The efficacy of erythropoietin in treating experimental traumatic brain injury: a systematic review of controlled trials in animal models

A review

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Object. Erythropoietin (EPO) shows promise as a neuroprotective agent in animal models of traumatic brain injury (TBI). However, clinical trials of the efficacy of EPO treatment in patients with TBI yield conflicting results. The authors conducted a systematic review and meta-analysis to assess the effect of EPO in experimental animal models of TBI, the goal being to inform the design of future clinical trials.

Methods. The authors identified eligible studies by searching PubMed, Web of Science, MEDLINE, Embase, and Google Scholar in October 2013. Data were pooled using the random-effects model, and results were reported in terms of standardized mean difference. Statistical heterogeneity was examined using both I² and chi-square tests, and the presence of small study effects was investigated with funnel plots and Egger tests. In-depth analyses were performed for lesion volume and neurobehavioral outcome, and the studies’ methodological quality was also evaluated.

Results. Of a total of 290 studies, 13 found an effect of EPO on lesion volume and neurobehavioral outcome. Overall, the methodological quality of the studies was poor, and there was evidence of statistical heterogeneity among the publications as well as small-study effects. However, in-depth analyses showed statistically significant findings in favor of a beneficial effect of EPO after TBI.

Conclusions. Despite limitations of this systematic review that may have influenced the findings, the authors conclude that EPO might be beneficial in treating experimental TBI in terms of reducing lesion volume and improving neurobehavioral outcome. However, this review also indicates that more well-designed and well-reported animal studies are needed.

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KEY WORDS • efficacy • erythropoietin • systematic review • traumatic brain injury

SYSTEMATIC reviews and meta-analyses are fundamental tools used to interpret the effectiveness of a treatment across many studies. Recently, the need to conduct systematic reviews and meta-analyses of animal studies modeling clinically relevant problems has been highlighted.9,13,36 In particular, systematic reviews of animal experiments will allow decisions regarding the design and conduct of subsequent clinical trials to be based on the entirety of the existing evidence that is synthesized in an unbiased manner. Moreover, systematic reviews permit a more objective appraisal of evidence than is allowed by the traditional narrative-style reviews that are more commonly associated with animal research.

Traumatic brain injury (TBI), which is the leading cause of long-term disability in children and young adults worldwide,17 causes a variety of cognitive, emotional, and behavioral problems that can occur either individually or in combination.20,40 In the US, a case of TBI occurs every 15 seconds, resulting in 1.7 million new head injury victims per year. Each year, on average, these events are responsible for 50,000 deaths, leave 80,000 individuals with permanent disabilities, and cost more than US$77 billion.34 However, there are no pharmacological treat-
ment options for TBI because the translation of neuroprotective effects from preclinical studies to clinical practice has so far failed.17,23

Erythropoietin (EPO), a secreted 30-kD glycoprotein, is a multifunctional tissue-protecting agent that exerts antiapoptotic, antiinflammatory, antioxidative, angiogenic, and neurotrophic effects.7,28 Recently, several investigations have shown that EPO and its analogs provide substantial benefits to rodents after TBI.13,15,22 However, a pilot randomized trial in humans demonstrated that EPO does not reduce neuronal cell death compared with placebo, although TBI severity was worse in the EPO group, making it difficult to rule out a treatment effect.29 This discrepancy between animal and human studies might be due to bias in the conduct or reporting of animal experiments.9 Therefore, systematic reviews and meta-analyses can point to the cause of bias in animal experiments and provide a clear description of the circumstances under which the treatment is efficacious.

Some argue for the need to perform rigorous systematic reviews and meta-analyses of all available animal data before proceeding to clinical trials9,25 not only to reveal the specific conditions under which a drug can have neuroprotective effects but also to provide insights into potential limitations of the drug that may influence its clinical usefulness.13,18 A systematic review and meta-analyses of the efficacy of EPO and its analogs in treating TBI in experimental animal studies have not yet been conducted. Therefore, our main objective was to systematically investigate whether the evidence from animal studies favors a beneficial effect of EPO in reducing lesion volume and improving neurobehavioral outcome after TBI.

Methods

Literature Search

In October 2013 we searched 5 electronic databases (PubMed, Web of Science, MEDLINE, Embase, and Google Scholar) using the terms EPO (or erythropoietin) and TBI (or traumatic brain injury), limiting results to animal studies. Reference lists from the resulting research articles and reviews were used to identify further relevant publications.

Two investigators assessed the titles and abstracts of studies, and obtained copies of articles that described controlled studies of EPO or its analogs in animal models of TBI and that measured both lesion volume and neurobehavioral outcome. We included all studies in which neurobehavioral measurements were conducted not only after but also before TBI, thereby establishing a baseline for normal or pre-TBI function. Prespecified exclusion criteria were as follows: administration of any other neuroprotective agent in the treatment group, no control group, or data presented in duplicate by additional publications. Disagreements between investigators were resolved by consensus after discussion (Fig. 1).

Statistical Analysis

Results were calculated as the standardized mean difference ([SMD]; reported in units of standard deviation), which allows data measured on different scales to be merged, and 95% confidence intervals. The within- and between-study variation or heterogeneity was assessed using the Cochrane $Q$-statistic,10,35 with a significant $Q$-statistic ($p < 0.10$) indicating heterogeneity among studies. Heterogeneity was also quantified with the $I^2$ statistic, which allows data measured on different scales to be assessed whether the preclinical EPO studies met the current Stroke Therapy Academic Industry Roundtable (STAIR) recommendations.12,26

Methodological Quality of Studies

The methodological quality of individual studies was assessed using an 8-point rating scale as previously described.13 One point was given for written evidence of each of the following criteria: presence of randomization; monitoring of physiological parameters; assessment of dose-response relationship; assessment of optimal treatment time window; blind measurements of outcomes; assessment of outcome on Days 1–3; assessment of outcome on Days 1–30; and combined measurement of lesion volume and neurobehavioral outcome. We also evaluated whether the preclinical EPO studies met the current Stroke Therapy Academic Industry Roundtable (STAIR) recommendations.12,26

Results were calculated as the standardized mean difference ([SMD]; reported in units of standard deviation), which allows data measured on different scales to be merged, and 95% confidence intervals. The within- and between-study variation or heterogeneity was assessed using the Cochrane $Q$-statistic,10,35 with a significant $Q$-statistic ($p < 0.10$) indicating heterogeneity among studies. Heterogeneity was also quantified with the $I^2$ statistic, which allows data measured on different scales to be assessed whether the preclinical EPO studies met the current Stroke Therapy Academic Industry Roundtable (STAIR) recommendations.12,26

For studies comparing different doses and/or timing of drug administration with a single control group, we pooled data from all experimental groups to compare with the control group. Pooled effect size was estimated using fixed and random-effects models. When there was heterogeneity among studies, pooled effect size was estimated using a random-effects model.13,53 The presence of small study effects was investigated with funnel plots and Egger tests. For Egger tests, a p value of <0.10 was considered to indicate the presence of small study effects.10
Finally, we assessed the impact of several variables (type of EPO treatment, treatment dose, timing and duration of treatment, animal species) on the efficacy of EPO by using meta-regression when substantial or considerable heterogeneity existed. All statistical analyses were performed using Stata software (version 12.0).

Results

Description of Studies

We identified 290 studies (35 from PubMed, 94 from MEDLINE, 102 from Web of Science, 0 from Embase, and 59 from Google Scholar). On the basis of predefined criteria, 277 studies were excluded, leaving 13 studies for systematic review. One study was excluded from the meta-analysis because sample sizes were not stated, and we were unable to obtain this information from the authors. Another study was excluded because lesion volume was measured in cubic millimeters and MWM performance was measured in seconds. Therefore, the meta-analysis ultimately included 11 studies.

Among the included studies (Table 1), treatment outcomes were measured for 1 to 90 days after TBI. Ten studies used rats, and 3 studies used mice. All studies assessed lesion volume and neurobehavioral function; 12 used the MWM to assess cognitive function, 7 used the mNSS to assess neurological function, and 9 used the foot-fault test to assess sensorimotor function.

Methodological Quality of Studies

The median quality score of included studies was 6 of 8 (range 2–8). Two studies had a score of 8, 1 study had a score of 7, and 5 studies had a score of 6. Animals were randomly allocated to treatment groups in 7 studies. Only 1 study did not report monitoring of physiological parameters (although the majority of the other studies only monitored body or rectal temperature). Only 3 studies assessed dose-
TABLE 1: Characteristics of the 13 studies included in the systematic review*

<table>
<thead>
<tr>
<th>Authors &amp; Year</th>
<th>Animal Species &amp; Sex (no.)</th>
<th>Type of Model</th>
<th>Drug (treated/ control)</th>
<th>Main Experimental Groups</th>
<th>Method of EPO Administration</th>
<th>Outcome Measures</th>
<th>Time of Measurement of Outcome</th>
<th>Quality Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grasso et al., 2007</td>
<td>M SDRs (66)</td>
<td>cryogenic cerebral injury</td>
<td>rhEPO/vehicle</td>
<td>1) sham op; 2) TBI plus placebo; 3) TBI plus rhEPO</td>
<td>1000 IU/kg rhEPO administered ip at 5 min after injury, &amp; continued every 8 hrs up to 14 days</td>
<td>MWM test, brain edema, BBB integrity, lesion vol</td>
<td>1–14 days</td>
<td>2</td>
</tr>
<tr>
<td>Xiong et al., 2007</td>
<td>young adult M &amp; F C57BL/6 mice (56)</td>
<td>CCI injury</td>
<td>rhEPO/sal</td>
<td>1) M sal group; 2) M EPO group; 3) F sal group; 4) F EPO group; 5) sham group</td>
<td>5000 U/kg body wt rhEPO administered ip at 6 hrs &amp; at 3 &amp; 7 days (total dosage 15,000 U/kg) after TBI</td>
<td>MWM test, foot-fault test, lesion vol</td>
<td>1–35 days</td>
<td>4</td>
</tr>
<tr>
<td>Xiong et al., 2008</td>
<td>adult M C57BL/6 mice (37)</td>
<td>CCI injury</td>
<td>rhEPO/sal</td>
<td>1) sal group; 2) EPO group; 3) sham group</td>
<td>5000 U/kg body wt rhEPO administered ip at 6 hrs &amp; at 3 &amp; 7 days post-TBI (total dosage 15,000 U/kg)</td>
<td>MWM test, foot-fault test, lesion vol</td>
<td>1–35 days</td>
<td>4</td>
</tr>
<tr>
<td>Zhang et al., 2009</td>
<td>young adult M Wistar rats (25)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham group; 2) TBI + sal group; 3) TBI + EPO group; 4) TBI + EPO + hemodilution group</td>
<td>5000 U/kg EPO administered ip at Days 1, 2, &amp; 3 postinjury</td>
<td>mNSS, foot-fault tests, MWM test, lesion vol</td>
<td>1–35 days</td>
<td>6</td>
</tr>
<tr>
<td>Xiong et al., 2010</td>
<td>young adult M Wistar rats (25)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham group; 2) TBI + sal group; 3) TBI + EPOx1 group; 4) TBI + EPOx3 group</td>
<td>5000 U/kg EPO administered ip at 1 day (EPOx1 group) or at Days 1, 2, &amp; 3 (EPOx3 group) postinjury</td>
<td>mNSS, foot-fault test, MWM test, lesion vol</td>
<td>1–35 days</td>
<td>8</td>
</tr>
<tr>
<td>Xiong et al., 2010*</td>
<td>young adult F EPOR-null &amp; wild-type mice (72)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham group; 2) TBI + sal group; 3) TBI + EPO group</td>
<td>5000 U/kg EPO administered ip at 6 hrs &amp; 3 &amp; 7 days postinjury</td>
<td>MWM test, foot-fault test, lesion vol</td>
<td>1–35 days</td>
<td>6</td>
</tr>
<tr>
<td>Zhang et al., 2010</td>
<td>young adult M Wistar rats (24)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham control; 2) TBI + sal; 3) TBI + EPO</td>
<td>5000 U/kg EPO administered ip at Days 1, 2, &amp; 3 postinjury</td>
<td>mNSS, foot-fault test, lesion vol</td>
<td>1–35 days</td>
<td>6</td>
</tr>
<tr>
<td>Meng et al., 2011</td>
<td>young adult M Wistar rats (48)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham; 2) TBI/sal group; 3) TBI + EPO1K; 4) TBI + EPO3K; 5) TBI + EPO5K; 6) TBI + EPO7K</td>
<td>EPO at doses of 0 (sal), 1000 (EPO1K), 3000 (EPO3K), 5000 (EPO5K), &amp; 7000 (EPO7K) U/kg body wt was administered ip at 24, 48, &amp; 72 hrs after TBI</td>
<td>mNSS, foot-fault test, MWM test, lesion vol</td>
<td>1–35 days</td>
<td>8</td>
</tr>
<tr>
<td>Ning et al., 2011</td>
<td>young M Wistar rats (23)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sal group; 2) EPO 6-hr group; 3) EPO 24-hr group</td>
<td>5000 U/kg EPO in sal was administered ip at 6 hrs &amp; at 1 &amp; 2 days (EPO 6-hr group) or at 1, 2, &amp; 3 days (EPO 24-hr group) postinjury</td>
<td>mNSS, foot-fault test, MWM test, lesion vol</td>
<td>1–90 days</td>
<td>7</td>
</tr>
<tr>
<td>Xiong et al., 2011*</td>
<td>young adult M Wistar rats (32)</td>
<td>CCI injury</td>
<td>CEPO/sal</td>
<td>1) sham; 2) TBI + vehicle; 3) TBI + CEPO x 1; 4) TBI + CEPO x 3</td>
<td>50 μg/kg body wt CEPO administered ip at 6 hrs (for the CEPO x 1 group) or at 6, 24, &amp; 48 hrs (for the CEPO x 3 group) after TBI</td>
<td>mNSS, foot-fault test, MWM test, lesion vol</td>
<td>1–35 days</td>
<td>6</td>
</tr>
<tr>
<td>Xiong et al., 2011*</td>
<td>young adult M Wistar rats (64)</td>
<td>CCI injury</td>
<td>EPO/sal</td>
<td>1) sham; 2) sal; 3) EPO + DMSO; 4) EPO + SU5416</td>
<td>5000 U/kg body wt EPO administered ip at 24, 48, &amp; 72 hrs after TBI</td>
<td>mNSS, MWM test, lesion vol</td>
<td>1–35 days</td>
<td>5</td>
</tr>
</tbody>
</table>

(continued)
response relationships, and 3 studies investigated the optimal time window for administering EPO. Four studies did not report that outcome measures were made by researchers who were blind to animal treatment. All 13 studies assessed treatment outcome at Days 1–3, and 12 studies also assessed outcome at Days 1–30. Overall, studies incompletely followed the STAIR recommendations (Table 2).

**Lesion Volume**

Within 10 studies there were 17 comparisons (involving 264 animals) of lesion volume 35 days after TBI. Pooled analysis indicated that animals in the treatment groups had significantly reduced lesion volume compared with animals in the control groups (SMD = −1.715, 95% CI −2.359 to −1.071, p < 0.0001).

There was evidence of considerable heterogeneity among studies (χ² = 76.27, df = 16 [p < 0.0001], I² = 79.0%) and small study effects (Egger test bias coefficient = −4.08, 95% CI −10.06 to −3.16, p = 0.001) (Fig. 2).

**Foot-Fault Test**

Nine studies evaluated sensorimotor function by using the foot-fault test 14 days after TBI. Pooled analysis indicated that animals in the treatment groups showed a significant improvement in sensorimotor function compared with animals in the control groups (overall: SMD = −2.813, 95% CI −3.15 to −2.473; forelimb: SMD = −3.153, 95% CI −3.635 to −2.670, p < 0.0001; hindlimb: SMD = −2.483, 95% CI −2.919 to −2.046, p < 0.0001).

There was acceptable heterogeneity among studies (overall: χ² = 61.33, df = 33 [p = 0.002], I² = 46.2%; forelimb: χ² = 26.41, df = 16 [p = 0.049], I² = 39.4%; hindlimb: χ² = 27.75, df = 16 [p = 0.034], I² = 42.5%); and small study effects for foot-fault test (Egger test bias coefficient = −4.88, p < 0.0001) (Fig. 3).

**The mNSS**

Within 7 studies there were 13 comparisons (involving 185 animals) of neurological function using the mNSS 14 days after TBI. Pooled analysis indicated that animals in the treatment groups had significantly lower mNSSs (that is, better neurological function) than animals in the control groups (SMD = −2.930; 95% CI −3.527 to −2.334, p < 0.0001).

There was acceptable heterogeneity among studies (overall: χ² = 22.66, df = 12 [p = 0.031], I² = 47.0%); and small study effects (Egger test bias coefficient = −10.78, 95% CI −6.93 to −4.58, p < 0.0001) (Fig. 4).

**The MWM Test**

Within 10 included trials there were 18 comparisons (involving 296 animals) of spatial learning and memory deficits performed using the MWM 35 days after TBI. Pooled analysis indicated that animals in the treatment groups significantly improved spatial learning performance (that is, greater percentage of time spent in the correct quadrant) compared with animals in the control groups (SMD = 3.933; 95% CI 3.35–4.512, p < 0.0001).
There was acceptable heterogeneity among studies ($\chi^2 = 32.55$, df = 17 [p = 0.013], $I^2 = 47.8\%$) and small study effects (Egger test bias coefficient = 10.13 (95% CI 3.710158–5.675137, p < 0.0001) (Fig. 5).

**Meta-Regression Analysis**

A multivariate, random-effects regression model considering EPO treatment type, dose, timing and duration, and animal species explained 89.99% of the variance in the effects of EPO on lesion volume among 10 studies (Tables 2 and 3).

**Discussion**

It is important to assess the benefits of candidate neuroprotective drugs by considering both histopathological and functional outcome. We present the first systematic review and meta-analysis of the efficacy of EPO and its analogs in treating animal models of TBI. Although small study effects and statistical heterogeneity were present among studies, we found that EPO and its analogs potentially exert neuroprotective effects in terms of reducing lesion volume and improving neurobehavioral outcome after TBI. These findings extend previous systematic reviews of controlled trials in animal models, which report that progesterone and beta-2 receptor antagonists also exert neuroprotective effects against TBI.

Critical appraisal of the methodological quality of studies is an essential part of systematic reviews. We assessed methodological quality in accordance with previously described standards for preclinical development of neuroprotective drugs that were established by a panel of expert stroke researchers to address the failure of many clinical trials of these drugs for acute ischemic stroke. Overall, we found that the methodological quality of the included EPO/TBI studies was poor; many failed to accurately report randomization of group assignment, blinded assessment of outcome, investigation of the optimal time window for treatment, and determination of a dose-response relationship, which are important issues that are generally required in clinical studies. Moreover, none of the included studies reported calculations of optimal sample size or criteria for inclusion/exclusion. Also, EPO treatment efficacy in rabbits, cats, or gyrencephalic primates has not yet been tested and should be considered in future studies.

This systematic review has some limitations. First, our objective was to assess the overall efficacy of EPO and its analogs, and we did not undertake analyses to investigate the presence of dose-response relationships or effects related to the timing of drug administration, such as those performed in a previously published study. It should also be noted that our pooling of all doses may lead to an underestimation of treatment efficacy, assuming the existence of a true dose-response relationship.

Second, publication bias, which is considered a potential threat to the validity of all systematic reviews of experimental studies, should also be considered. Although we made an extensive effort to identify all relevant published and unpublished studies, we were only able to include data that were published in some form; hence our analysis did not take unpublished data into account. Because studies with negative results are less likely to be published, our results may overstate overall effect size. The funnel plots and the Egger tests suggest the possibility of publication bias or other small-study biases, consistent with that observed in previous systematic reviews of animal studies.

Third, nonpublication of studies serves to limit available information on the effect of treatment under certain testing conditions, such as specific doses or timing of treatment. Extracting multiple pieces of information from a single publication has the potential to introduce bias into systematic reviews because the results were generated by the same investigators. As found in this review,
the existence of true heterogeneity should be considered as a potential explanation.

Fourth, there were differences among studies in terms of animal species, physiological parameters, methods of drug administration, and experimental protocols. We used regression analysis to explore sources of variation in the effect of EPO on lesion volume. Unfortunately, it was not possible to judge accurately whether the relationships we observed were independent of these factors, which also led to statistical heterogeneity and made the analysis less reliable.

Fifth, although a fundamental assumption is that the results of animal studies, if performed well enough, will predict effects in humans, promising neuroprotective drugs previously identified as effective in animal TBI models have failed in Phase II or III clinical trials.45

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**Fig. 2.** Upper: Forest plot showing results from meta-analysis of lesion volume. Pooled analysis indicated that animals in the treatment groups had significantly reduced lesion volume compared with animals in the control groups (SMD = -1.715, 95% CI -2.359 to -1.071, p < 0.0001). Lower: Begg funnel plot of lesion volume. There was evidence of small study effects (Egger test bias coefficient = -4.08, 95% CI -10.0636 to -3.160554, p = 0.001). s.e. = standard error.
Fig. 3. **Upper:** Forest plot showing results from meta-analysis of foot-fault test. Pooled analysis indicated that animals in the treatment groups showed a significant improvement in sensorimotor function compared with animals in the control groups (overall: SMD = -2.813, 95% CI: -3.15 to -2.473; forelimb: SMD = -3.153, 95% CI: -3.6385 to -2.670, p < 0.001; hindlimb: SMD = -2.483, 95% CI: -2.919 to -2.0546, p < 0.0001). **Lower:** Begg funnel plot of foot-fault test. There was evidence of small study effects (Egger test bias coefficient = -4.88, p < 0.0001).
Finally, there were several instances in which numerical data were not readily available and had to be derived from graphs. Although we enlarged the graphs, and data were independently extracted by 2 investigators, this technique can be imprecise. Therefore, our findings should be interpreted carefully.

To improve the transition from animal experiments to human clinical trials, future animal studies should report full methodological details to allow others to reproduce and validate their results and to enable more accurate reviews and meta-analyses, which would further scientific progress. Researchers are strongly encouraged to consult and follow the ARRIVE (Animal Research: Reporting In Vivo Experiments) guidelines when designing studies and writing up results, which is considered an important step forward in improving scientific reporting standards for animal research.8,21

Also, this systematic review focuses only on the overall effect of EPO on long-term outcomes following TBI, largely due to insufficient data regarding other short-term outcomes such as brain edema and blood-brain barrier permeability, which were reported in only one of the included studies.15 Moreover, most included studies used the controlled cortical impact injury model of TBI to investigate the effect of EPO; therefore, these limited results might be inadequate for predicting the treatment
Fig. 5. Upper: Forest plot showing results from meta-analysis of MWM test. Pooled analysis indicated that animals in the treatment groups showed significantly improved spatial learning (i.e., greater percentage of time spent in the correct quadrant) compared with animals in the control groups ($SMD = 3.933$, $95\% CI 3.355–4.512$, $p < 0.0001$). Lower: Begg funnel plot of MWM test. There was evidence of small study effects (Egger test bias coefficient $= 10.13$, $95\% CI 3.710501–5.675137$, $p < 0.0001$).

<table>
<thead>
<tr>
<th>Study ID</th>
<th>SMD (95% CI)</th>
<th>Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meng et al. 2011-1</td>
<td>4.23 (2.38, 6.08)</td>
<td>5.47</td>
</tr>
<tr>
<td>Meng et al. 2011-2</td>
<td>5.81 (3.45, 8.17)</td>
<td>4.03</td>
</tr>
<tr>
<td>Meng et al. 2011-3</td>
<td>7.55 (4.59, 10.51)</td>
<td>2.91</td>
</tr>
<tr>
<td>Meng et al. 2011-4</td>
<td>3.20 (1.66, 4.74)</td>
<td>6.60</td>
</tr>
<tr>
<td>Ning et al. 2011-1</td>
<td>3.74 (1.98, 5.51)</td>
<td>5.76</td>
</tr>
<tr>
<td>Ning et al. 2011-2</td>
<td>2.77 (1.30, 4.24)</td>
<td>6.66</td>
</tr>
<tr>
<td>Xiong et al. 2007-1</td>
<td>3.88 (2.14, 5.62)</td>
<td>5.84</td>
</tr>
<tr>
<td>Xiong et al. 2007-2</td>
<td>3.26 (2.04, 4.49)</td>
<td>7.95</td>
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<tr>
<td>Xiong et al. 2008</td>
<td>2.31 (1.28, 3.33)</td>
<td>8.89</td>
</tr>
<tr>
<td>Xiong et al. 2010-1*</td>
<td>3.13 (1.35, 4.91)</td>
<td>5.71</td>
</tr>
<tr>
<td>Xiong et al. 2010-2*</td>
<td>5.72 (3.09, 8.34)</td>
<td>3.49</td>
</tr>
<tr>
<td>Xiong et al. 2010-1*</td>
<td>4.20 (2.57, 5.83)</td>
<td>6.25</td>
</tr>
<tr>
<td>Xiong et al. 2010-2*</td>
<td>6.48 (3.89, 9.08)</td>
<td>3.55</td>
</tr>
<tr>
<td>Xiong et al. 2011-1*</td>
<td>3.56 (1.92, 5.21)</td>
<td>6.19</td>
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<td>Xiong et al. 2011-2*</td>
<td>5.97 (3.55, 8.39)</td>
<td>3.91</td>
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<tr>
<td>Xiong et al. 2011*</td>
<td>4.46 (2.54, 6.38)</td>
<td>5.24</td>
</tr>
<tr>
<td>Zhang et al. 2009</td>
<td>4.25 (2.07, 6.44)</td>
<td>4.48</td>
</tr>
<tr>
<td>Zhang et al. 2012</td>
<td>2.78 (1.31, 4.25)</td>
<td>6.86</td>
</tr>
<tr>
<td>Overall (I-squared = 47.8%, $p = 0.013$)</td>
<td>3.93 (3.35, 4.51)</td>
<td>100.00</td>
</tr>
</tbody>
</table>

Table 3: Meta-regression analysis *

| Covariate                  | Coefficient | SE     | t      | $p > |t|$ | 95% CI       |
|---------------------------|-------------|--------|--------|-------|-------------|
| treatment dose            | -0.7318648  | 0.4506005 | -1.62  | 0.130 | -1.713639   | 0.2499093             |
| type of EPO treatment     | -1.883973   | 0.7043093 | -2.67  | 0.020 | -3.418531   | -0.3494147            |
| timing & duration of treatment | 0.5522269  | 0.7554 | 0.73   | 0.479 | -1.093648   | 2.198102              |
| animal species             | -1.744021   | 0.345596 | -5.05  | 0.000 | -2.49701    | -0.9910316            |
| _cons                      | 4.540593    | 2.514451 | 1.81   | 0.096 | -0.9379261  | 10.01911              |

* Residual maximum likelihood estimate of between-study variance, $\tau^2 = 0.2108$; % residual variation due to heterogeneity, $I^2_{res} = 40.76\%$; proportion of between-study variance explained as adjusted $R^2 = 89.99\%$; joint test for all covariates Model $F(4,12) = 9.42$, with Knapp-Hartung modification probably $> F = 0.0011$. _cons = constant; _ES = effect size; _res = residual; t = test for coefficient.
response in patients. With that in mind, prior to making any clinical practice recommendations, more appropriate and standardized experimental studies are needed to better evaluate the impact of this promising pharmacological intervention for TBI.

Conclusions

Despite its limitations, this systematic review and meta-analysis show that EPO and analogs can reduce lesion volume and improve neurobehavioral outcomes in animal models of TBI. However, without rigorous, robust, and detailed preclinical evaluation, it is unlikely that novel neuroprotective drugs will prove effective when tested in large, time-consuming, and expensive human clinical trials. Thus, more well-designed and well-reported experimental animal studies are needed.

Disclosure

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