Impact of Erythropoiesis-Stimulating Agents on Behavioral Measures in Children Born Preterm

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Objective To evaluate the impact of erythropoiesis-stimulating agents (ESAs) administered during initial hospitalization and family demographic factors on behavior at 3.5-4 years of age.

Study design Children were enrolled who had previously participated in a randomized study of ESAs (n = 35) or placebo (n = 14) in infants born preterm with birth weights of 500-1250 g. A term healthy control group (n = 22) also was recruited. Behavior was evaluated by parent report with the Behavioral Assessment System of Children-2. Principal component analyses identified 2 demographic factors, a Socioeconomic Composite (SEC) and a Family Stress Composite. A multivariate general linear model evaluated the impact of study group and sex on the 4 composite scales of the Behavioral Assessment System of Children-2. Demographic factors were treated as covariates and interactions with study group (ESA, placebo, and term) were examined.

Results The ESA group had significantly better scores than the placebo group on behavioral symptoms ($P = .04$) and externalizing scales ($P = .04$). An interaction was observed between study group and SEC ($P = .001$). A beneficial effect of ESAs was maximal in the children with lower SEC scores.

Conclusions The beneficial effects of ESAs on childhood behavior were maximal in children with lower SEC scores. ESAs seemed to ameliorate the adverse impact of lower SEC on behavioral domains seen in the placebo group. This effect was independent of the beneficial effect of ESAs on global cognition we reported previously. *(J Pediatr 2017;184:75-80).*

Trial registration ClinicalTrials.gov: NCT01207778 and NCT00334737.

Infants born preterm are at risk for a variety of adverse outcomes, including motor, cognitive, and behavioral problems.[1] In a national cohort study from Finland, parents of 5-year-old children born with very low birth weight reported greater rates of behavioral difficulties including internalizing behavior (ie, anxiety, depression, withdrawal) and externalizing behavior (ie, conduct problems, aggression, hyperactivity) compared with children born term.[2] Behavioral problems in children with very low birth weight have been shown to persist through early childhood and adolescence and are not lessened by higher intelligence scores.[3] Several studies have shown that behavior problems in early childhood often are accompanied by cognitive, executive function, and motor difficulties, raising concerns about long-term outcomes.[4,5]

Erythropoiesis-stimulating agents (ESAs) such as erythropoietin (Epo) or darbepoetin (Darbe) have been used to increase red cell production and decrease transfusions in infants born preterm. Recent studies in animals and humans in which investigators evaluated the nonhematopoietic effects of ESAs suggest a neuroprotective potential through mechanisms of increased oligodendrogenesis, decreased inflammation, decreased oxidative injury, and decreased apoptosis.[6] Our group reported previously that the administration of ESAs (Epo or Darbe) shortly after birth significantly improved cognition and object permanence scores and decreased neurodevelopmental impairments at 2 years of age[7] in a randomized trial of children with birth weights of 500-1250 g.[8] These findings persisted at 3.5-4 years of age.[9]

The purpose of this study was to describe the effects of ESAs on behavioral and emotional functioning at 3.5-4 years of age. Because socioeconomic status has diverse and prominent effects on brain development,[10] the role of socioeconomic and family factors were investigated as possible modifiers of ESA effects.

Darbe	Darbepoetin alfa
Epo	Erythropoietin
ESA	Erythropoiesis-stimulating agents
BASC-2	Behavioral Assessment System of Children – Second Edition
SEC	Socioeconomic Composite

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Methods

Former infants born preterm (500-1250 g birth weight), enrolled at ≤48 hours of age in the original study[7] were eligible for the current BRITE study (Brain Imaging and Developmental Follow-up of Infants Treated with Erythropoietin; Clinicaltrials.gov: NCT01207778). Infants with genetic disorders, significant congenital anomalies (including known neurologic anomalies), hypertension, seizures, thrombosis, hemolytic disease, or who were already receiving Epo were ineligible for the original study. The number of children eligible for the study and evaluated at 3.5-4 years of age is shown in shown in **Figure 1** (available at www.jpeds.com).

A total of 49 former children born preterm from the original randomized controlled trial were recruited successfully. A group of children born at full term (n = 24) who had an uneventful newborn course also were recruited at age 3.5-4 years at the New Mexico site for the BRITE study. Questionnaires were missing from 2 of the term group and 4 of the preterm group; those subjects and were omitted from this analysis. The parents and examiners continued to be masked to the treatment assignment of the children. The study was approved by the institutional review boards at the University of Utah and the University of New Mexico, and informed consent was obtained from parents. Demographic and medical history data were obtained from the family.

Initial Study Procedures

Randomization of the participants born preterm to ESA vs placebo groups originally was performed during their initial hospitalization with the use of a computer-generated permuted block method, stratified by center. Twins were assigned to the same treatment group. Caregivers and investigators (except the research pharmacists and coordinators administering the study medicine) were masked to the treatment assignment. An investigational new drug application was approved by the Federal Drug Administration (IND #100138), and the study was registered at ClinicalTrials.gov (NCT00334737).

Dosing of Study Drug and Supplements

Infants were randomized to 1 of 3 groups: Epo, 400 units/kg, given subcutaneously 3 times a week; Darbe, 10 μg/kg, given subcutaneously once a week, with sham dosing 2 other times per week; or placebo, consisting of 3 sham doses per week. Dosing continued until 35 weeks of postmenstrual age, discharge, transfer to another hospital, or death. All infants received supplemental parenteral and enteral iron, folate, and vitamin E. All infants received study drug dosing through 35 weeks of postmenstrual age, for an average dosing length of 7.2 ± 1.8 weeks. After planned analyses showed no significant differences between Epo and Darbe recipients, they were combined as a single ESA group.

Present Study

The Behavioral Assessment of Children Scale–Second Edition (BASC-2) is a norm-referenced parent rating scale that assesses childhood behavioral and emotional issues.[11] The BASC-2 consists of 4 composite scales: adaptive skills (social skills, activities of daily living), behavioral symptoms (withdrawal or atypical behaviors), externalizing problems (hyperactivity, aggression, conduct problems), and internalizing problems (anxiety, depression, somatization). For the adaptive skills composite, greater scores are better; for other scales, greater scores are indicative of problems. Standardized t scores were used for analyses.

The Wechsler Preschool and Primary Scale of Intelligence – Third Edition, a widely used standardized scale of general cognitive abilities, was administered to each child.[12] Socioeconomic and demographic variables were obtained through maternal interviews, including maternal age and education, income level, the number of times the family moved since the index child's birth, the number of children in the household <6 years of age, and the primary language spoken in the home.

Data Analyses

The hypothesis of beneficial ESA treatment effects across all BASC-2 scales was tested with a multivariate general linear model with covariates (MANCOVA). With respect to the 4 dependent variables (BASC-2 scores) in the MANCOVA, preliminary analyses revealed that 3 of the 4 variables were not distributed normally, according to the Shapiro-Wilks test (all except the adaptive skills scale). A log transformation was performed for each BASC-2 scale; all transformed values correlated with the original t scores at r = 0.99.

Effect sizes from the MANCOVA are presented as partial eta-squared values with statistical significance of $P < .05$ for the Wilks' lambda multivariate tests. Fixed factors were study group and sex. Given the importance of demographic factors for BASC-2 scales,[13] these influences were considered systematically in the statistical model. Rather than including each of the 7 specific demographic variables in the model, which would reduce statistical power and complicate interpretation, these variables were reduced to 2 composites via the use of principal components analysis with direct oblimin rotation. Ethnicity was coded as either "Hispanic" or "non-Hispanic." Two factors emerged with eigenvalues >1, and these captured 37% and 23% of the shared variance (**Table I**, available at www.jpeds.com, provides details on factor loadings). For summary purposes, the first of these factors was termed "Socioeconomic Composite" (SEC) and the second "Family Stress Composite." Greater scores on the SEC indicated relatively greater income and education, and greater scores on the Family Stress Composite indicated more family moves, more children in the home, and younger maternal age. Preliminary analyses revealed that the second demographic factor was not related to any BASC-2 scale and did not interact with other factors, so this variable was dropped from further analysis.

In the MANCOVA, sex and study group (placebo, ESA, term) were fixed factors and SEC was a covariate. Interactions of sex with study group and SEC with study group were included because of concern that effects of ESA might be moderated by these factors. Planned comparisons contrasted the placebo and ESA groups (to reveal the effects of ESAs) and the ESA and term groups (to reveal whether ESA treatment improved scores