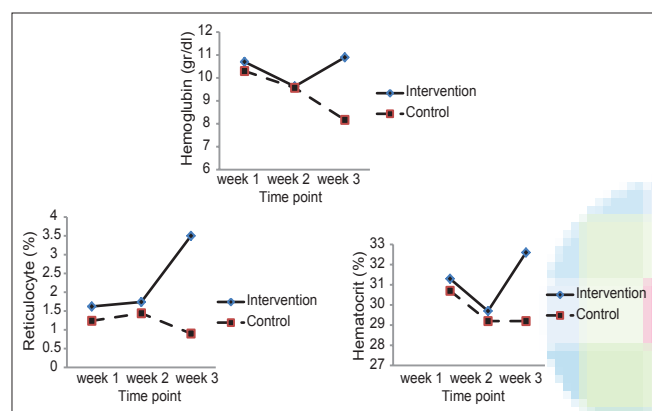Data are presented as mean ± SD or number (%)

Figure 1: Changes in hemoglobin, hematocrit and reticulocyte within weeks 1-3 in the two groups

rhEpo administration in the early neonatal period. These efforts are due to considerable falling hemoglobin after birth in VLBW infants. The number of red blood cells may decrease after birth due to the natural breakdown of erythrocytes. In this context, VLBW neonates are the main groups of patients requiring blood transfusions hence that about 60-100% of these infants receive multiple transfusions, mostly during the first 2 weeks of life.[13-16] However, because of some problems such as small sample size, few randomized studies and the use of different rhEpo doses and duration of administration, the results of the studies are very varied. In the present trial, we tried to assess the effectiveness of early rhEpo administration and whether this intervention improves hematopoietic indices in VLBW neonates. Our study confirmed the effectiveness of rhEpo on improving blood indicators of hemoglobin, hematocrit and reticulocyte count in premature infants without any observed side-effects. This improvement was achieved within 3 weeks of drug administration. On the other hand, the trend of the changes in the indicators was

similar between the intervention and trail groups within the first 2 weeks of the study. This result is similar to results in some previous studies.[17-20]

Furthermore, in our study, despite stimulated erythropoiesis following rhEpo administration, the number of transfusions was not changed with rhEpo therapy. This result is similar to results from some previous studies.[21-23] Although the studies are in agreement with the beneficial effects of the early use of rhEpo on erythropoiesis stimulation, it's the effects on transfusion requirements remain controversial. In a study by Carnielli *et al*.[24] rhEpo, especially in combination with iron supplementation was effective in reducing the need for blood transfusions in the premature infant, however contrarily; Donato *et al*.[15] revealed that the early administration of rhuEpo induced a rise in reticulocyte counts, but not enough to reduce the transfusion requirement. In fact, the length of rhEpo treatment and doses varied among the studies reviewed and no agreement exists for this consideration. We proposed to limit rhEpo treatment of VLBW neonates to a 3 week course and reported a difference by the 3rd week of treatment.

In our study, no serious side-effects were observed in both rhEpo and placebo groups. In reviewing the literature, few side-effects have been reported. These include neutropenia or thrombocytosis.[25] More rare side-effects include high blood pressure, flu symptoms, joint and bone pain, tremors/chills, injection site inflammation (resolves after a few days) and headaches.[26] However, it seems that the reported side-effects in adults can be associated with the dose and duration of drug administration.

CONCLUSION

Our study suggested that rhEpo is an effective drug for improving levels of erythropoiesis parameters within 3 weeks of its administration in VLBW infants regardless its effect on the need for blood transfusions.

ACKNOWLEDGMENTS

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