Erythropoietin (Epo) Neuroprotection - Has its Time Come?

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Disclosures

• I have no financial relationships to disclose or conflicts of interest to resolve

• All studies were approved by the UW IACUC or IRB, as appropriate

• Use of Erythropoietin (Epo) for neuroprotection is off-label
Goals

• Review target neonatal populations that might benefit from Epo neuroprotection
• Brief review of basic research findings of Epo neuroprotection
• Preclinical Epo trials
• Update of clinical trials, ongoing and planned
Term Infants

- Acute Injury
- Typically Gray Matter
  - Perinatal asphyxia (3-5/1000 live births)
    - 23% neonatal deaths globally
  - Stroke

MCA stroke, 15 m

Basal ganglia

Lawn 2005, Black 2010
Neutrophils
Β2 integrin
Microglia / Macrophages
Astrocytes
Lymphocytes
NK cells
Mast cells

Immuno-inflammatory response after neonatal HI

Hagberg et al
Prematurity

• Births (USA)
  o ~ 4 million babies/year
  o ~ 500,000/year are preterm
  o ~ 50,000/year are ELBW
Preterm Brain Injury

- Principal causes
  - Interruption of normal development
  - Damage to existing tissues
    - Infection/Inflammation
    - Increases vulnerability to HI
  - Hemorrhage (IVH)
  - Hypoxia-ischemia

- White matter injury
  - PVL

- Gray matter injury
  - Smaller brain volumes
  - Deep gray matter

- Cerebellar injury

In: Epo and the nervous system
Outcomes for Infants <27 weeks

- Severe: 25%
- Moderate: 25%
- Mild or Normal: 50%

Neurodevelopmental Impairment

- Cerebral palsy
- Mental retardation
- Deafness
- Blindness

- School failure
- Autism
- ADHD
Neuroprotective Intervention

- Ideal treatment would be:
  - Safe
  - Readily available
  - Relatively inexpensive
  - Require no special equipment
  - Effective after injury
  - Protect neurons and glia (oligodendrocytes)
  - Reduce inflammation
  - Reduce apoptosis
  - Improve outcome
Erythropoietin (Epo)

- Produced in kidney/liver
- Regulates hematopoiesis
  - 55 kDa cytokine receptor
  - JAK/Stat 5 signaling
  - Blocks apoptosis of erythrocyte precursors
- Treatment for anemia
  - Cloned in 1985
Summary of Basic Research
## Non-hematopoietic Effects of Epo

<table>
<thead>
<tr>
<th>Growth Factor</th>
<th>Protective</th>
</tr>
</thead>
<tbody>
<tr>
<td>↑ Neurogenesis</td>
<td>↓ Apoptosis</td>
</tr>
<tr>
<td>↑ Glial cell proliferation</td>
<td>↓ Inflammation</td>
</tr>
<tr>
<td>↑ Angiogenesis</td>
<td>↓ Oxidative injury</td>
</tr>
<tr>
<td></td>
<td>↓ Nitric oxide toxicity</td>
</tr>
<tr>
<td></td>
<td>↓ Glutamate toxicity</td>
</tr>
</tbody>
</table>

Epo in CNS

- Receptors on neurons and glia
- Released by astrocytes
- Trophic effects
  - Astrocyte proliferation
  - Oligodendrocyte maturation
  - Blocks neuronal apoptosis
- Proposed to treat:
  - Spinal cord injury (Vitellaro-Zuccarello, 2007)
  - TBI (Grasso, 2007; Mammis, 2009)
  - Ischemic stroke (Ehrenreich, 2002)
  - Multiple sclerosis (Ehrenreich, 2007)
  - Perinatal asphyxia (Juul, 1997)
Epo Decreases Brain Injury

Gross Injury

Apoptosis

Gliosis

Conclude 5,000 U/kg x3 preferred dose

Epo Downregulates Inflammatory and Apoptotic Pathways

- Neonatal mice ± H-I brain injury ± Epo Tx
- Injury increased mRNA related to inflammation and apoptosis
- Epo treatment decreased expression of mRNA for these proteins

Epo Reduces Inflammatory Gene Expression

- Microarray analysis of hippocampus
- Gene expression “heat map”
- Inflammation-related gene set
- Red = more
- Blue = less
- Compare patterns
- Epo reduced the response to H-I

Epo Crosses the Blood Brain Barrier

Estimate that < 2% of systemic Epo crosses BBB

Summary of Results

• Epo:
  o Crosses the BBB into CSF and brain
  o Decreased inflammation and apoptosis
  o Protects dopaminergic neurons/decreases structural brain injury after HI
  o Reduces learning impairment due to brain injury
  o Neuroprotection is dose-dependent
  o Does not increase experimental ROP
  o Is safe in experimental models
Retrospective Studies
Higher cumulative doses of Epo and developmental outcomes in preterm infants

- **Retrospective cohort** of 82 infants born <1500g, ≤30 wk, evaluated at median age 25 mo

- Multivariate linear regression analyses: Are Epo and Bayley Scales PDI and MDI scores associated?

- Median 6–wk cumulative Epo dose was 3750 U/kg

- Higher 6 week Epo dose was associated with:
  - Higher MDI
  - No difference in PDI
Erythropoietin Improves Neurodevelopmental Outcome of Extremely Preterm Infants

• Retrospective review: ELBW infants treated with Epo
• 89 Epo–treated (10–13 years of age) vs. 57 untreated
• Results:
  o 55% Epo group assessed as normal vs. 39% untreated ($p<0.05$)
  o IQ score: 90.8 in Epo group vs. 81.3 in untreated ($p<0.005$)
  o Children with IVH: Epo–treated scored better than untreated children (52% vs 6% normally developed, composite HAWIK–III IQ score, 90.3 vs 67.0)
  o Children without IVH: no difference

Erythropoietin Concentrations and Neurodevelopmental Outcome in Preterm Infants
Ryann Bierer et al. *Pediatrics* 2006;118;e635-e640
Prospective Clinical Trials
60 ELBW infants born < 28 weeks and < 1000 grams
Evaluated safety and pharmacokinetics after Epo doses

Safety
- No harmful effects on:
  - ROP
  - Blood pressure
  - Clotting disorders
  - Hemorrhage
  - Kidney function
  - Liver function
  - Sepsis
  - Transfusions
  - Hospital stay

Human vs. Rat Pharmacokinetics

## Follow Up

<table>
<thead>
<tr>
<th></th>
<th>Epo</th>
<th>Control</th>
<th>Relative Risk</th>
<th>P</th>
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</thead>
<tbody>
<tr>
<td><strong>Low BSID Scores</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MDI ≤ 70</td>
<td>6% (1/17)</td>
<td>16% (3/18)</td>
<td>0.35</td>
<td>0.32</td>
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<tr>
<td>MDI ≤ 85</td>
<td>47% (8/17)</td>
<td>61% (11/18)</td>
<td>0.77</td>
<td>0.41</td>
</tr>
<tr>
<td><strong>Severity of NDI</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>24% (4/17)</td>
<td>21% (4/19)</td>
<td>1.12</td>
<td>0.86</td>
</tr>
<tr>
<td>NDI Moderate</td>
<td>47% (8/17)</td>
<td>32% (6/19)</td>
<td>1.49</td>
<td>0.35</td>
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<tr>
<td>NDI Severe</td>
<td>29% (5/17)</td>
<td>47% (9/19)</td>
<td>0.62</td>
<td>0.28</td>
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</table>

McAdams, Unpublished data
## Linear regression analysis of BSID scores

<table>
<thead>
<tr>
<th>Score</th>
<th>Factor</th>
<th>B</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>MDI</td>
<td>Epo</td>
<td>1.63</td>
<td>0.49</td>
</tr>
<tr>
<td></td>
<td>GA birth</td>
<td>0.59</td>
<td>0.54</td>
</tr>
<tr>
<td></td>
<td>IVH</td>
<td>-2.85</td>
<td>0.381</td>
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<tr>
<td></td>
<td>PMA</td>
<td>-0.05</td>
<td>0.711</td>
</tr>
<tr>
<td>PDI</td>
<td>Epo</td>
<td>7.84</td>
<td>0.022</td>
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<tr>
<td></td>
<td>GA birth</td>
<td>4.18</td>
<td>0.003</td>
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<tr>
<td></td>
<td>IVH</td>
<td>-7.59</td>
<td>0.088</td>
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<tr>
<td></td>
<td>PMA</td>
<td>0.13</td>
<td>0.498</td>
</tr>
<tr>
<td>Language</td>
<td>Epo</td>
<td>2.06</td>
<td>0.488</td>
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<tr>
<td></td>
<td>GA birth</td>
<td>0.24</td>
<td>0.824</td>
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<tr>
<td></td>
<td>IVH</td>
<td>-9.75</td>
<td>0.004</td>
</tr>
<tr>
<td></td>
<td>PMA</td>
<td>-0.15</td>
<td>0.322</td>
</tr>
</tbody>
</table>

The unstandardized regression coefficient B indicates the magnitude of the effect of each factor on the outcome score.
High-dose Epo Trial in VLBW Infants

• 45 VLBW Infants < 32 weeks < 1500 g
• Randomized double-masked trial
• 3x Epo (3000 U/kg) vs. placebo
• Safety: no differences reported for ...
  o ROP
  o Intracranial hemorrhage
  o Sepsis
  o NEC
  o BPD
  o Blood pressure
  o CBC indices: hemoglobin, leukocyte, platelets

Follow-up plan:
420 infants
2 years of age

Does Epo Improve Outcome In Very Preterm Infants?
Swiss Neonatal Network. Presented ESPR 2011

• 270 of 420 enrolled
  o Mean gestational age: 29 weeks
  o Mean birth weight: 1200 gm

• No difference in
  o Sepsis, BPD, ROP, IVH, adverse events or death

• Preliminary MRI Results
  o White matter injury scores were lower in Epo treated infants compared to controls
Phase I Trial of Neonatal Epo in Perinatal HIE

Methods: Epo dose-escalation open-label study, N=24
All subjects met criteria for moderate HIE and were cooled
• 250 (N=3),
• 500 (N=6),
• 1000 (N=7)
• 2500 U/kg/dose (N=8)
Infants received up to 6 doses of Epo IV QOD starting at <24h of age

Wu et al, unpublished data
Phase I Trial of Neonatal Epo in Perinatal HIE

Patients received an average of 4.7 doses
Mean length of hospital stay was 12 days

Safety
No deaths
No SAE
1st Clinical Trial of Epo for HIE 2009

- Epo (n=83) vs. conventional (n=84) treatment
- Epo 300 U/kg (n=52) or 500 U/kg (n=31), Q48h x7
- Epo improved neurologic signs at 7, 14, and 28 days as assessed by Thompson Score
- Epo reduced disability for moderate HIE
- Epo decreased the number of MDI scores below 70
- Epo reduced incidence of CP at 18 months
- Death or disability at 18 months was: 44% control vs. 25% Epo–treated ($p < 0.02$)
- No adverse effects of Epo.

Human Recombinant Erythropoietin in Asphyxia Neonatorum: Pilot Trial

• Prospective case–control study with 45 neonates in 3 groups:
  o Normal healthy group: N = 15
  o Mild/mod HIE: Epo 2500 U/kg, s.c. daily x 5 d, N=15
  o Mild/mod HIE: no Epo, N=15

• HIE groups had greater blood NO than normals (P<0.001)
  o NO ↓ in the HIE–Epo group vs. HIE–control group (P<0.001)

• EEG: 10 HIE–Epo vs 3 HIE–ctrl had normal backgrounds (P=0.01)
• MRI did not differ between groups
• HIE–Epo had fewer neurologic (P=0.03) and developmental abnormalities (P=0.03) at 6 months

Elmahdy et al. Pediatr 2010
Epo for Reduction of Perinatal Arterial Ischemic Stroke (PAS): A Feasibility and Safety Study

M Benders, L De Vries, M Roks, P Lemmers, N Van der Aa, F Groenendaal, I. Van Straaten, J. Smal. Van Bel

- 17 neonates with MRI-proven PAS
- 3 doses of Epo of 1000 IU/kg: immediately after diagnosis of PAS, 24 and 48 hrs after the first dose
- MRI/MRA at 1 wk and 3 months
- Safety parameters studied

- Results: HR, BP, Hct and coags were all in normal range, as were liver and renal functions
- No efficacy data yet
Epo Risks

• Risks
  o Hypertension
  o Clotting, Polycythemia
  o RBC aplasia
• Increased cardiovascular problems in pts with chronic renal failure
• Increased risk of DVT in adults undergoing spinal surgery
• Increased tumor growth in chemotherapy pts treated for anemia
• Increased risk of death (16% vs. 9%) in elderly patients treated for acute stroke (Ehrenreich, Stroke 2009;40 (12):e647–56)

• These risks have never been reported in infants
• So risks may be negligible when considering an acute regimen as a rescue for brain injury in infants
• For preterm infants the risk of ROP must be assessed
<table>
<thead>
<tr>
<th>Identifier</th>
<th>Status</th>
<th>Study</th>
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</thead>
<tbody>
<tr>
<td>NCT00719407</td>
<td>Enrollment complete</td>
<td>Neonatal Erythropoietin in Asphyxiated Term Newborns (NEAT) University of California, San Francisco, USA (Yvonne W Wu, MD, MPH)</td>
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<tr>
<td>NCT00910234</td>
<td>?</td>
<td>Recombinant Epo for Neuroprotection in Very Preterm Infants China Medical University (Bai-Horng Su, MD, PhD)</td>
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<tr>
<td>NCT00413946</td>
<td>Ongoing</td>
<td>Does Erythropoietin Improve Outcome in Very Preterm Infants? Swiss Neonatal Network (Hans U Bucher, Prof. University of Zurich)</td>
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<tr>
<td>NCT00413946</td>
<td>Ongoing</td>
<td>Brain Imaging and Developmental Follow up of Infants Treated With Erythropoietin (BRITE) Univ. of New Mexico (Robin K Ohls, MD)</td>
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<tr>
<td>NCT01378273</td>
<td>Not yet enrolling</td>
<td>Trial of Erythropoietin Neuroprotection in Extremely Preterm Infants (PENUT) University of Washington (Sandra Juul, MD, PhD)</td>
</tr>
<tr>
<td>NCT00513240</td>
<td>Ongoing</td>
<td>Epo for Brain Protection in Neonatal Open Heart Surgery Baylor College of Medicine/Texas Children's Hospital (Dean B. Andropoulos, MD)</td>
</tr>
</tbody>
</table>
Other Planned Trials

Term HIE (NE)
- Epo + hypothermia (NEAT 2 trial)
- Darbe + hypothermia (DANCE trial)
- Neonatal Network trial?

Preterm Neuroprotection
- PENUT trial
- Neonatal Network trial?
Thanks to...

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- CHDD
- UW NICU & SCH
- Our patients and families
- Investigators who have shared their data

NICHD, NINDS
March of Dimes
RoFAR