Prophylactic Early Erythropoietin for Neuroprotection in Preterm Infants: A Meta-analysis

Hendrik S. Fischer, MD, Nora J. Reibel, MSc, Christoph Bührer, MD, Christof Dame, MD

CONTEXT: Recombinant human erythropoietin (rhEPO) is a promising pharmacological agent for neuroprotection in neonates.

OBJECTIVE: To investigate whether prophylactic rhEPO administration in very preterm infants improves neurodevelopmental outcomes in a meta-analysis of randomized controlled trials (RCTs).

DATA SOURCES: Medline, Embase, and the Cochrane Central Register of Controlled Trials were searched in December 2016 and complemented by other sources.

STUDY SELECTION: RCTs investigating the use of rhEPO in preterm infants versus a control group were selected if they were published in a peer-reviewed journal and reported neurodevelopmental outcomes at 18 to 24 months’ corrected age.

DATA EXTRACTION: Data extraction and analysis followed the standard methods of the Cochrane Neonatal Review Group. The primary outcome was the number of infants with a Mental Developmental Index (MDI) <70 on the Bayley Scales of Infant Development. Secondary outcomes included a Psychomotor Development Index <70, cerebral palsy, visual impairment, and hearing impairment.

RESULTS: Four RCTs, comprising 1133 infants, were included in the meta-analysis. Prophylactic rhEPO administration reduced the incidence of children with an MDI <70, with an odds ratio (95% confidence interval) of 0.51 (0.31–0.81), \( P < .005 \). The number needed to treat was 14. There was no statistically significant effect on any secondary outcome.

CONCLUSIONS: Prophylactic rhEPO improved the cognitive development of very preterm infants, as assessed by the MDI at a corrected age of 18 to 24 months, without affecting other neurodevelopmental outcomes. Current and future RCTs should investigate optimal dosing and timing of prophylactic rhEPO and plan for long-term neurodevelopmental follow-up.
Improving neurodevelopmental outcomes is a major goal in neonatology, especially with regard to the increasing survival rate of the most immature infants.1-4 In particular, extremely low birth weight (ELBW) infants may develop cognitive delay or cerebral palsy later in life, even if the cranial ultrasound examination at discharge is normal.4 Recombinant human erythropoietin (rhEPO) is 1 of the most promising pharmacological substances for neuroprotection in newborn infants and may be beneficial for regeneration and repair after neonatal brain injury.5,6 Interestingly, the endogenous erythropoietin receptor system activates in response to hypoxic–ischemic injury. This means that rhEPO may be particularly potent for neuroprotection and neuronal regeneration if administered timely after an acute brain injury.7-9 Conversely, if rhEPO is administered prophylactically, higher doses may be needed to cross the blood–brain barrier.10,11 Numerous in vitro and in vivo experiments (including models of the fetal or immature brain) have shown that rhEPO has antioxidant, antixcitotoxic, and anti-inflammatory properties, prevents neuronal apoptosis,12 and promotes neurogenesis and angiogenesis.7,13-15 Debate continues over whether premature infants at high risk of neurodevelopmental impairment may benefit from prophylactic rhEPO.16 Most clinical follow-up studies on preterm infants primarily treated with rhEPO (or its higher-glycosylated derivate darbepoetin) for anemia of prematurity indicated improved neurodevelopmental outcomes,17-21 whereas only a few studies have shown no effect.22,23 In 2005, the first phase I and II trials were initiated to establish the safety of early high-dose rhEPO for neuroprotection.24,25 So far, only a limited number of randomized controlled trials (RCTs) have reported data on the neuroprotective effects of rhEPO in preterm infants. This limitation was reflected in 2 previous meta-analyses26,27 that included several quasi-RCTs25,28-30 and a cohort study.19 Recently, neurodevelopmental outcomes at a corrected age of 18 to 24 months from 2 prospective RCTs, intended primarily to evaluate the neuroprotective effects of rhEPO in a total of 978 very preterm infants, were published almost simultaneously.31,32 Therefore, the present meta-analysis aimed to study whether prophylactic rhEPO improves the neurodevelopmental outcomes of very preterm infants at 18 to 24 months’ corrected age, solely including data from previous and recent RCTs.

METHODS

Criteria for Considering Studies for This Review

All RCTs that investigated the effect of prophylactic rhEPO in preterm infants were eligible for the meta-analysis. Included trials had to compare an rhEPO-treated group with a control group that received no treatment or placebo, and they had to report neurodevelopmental outcomes. Only studies published in full in peer-reviewed journals were accepted. No language restrictions were applied. Very key... you should always look for this Types of Outcome Measures

Outcome measures were specified beforehand in a review protocol, considering the outcomes reported by recently published RCTs.31,32

The primary outcome of the meta-analysis was the number of infants with a Mental Developmental Index (MDI) <70 on the Bayley Scales of Infant Development, Second Edition (BSID-II), at 18 to 24 months’ corrected age.33 If infants were assessed according to the Third Edition (BSID-III),24 we used cognitive scores <85 as the primary outcome, because a recent study showed that a cognitive score <85 (BSID-III) predicts an MDI <70 (BSID-II), with an overall agreement of 97.3%.35 A planned subgroup analysis assessed the incidence of an MDI <70 (BSID-II) or cognitive score <85 (BSID-III) in infants <28 weeks’ gestational age. Other predetermined secondary outcomes were the number of infants with a Psychomotor Developmental Index (PDI) <70 (BSID-II), cerebral palsy, severe visual impairment, and severe hearing impairment, respectively. After the literature search, we added the secondary outcome “any neurodevelopmental impairment at 18 to 24 months’ corrected age.”

Search Methods for Identification of Studies

For the literature search, we followed the standard search methods of the Cochrane Collaboration.36 Two authors independently searched the databases Medline, Embase, and the Cochrane Central Register of Controlled Trials (CENTRAL). The results were combined with cross-referencing of previous reviews and trials and with the use of expert information. Additional information about ongoing trials was sought through the search portal of the International Clinical Trials Registry Platform.37 Medline, Embase, and CENTRAL were searched on May 31, 2016. The search was updated on December 18, 2016. A highly sensitive search strategy was applied, as outlined in the Cochrane Handbook for Systematic Reviews of Interventions, and complied with specific recommendations for identifying RCTs in Medline and Embase.36,38 A variety of search terms were applied to identify RCTs concerned with rhEPO use in premature infants (search concept: premature infant AND erythropoietin AND randomized controlled trial) or with follow-up studies on children of all ages that reported neurodevelopmental outcomes.
After rhEPO treatment (search concept: child AND erythropoietin
AND [RCT OR follow-up study]
AND [neurodevelopmental testing
OR neurodevelopmental outcomes
OR neuroprotection]). Altogether,
we used >60 free-text and indexed
search terms, including many from
the Medical Subject Headings (MeSH)
and Excerpta Medica Tree indexing
systems. The detailed search strategy
is available in the Supplemental
Information. We followed the
Preferred Reporting Items for
Systematic Reviews and Meta-
analyses (PRISMA) Statement.39,40

For methodological reasons, however, the latter was not
included in the meta-analysis.17 Another
RCT targeted both hematologic and
neurodevelopmental outcomes,
comparing rhEPO, placebo, and a
third intervention group receiving
darbepeotin.20 For methodological
reasons, however, the latter was not
included in the meta-analysis.

To obtain additional data or
information, we contacted the first
or senior authors of all included RCTs.
We highly appreciate the cooperation
of another study already included

Ohls: outcome assessed too late

Song '16

Natalucci 2016

Ohls '14

4

3

Ohls '04

42,43

A third RCT
(NCT01207778), recently published
by Ohls et al,21 was excluded because
it reported neurodevelopmental
outcomes at 3.5 to 4 years.

INCLUDED

EXCLUDED

Included Studies

Four RCTs, which assessed a total of
1133 patients at 18 to 24 months'
corrected age, were included in the
meta-analysis; their main
characteristics are summarized
in Table 1. All studies included
preterm infants of <32 0/7 weeks’
gestational age, but only 2 used the
infants’ birth weight to define the
inclusion criteria.17,20 Notably, the
timing, dosing, and duration of rhEPO
administration varied. Whereas 1
RCT applied early high-dose rhEPO
only (first dose within 3 hours after
birth, cumulative dose of 9000 IU/kg
within the first 42 hours),32 the other
3 RCTs started rhEPO later (within
the first 48–96 hours), applied lower
doses (1200–1750 IU/kg per week),
and prescribed sustained treatment
of several weeks.17,20,31 One RCT was
designed primarily to prevent red
blood cell transfusions by reducing
the anemia of prematurity.17 Another
RCT already combined rhEPO
concentrations with
neurodevelopmental outcomes at
18 to 22 months’ corrected age in
a single-center follow-up study on
an RCT of rhEPO versus placebo,
but the study was excluded because
the population was a subset of the
Eunice Kennedy Shriver National
Institute of Child Health and
Human Development Neonatal
Research Network Trial included
in our meta-analysis.17
of 1 author who provided us with previously unpublished data for infants <28 0/7 weeks’ gestational age.32

**Risk of Bias in the Included Studies**

**Selection Bias**

Random sequence generation and allocation concealment were adequate in all studies (low risk).

**Performance Bias**

In 3 RCTs, considerable efforts were made to blind parents and personnel to the study intervention (low risk).17,20,32 By contrast, doctors and nurses responsible for the treatment were not blinded in the trial by Song et al31 (high risk).

**Detection Bias**

Investigators performing long-term outcome assessments were blinded to the patients’ group allocation in all studies (low risk).

**Attrition Bias**

The proportion of the surviving study patients who completed follow-up varied from 71% to 91%, according to the particular study and group, and the maximum difference in follow-up between intervention and control group was 91% versus 80% in 1 study20 (unclear risk).

**Reporting Bias**

Two RCTs were registered at ClinicalTrials.gov before or shortly after recruitment began (low risk),20,32 and 1 RCT was not registered (unclear risk).17 The study by Song et al31 was registered 6 months after enrollment was completed, and the registered inclusion criteria and primary outcomes differed from those reported in the publication (high risk).

**Other Bias**

Two RCTs were co-funded by pharmaceutical companies (low risk).17,32 Visual inspection of the funnel plots did not reveal significant asymmetries in the distribution of effect sizes around the mean, making publication bias less likely. The funnel plots, detailed characteristics of the included studies, and the complete risk of bias assessment are available as Supplemental Information (Supplemental Tables, Supplemental Fig 4).

![FIGURE 1](https://example.com/fig1.png)

**Effects of Intervention**

The effects of rhEPO on MDI <70 and PDI <70 are shown in Fig 2. Prophylactic rhEPO in very preterm infants significantly reduced the incidence of an MDI <70 at 18 to 24 months’ corrected age, with an OR (95% CI) of 0.51 (0.31–0.81), P = .005. Statistical measures did not indicate a heterogeneity problem (χ² = 4.01, P = .26, I² = 25%). The absolute risk of an MDI <70 was reduced from 15.7% to 8.4%, corresponding to an NNT of 14. In a sensitivity analysis excluding the data of the largest trial by Song et al31 the
calculated effect size of rhEPO on MDI <70 remained in favor of rhEPO but was not longer statistically significant, with an OR (95% CI) of 0.68 (0.4–1.18), P = .17 (data not shown). RhEPO had no significant impact on MDI <70 in infants <28 weeks’ gestational age (Fig 2B) or on PDI <70 (Fig 2C). The definitions of the other secondary outcomes (cerebral palsy, severe visual impairment, severe hearing impairment, and any neurodevelopmental impairment) all showed some variation between the included studies (see Supplemental Tables for details). As shown in Fig 3, there was no significant effect of rhEPO on any of these outcomes in the meta-analysis. The combined OR (95% CI) of any neurodevelopmental impairment was 0.55 (0.28–1.08), P = .08, with substantial statistical heterogeneity between the included studies (χ² = 10.38, P = .02, I² = 71%).

don’t ever write ‘first’ or ‘largest’ in one of your manuscripts. Hubris. Let an editorial say it.

**DISCUSSION**

This is the largest meta-analysis to date to investigate the neuroprotective effects of prophylactic rhEPO in very preterm infants of 18 to 24 months’ corrected age and the first to include only RCTs. Our analysis is highly topical, because 2 of the 4 RCTs, which included 978 of the 1133 patients studied, had only been reported recently.31,32

The meta-analyzed data showed that rhEPO reduced the number of patients with an MDI <70 at 18 to 24 months’ corrected age but had no significant effect on motor development, hearing, or vision.

**Quality of Evidence**

The included RCTs were mostly of high methodological quality, with a low risk of bias (Supplemental Tables). The only major problems related to the RCT by Song et al31 were the lack of blinding of personnel and indications of selective reporting. Incomplete outcome data were considered as a minor point in all studies. However, the reported follow-up rates at 18 to 24 months’ corrected age were satisfactory from a practical point of view. In accordance with Cochrane methods, we used funnel plots to assess for publication bias,36 but we did not identify major asymmetries (Supplemental Fig 4). However, we were aware that this method had low power to detect bias, because only 4 RCTs were included in the meta-analysis. More importantly, there remains a possibility of unpublished data, in that 86 RCTs of rhEPO were excluded from the meta-analysis because they did not report neurodevelopmental outcomes (Fig 1).

**Effect on Cognitive Outcome**

The beneficial effect of prophylactic rhEPO on the incidence of MDI <70 at 18 to 24 months’ corrected age was consistent across all included RCTs (Fig 2A), although timing and dosing of rhEPO administration varied between trials. With an absolute risk reduction from 15.7% to 8.4% and an NNT of 14 patients, the estimated effect is clinically relevant. The observed benefits of rhEPO are robust, because they are in agreement with available evidence from longer-term follow-up studies, which also reported improved cognitive outcomes. In the second follow-up on the previously mentioned RCT of rhEPO, darbepoetin, or placebo, Ohls et al21,47 assessed 53 children via the Wechsler Preschool and Primary Scale of Intelligence, Third Edition at the age of 3.5 to 4 years. The study compared the pooled data of the rhEPO group (n = 24) and darbepoetin group (n = 15) with the placebo group (n = 14) and showed that the children treated by erythropoiesis-stimulating agents scored significantly better for full-scale IQ, performance IQ, overall executive function, and working memory. When cognitive function was analyzed, results at 18 to 22 months and 4 years correlated significantly between BSID-II composite score and full-scale IQ.20,21 Neubauer et al19 analyzed neurodevelopmental outcomes at the age of 10 to 13 years in a cohort study of 89 ELBW infants who received rhEPO from the first week of life to near-term and 57 untreated ELBW infants. rhEPO-treated infants attained higher verbal, nonverbal, and composite

**TABLE 1 Characteristics of Included Studies**

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>N</th>
<th>Gestational Age, Birth Wt</th>
<th>Time Point of Intervention</th>
<th>Intervention</th>
<th>Recruitment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ohls et al37</td>
<td>2004</td>
<td>102</td>
<td>≤32 0/7, ≤1000 g</td>
<td>24–86 h of age</td>
<td>rhEPO 400 IU/kg IV or SC, 3 times per wk until 35/7 wk gestation</td>
<td>1997–1998</td>
</tr>
<tr>
<td>Ohls et al32</td>
<td>2014</td>
<td>53a</td>
<td>Any GA, 500–1250 g</td>
<td>≤48 h of age</td>
<td>rhEPO 400 IU/kg SC, 3 times per wk until 55/7/wk gestation</td>
<td>2006–2010</td>
</tr>
<tr>
<td>Natalucci et al32</td>
<td>2016</td>
<td>365</td>
<td>26 0/7–31 6/7, any BW</td>
<td>&lt;3 h of age</td>
<td>rhEPO 3000 IU/kg IV at &lt;3 h, 12–18 h and 36–42 h of age</td>
<td>2005–2012</td>
</tr>
<tr>
<td>Song et al31</td>
<td>2018</td>
<td>613</td>
<td>≤32 0/7, any BW</td>
<td>&lt;72 h of age</td>
<td>rhEPO 500 IU/kg IV every other day for 2 wk</td>
<td>2009–2013</td>
</tr>
</tbody>
</table>

BW, birth wt; GA, gestational age; IV, intravenously; SC, subcutaneously.

a This study had 3 groups: rhEPO (n = 29) versus placebo (n = 24) versus darbepoetin (n = 27). The darbepoetin group was not included in the meta-analysis.

b Median (interquartile range) GA of included infants 28–29 wk; study entry criteria: preterm infants with a birth wt of 500–1250 g.

One of these studies was judged the riskiest in terms of BIAS. Which one was it? Text your answer plus your favorite Justin Bieber, Taylor Swift, or One Direction song to Dr. McDonnell (440) 258-6938 ASAP before he separates from the US Navy.

Downloaded from by guest on May 15, 2017
IQ scores in the Hamburg–Wechsler Intelligence Test for Children III, and a subgroup analysis showed a particular benefit of rhEPO in infants with intraventricular hemorrhage. Because infants <28 0/7 weeks’ gestational age are at particularly high risk of poor developmental outcomes, the effects of rhEPO in this subpopulation deserve closer examination. In our planned subgroup analysis, there was no significant benefit of rhEPO on the outcome measure MDI <70 in such infants (Fig 2B). However, only stratified data from the recent Swiss and Chinese rhEPO neuroprotection RCTs were available for the subgroup analysis, and the resulting subset of 60 rhEPO-treated and 57 placebo-treated infants <28 0/7 weeks’ gestational age was less than the optimal information size. We are aware that a multicenter RCT in the United States is currently recruiting 941 preterm infants <28/0/7 weeks’ gestational age to answer this question (PENUT Trial, NCT01378273). The PENUT Trial enrolls infants within 24 hours after birth to investigate an early high-dose strategy of 6 doses of intravenous Epo 1000 U/kg per dose at 48-hour intervals, followed by subcutaneous Epo 400 U/kg per dose 3 times per week up to 32 6/7 weeks’ postmenstrual age. Results are expected in 2019 and will be

### FIGURE 2
Effects of rhEPO on neurodevelopment, as assessed by BSID-II or BSID-III at 18 to 24 months’ corrected age. Forest plots show the effects on the number of infants with an MDI <70 (BSID-II) or cognitive score <85 (BSID-III) in all infants (primary outcome, A), in infants <28 0/7 weeks’ gestational age (planned subgroup analysis, B), and on the number of infants with a PDI <70 as assessed by BSID-II (secondary outcome, C). M–H, Mantel–Haenszel.

#### A MDI <70

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>rhEPO</th>
<th>Placebo</th>
<th>Odds Ratio</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Events</td>
<td>Events</td>
<td>M–H, Random, 95% CI</td>
<td>M–H, Random, 95% CI</td>
</tr>
<tr>
<td>Ohis 2004</td>
<td>14</td>
<td>16</td>
<td>0.82 (0.34–1.97)</td>
<td></td>
</tr>
<tr>
<td>Ohis 2014</td>
<td>3</td>
<td>6</td>
<td>0.35 (0.08–1.57)</td>
<td></td>
</tr>
<tr>
<td>Natalucci 2016</td>
<td>12</td>
<td>15</td>
<td>0.71 (0.32–1.56)</td>
<td></td>
</tr>
<tr>
<td>Song 2016</td>
<td>19</td>
<td>49</td>
<td>0.34 (0.20–0.59)</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>48</td>
<td>86</td>
<td>0.51 (0.31–0.81)</td>
<td>1.00</td>
</tr>
</tbody>
</table>

Heterogeneity: $\chi^2 = 0.06; \chi^2 = 4.01; df = 3 (P = .26); P = .25%$

Test for overall effect: $Z = 2.80 (P = .005)$

#### B MDI <70 in infants <28 weeks’ gestational age

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>rhEPO</th>
<th>Placebo</th>
<th>Odds Ratio</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Events</td>
<td>Events</td>
<td>M–H, Random, 95% CI</td>
<td>M–H, Random, 95% CI</td>
</tr>
<tr>
<td>Ohis 2004</td>
<td>No stratified data available</td>
<td>Not estimable</td>
<td></td>
<td>1.00</td>
</tr>
<tr>
<td>Ohis 2014</td>
<td>No stratified data available</td>
<td>Not estimable</td>
<td>0.21 (0.02–1.98)</td>
<td></td>
</tr>
<tr>
<td>Natalucci 2016</td>
<td>1</td>
<td>4</td>
<td>0.58 (0.12–2.95)</td>
<td></td>
</tr>
<tr>
<td>Song 2016</td>
<td>4</td>
<td>12</td>
<td>0.41 (0.11–1.53)</td>
<td>1.00</td>
</tr>
<tr>
<td>Total</td>
<td>5</td>
<td>60</td>
<td>0.00</td>
<td>1.00</td>
</tr>
</tbody>
</table>

Heterogeneity: $\chi^2 = 0.00; \chi^2 = 0.52, df = 1 (P = .47); P = .00%$

Test for overall effect: $Z = 1.33 (P = .18)$

#### C PDI <70

<table>
<thead>
<tr>
<th>Study or Subgroup</th>
<th>rhEPO</th>
<th>Placebo</th>
<th>Odds Ratio</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Events</td>
<td>Events</td>
<td>M–H, Random, 95% CI</td>
<td>M–H, Random, 95% CI</td>
</tr>
<tr>
<td>Ohis 2004</td>
<td>14</td>
<td>6</td>
<td>2.04 (1.01–8.53)</td>
<td></td>
</tr>
<tr>
<td>Ohis 2014</td>
<td>No data (BSID III used)</td>
<td>Not estimable</td>
<td></td>
<td>1.00</td>
</tr>
<tr>
<td>Natalucci 2016</td>
<td>21</td>
<td>17</td>
<td>1.14 (0.58–2.24)</td>
<td></td>
</tr>
<tr>
<td>Song 2016</td>
<td>No data available</td>
<td>Not estimable</td>
<td>1.67 (0.67–4.13)</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>35</td>
<td>236</td>
<td>0.00</td>
<td>1.00</td>
</tr>
</tbody>
</table>

Heterogeneity: $\chi^2 = 0.24; \chi^2 = 2.16, df = 1 (P = .14); P = .54%$

Test for overall effect: $Z = 1.10 (P = .27)$
FIGURE 3
Effects of rhEPO on secondary outcomes at 18 to 24 months’ corrected age. Forest plots show the effects on cerebral palsy (A), severe visual impairment (B), severe hearing impairment (C), and any neurodevelopmental impairment (D). M-H, Mantel–Haenszel.
stratified by gestational age at birth (<26 or 26–27 6/7 weeks).48

**Effect on Motor Outcomes, Vision, and Hearing**

In the meta-analysis, prophylactic rhEPO had no significant effect on the incidence of PDI <70, cerebral palsy, severe visual impairment, or severe hearing impairment at 18 to 24 months’ corrected age (Figs 2C and 3A–C). Admittedly, these adverse outcomes are rare, and the meta-analysis may not be sufficiently powered to detect minor effects. The latter also applies to longer-term follow-up studies that did not detect prophylactic rhEPO to have any impact on gross and fine motor activity, cerebral palsy, vision, or hearing.19,21 Reassuringly, the lack of effect on visual outcome in the meta-analysis is consistent with previous meta-analyses showing that early rhEPO did not increase the risk of retinopathy of prematurity (ROP)49,50 and with experimental data suggesting that rhEPO may even protect the neuronal cells of the retina.51 Most recently, a meta-analysis by Fang et al52 evaluated the influence of rhEPO (independent of early or late treatment initiation) on ROP and found no impact on the rates of any stage ROP, severe ROP, or ROP necessitating intervention.

**Effect on Any Neurodevelopmental Impairment**

Despite the robust beneficial effect of rhEPO on cognitive development, this meta-analysis showed only a trend toward lowering the combined outcome of any neurodevelopmental impairment (OR 0.55; 95% CI, 0.28–1.08; P = .08; Fig 3D). One possible explanation is that rhEPO targets only a subset of cells or certain areas of the brain responsible for cognitive development. Alternatively, the aforementioned discrepancy may result from the fact that data on the incidence of PDI <70 were not available for 2 RCTs.28,31 In the meta-analysis, this limitation may have biased the calculated effect of rhEPO on PDI <70 and on any neurodevelopmental impairment in favor of the placebo (Figs 2C and 3D). Notably, Song et al31 reported absolute PDIs, which were significantly higher in their rhEPO group compared with placebo (median PDI 99 vs 90; P < .001).

**Future Directions**

The results of the present meta-analysis clearly indicate that more clinical trials are warranted to investigate open questions about prophylactic rhEPO administration in preterm infants. Most importantly, major variations in the study protocols of the 4 RCTs included in the meta-analysis indicate that the optimal timing and dosing of rhEPO for neuroprotection in preterm infants are still unknown. It is therefore interesting that the follow-up on a previous phase I and II dose escalation trial of early high-dose rhEPO showed that rhEPO improved cognitive and motor outcome measures at 4 to 36 months’ corrected age.53 The protocol of the Swiss RCT (first dose within 3 hours after birth, cumulative dose of 9000 IU/kg within the first 42 hours)32 was the only RCT that closely followed experimental data, arguing for early high-dose rhEPO to achieve therapeutic levels in the brain within the most vulnerable early postnatal period.54,55 Because a subset of the study patients who were examined by cranial MRI scans at term-corrected age showed significantly lower white and gray matter injury scores after rhEPO treatment,56 it was disappointing that BSID-II scores at 18 to 24 months were not improved, neither in the subset nor in the entire study population.32 However, benefits in neurodevelopmental outcome may not be ascertainable until school age or even later.19,57,58 Looking at the encouraging outcome data of preterm rhEPO-treated infants in the retrospective study of Neubauer et al19 which extend the follow-up examinations from 2 to 13 years of age, the 2-year time point was the only one that showed no effects of rhEPO. Therefore, patients from the 4 analyzed RCTs need long-term neurodevelopmental follow-up examinations at 5 to 6 and 10 to 13 years of age, and additional funding should be provided for this purpose.

The fact that in the other 3 RCTs, repeated low-dose rhEPO treatment (started between 24 and 96 hours after birth, in doses of 1200–1750 IU/kg per week, sustained for 2 weeks51 or longer17,20) significantly improved neurologic outcomes provides an indication that continued treatment may be essential. So far, it is unclear whether this effect is caused by a direct impact of rhEPO on the premature brain or by stimulated erythropoiesis and reduced red blood cell transfusions. However, the question deserves more research, particularly considering pharmacodynamically optimized rhEPO treatment to prevent transfusions.59 To date, clinical data from meta-analyses and studies of early high-dose rhEPO indicate no increased risk for typical disorders in very preterm infants such as necrotizing enterocolitis, bronchopulmonary dysplasia, and other adverse outcomes.24,25,27,32,49,50,53 Two cohort studies even found a lower incidence of bronchopulmonary dysplasia in infants treated with rhEPO to prevent red blood cell transfusions.60,61 However, more skin hemangiomas were reported in 2 other cohort studies.25,62

Future RCTs of prophylactic rhEPO in preterm infants should investigate treatment protocols that combine an early high-dose strategy with continued treatment. The aforementioned PENVUT trial in the United States (NCT01378273)48...
Yay! In early 2019, you can re-do this MA using PENUT, the Chinese trial, and the EPO REPAIR study. Then you’ll have 7 studies in your Forest Plot.

and another ongoing trial in China (NCT02601872, NCT02550054) apply such strategies. Moreover, the EPO REPAIR trial in Switzerland currently investigates a similar dosing protocol for the rescue treatment of preterm infants with intraventricular hemorrhage (NCT02076373). However, the results of these studies will not be available before the end of 2018.

**Limitations**
The limitations of the available evidence are discussed above. Importantly, the RCT by Song et al had a substantial impact on the primary outcome but involved a high risk of bias in 2 domains. In addition, there were limitations at the meta-analysis level; as a primary outcome, we accepted a cognitive score <85 (BSID-III) as equivalent to an MDI <70 (BSID-II), as recommended by Johnson et al. However, authors of other studies found slightly different cutoff levels. With regard to the secondary outcomes, visual impairment, hearing impairment, and any neurodevelopmental impairment, we deliberately accepted minor deviations of the respective outcome definitions between the included studies (Supplemental Tables).

**CONCLUSIONS**
The current meta-analysis of 4 RCTs, including 574 rhEPO-treated and 547 placebo-treated premature infants, indicates that prophylactic rhEPO improves neurocognition, as assessed by the MDI at 18 to 24 months’ corrected age. By contrast, rhEPO had no significant effects on other neurodevelopmental outcomes. These findings demonstrate the promising potential of rhEPO for neuroprotection in very preterm infants. New data from current and future RCTs are needed to identify the optimal timing and dosing of prophylactic rhEPO administration in this age group. Ongoing RCTs and those already completed should be funded for long-term neurodevelopmental follow-up.

**ACKNOWLEDGMENTS**
We thank Dr Giancarlo Natalucci for providing previously unpublished data and additional information on his study. We appreciate the expert neuropediatric advice of Drs Elisabeth Walch and Gitta Reuner regarding the comparison of the different editions of the Bayley Scales of Infant Development. In addition, we thank Lukas Bomke, Judith Hollnagel, Christina Dame-Paasch, and Drs Makoto Kashiwabara, Paulina Aleksander, and Banu Orak for translating articles published in Bulgarian, Chinese, Italian, Japanese, Polish, and Turkish, respectively.

**ABBREVIATIONS**
BSID-II: Bayley Scales of Infant Development, Second Edition
BSID-III: Bayley Scales of Infant Development, Third Edition
CENTRAL: Cochrane Central Register of Controlled Trials
CI: confidence interval
ELBW: extremely low birth weight
MDI: Mental Developmental Index
MeSH: Medical Subject Headings
NNT: number needed to treat
OR: odds ratio
PDI: Psychomotor Development Index
PRISMA: Preferred Reporting Items for Systematic Reviews and Meta-analyses
RCT: randomized controlled trial
rhEPO: recombinant human erythropoietin
ROP: retinopathy of prematurity

**REFERENCES**


4. Johnson S, Moore T, Marlow N. Using the Bayley-III to assess neurodevelopmental delay: which...
cut-off should be used? Pediatr Res. 2014;75(5):670–674


49. Ohišson A, Aher SM. Early erythropoietin for preventing red cell transfusion in preterm and/or low birth weight infants. Cochrane Database Syst Rev. 2014;4:CD004863


57. Marlow N. Is survival and neurodevelopmental impairment at 2 years of age the gold standard outcome for neonatal studies? Arch Dis Child Fetal Neonatal Ed. 2015;100(1):F82–F84


### PRISMA 2009 Checklist

<table>
<thead>
<tr>
<th>Section/topic</th>
<th>#</th>
<th>Checklist item</th>
<th>Reported on page #</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>TITLE</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Title</td>
<td>1</td>
<td>Identify the report as a systematic review, meta-analysis, or both.</td>
<td></td>
</tr>
<tr>
<td><strong>ABSTRACT</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Structured summary</td>
<td>2</td>
<td>Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.</td>
<td></td>
</tr>
<tr>
<td><strong>INTRODUCTION</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rationale</td>
<td>3</td>
<td>Describe the rationale for the review in the context of what is already known.</td>
<td></td>
</tr>
<tr>
<td>Objectives</td>
<td>4</td>
<td>Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICO).</td>
<td></td>
</tr>
<tr>
<td><strong>METHODS</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Protocol and registration</td>
<td>5</td>
<td>Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.</td>
<td></td>
</tr>
<tr>
<td>Eligibility criteria</td>
<td>6</td>
<td>Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.</td>
<td></td>
</tr>
<tr>
<td>Information sources</td>
<td>7</td>
<td>Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.</td>
<td></td>
</tr>
<tr>
<td>Search</td>
<td>8</td>
<td>Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.</td>
<td></td>
</tr>
<tr>
<td>Study selection</td>
<td>9</td>
<td>State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).</td>
<td></td>
</tr>
<tr>
<td>Data collection process</td>
<td>10</td>
<td>Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.</td>
<td></td>
</tr>
<tr>
<td>Data items</td>
<td>11</td>
<td>List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.</td>
<td></td>
</tr>
<tr>
<td>Risk of bias in individual studies</td>
<td>12</td>
<td>Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.</td>
<td></td>
</tr>
<tr>
<td>Summary measures</td>
<td>13</td>
<td>State the principal summary measures (e.g., risk ratio, difference in means).</td>
<td></td>
</tr>
<tr>
<td>Synthesis of results</td>
<td>14</td>
<td>Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., I²) for each meta-analysis.</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Section/topic</th>
<th>#</th>
<th>Checklist item</th>
<th>Reported on page #</th>
</tr>
</thead>
<tbody>
<tr>
<td>Risk of bias across studies</td>
<td>15</td>
<td>Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).</td>
<td></td>
</tr>
<tr>
<td>Additional analyses</td>
<td>16</td>
<td>Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.</td>
<td></td>
</tr>
<tr>
<td><strong>RESULTS</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Study selection</td>
<td>17</td>
<td>Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.</td>
<td></td>
</tr>
<tr>
<td>Study characteristics</td>
<td>18</td>
<td>For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.</td>
<td></td>
</tr>
<tr>
<td>Risk of bias within studies</td>
<td>19</td>
<td>Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).</td>
<td></td>
</tr>
<tr>
<td>Results of individual studies</td>
<td>20</td>
<td>For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.</td>
<td></td>
</tr>
<tr>
<td>Synthesis of results</td>
<td>21</td>
<td>Present results of each meta-analysis done, including confidence intervals and measures of consistency.</td>
<td></td>
</tr>
<tr>
<td>Risk of bias across studies</td>
<td>22</td>
<td>Present results of any assessment of risk of bias across studies (see item 15).</td>
<td></td>
</tr>
<tr>
<td>Additional analysis</td>
<td>23</td>
<td>Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see item 16]).</td>
<td></td>
</tr>
<tr>
<td><strong>DISCUSSION</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Summary of evidence</td>
<td>24</td>
<td>Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).</td>
<td></td>
</tr>
<tr>
<td>Limitations</td>
<td>25</td>
<td>Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).</td>
<td></td>
</tr>
<tr>
<td>Conclusions</td>
<td>26</td>
<td>Provide a general interpretation of the results in the context of other evidence, and implications for future research.</td>
<td></td>
</tr>
<tr>
<td><strong>FUNDING</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Funding</td>
<td>27</td>
<td>Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.</td>
<td></td>
</tr>
</tbody>
</table>