Editorial

Erythropoietin and prenatal hypoxia-ischemia

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Discovered in the 1950s, erythropoietin (EPO) was originally known for its role in regulating the proliferation of immature erythrocyte precursors. Although EPO was believed to be produced only in the kidney and liver, Tan and colleagues\textsuperscript{28} showed in 1992 that other organs, including the brain, produced this cytokine in response to hypoxic-ischemic stress. This finding paved the way for a plethora of studies that increasingly confirmed a number of interrelated functional roles for EPO in the mammalian brain, some of which form the basis for the studies by Mazur and colleagues\textsuperscript{18} reported in this issue of \textit{Journal of Neurosurgery: Pediatrics}.

It is now appreciated that endogenously produced EPO reduces the extent of apoptotic injury following ischemia, trauma, and other insults,\textsuperscript{9,17,23} and astrocytes may be the primary cell type responsible for this production.\textsuperscript{3} However, all resident brain cells (neurons, astrocytes, microglia, and endothelial cells) express one or more EPO receptors (EPORs).\textsuperscript{12} Erythropoietin and EPORs play important roles in brain development.\textsuperscript{3} Very high expression levels of the EPOR predominate in the fetal brain, and, as in other tissues, levels diminish during fetal development, and then rapidly after birth.\textsuperscript{9} Expression of EPO itself increases over the time of gestation, but falls at birth as well.\textsuperscript{27} Generally speaking, the expression of both EPO and EPORs in adults are upregulated in a temporally distinct, cell-specific manner\textsuperscript{23,24} in response to many different stimuli, secondary to the stabilization of one or more isoforms of hypoxia-inducible factor, a transcription factor family responsible for inducing the expression of hundreds of survival-enhancing genes.\textsuperscript{21} Accumulating evidence suggests that endogenous EPO may be a primary inducer and effecter of “preconditioning”-induced cerebroprotection,\textsuperscript{6,10} as such, elucidating the molecular basis of protection afforded by the activation of innate neuroprotective pathways promises to advance therapeutics for many cerebrovascular disorders.

The elegant elucidation of the spatiotemporal features of sustained EPOR mRNA and protein upregulation in the prenatal brain in response to third trimester hypoxia-ischemia by Mazur et al. in the present study, in several cell types including oligodendroglial lineage cells, indicates that an oxygen- or metabolically driven, naturally self-regulating, EPO-based protection mechanism is likely operative, at least to some extent, in utero. But what Mazur and colleagues also discovered was that a corresponding increase in the expression of mRNA and protein levels of the ligand does not occur in the developing brain in response to prenatal hypoxia-ischemia. Based on the notion that unoccupied EPORs may predispose cells to apoptosis, the present investigators tested the hypothesis that supplying exogenous EPO systemically, starting several days after the hypoxic-ischemic event itself, may rescue the hypoxia-ischemia injured brain. Building on the seminal work by Sakanaka et al.,\textsuperscript{20} in the adult brain, a growing number of experimental studies have demonstrated EPO-based neuroprotection using pretreatment or immediate posttreatment protocols. However, the efficacy of delayed, postnatal EPO treatment for prenatal hypoxia-ischemia, and the wide, clinically relevant therapeutic window identified by Mazur et al. is an important advance, and certainly attractive from a clinical perspective.

Whereas it initially appeared unlikely that a large glycosylated molecule like EPO would be able to cross the blood-brain barrier (BBB), implying that blood-borne EPO would not affect brain function, several studies in both animals\textsuperscript{2} and humans\textsuperscript{23} demonstrated that this is not necessarily the case. These surprising pharmacokinetics underscore the success of a number of studies in adult animals using systemically administered EPO for ischemic\textsuperscript{19} and hemorrhagic\textsuperscript{11} stroke, as well as in animal models of postnatal hypoxia-ischemia, as described by Mazur et al. The dosing and timing of EPO administration in relation to the ischemic event bear prominently on the clinical relevance of these findings; delayed openings in the BBB, or age-dependent differences in barrier permeability and/or BBB transporters,\textsuperscript{23} might also contribute. In fact, the possibilities that the brain protective effects of systemically administered EPO result from endocytotic transport of EPO across an intact barrier, or the activation of luminally facing endothelial EPORs that trigger cascades of intercellular survival pathway signaling that culminate in neuronal protection, have yet to be ruled out. Regardless, the expanded therapeutic window identified by Mazur et al. is an exciting advancement for the periventricular leukomalacia field. Moreover, Mazur et al. demonstrate that EPO-treated newborn rats continued to exhibit evidence of lasting protection as adults, in the form of improved motor scores and lower seizure thresholds, confirming that this treatment did not simply delay the rate of injury progression, but in fact conferred permanent protection.

As alluded to by the authors, EPO and other protec-
different factors can act synergistically and mutually regulate one another’s activity; for example, EPO can activate vascular endothelial growth factor expression in endothelial cells. It will be important in future studies to address the participation of vascular endothelial growth factor and other downstream signaling molecules responsible for the EPO-mediated protection observed in the present study. Many reports have explored such regulatory schemes, downstream targets, and signaling pathways in detail in adult models, but few have identified whether these mechanisms, or completely unique ones, underlie EPO-mediated protection in the neonatal brain and, in turn, what defines the cerebral “EPO proteome” at this age. Moreover, whether “vasculoprotection” contributed to the improved outcomes observed by Mazur et al. was not addressed. But the finding that EPO improves vascular integrity and exerts ant apoptotic effects on cerebral endothelium, promotes angiogenesis, mobilizes endothelial progenitor cells from marrow, and augments the production of additional angiogenic factors, suggests this very well could be the case in the neonatal brain. Similarly, enhanced neuronal survival may also be secondary to an EPO-mediated protection of EPOR-expressing astrocytes, and/or the enhanced neuronal progenitor cell production from neural stem cells. Prior evidence that EPO augments the differentiation and maturation of oligodendrocyte precursors is in agreement with the current finding that EPORs are upregulated on ischemia-vulnerable oligodendroglial lineage cells. As exciting as the results of Mazur et al. appear, translating them to the clinic will not be without significant hurdles. For one, it may be necessary to activate cerebral EPORs by nonhematopoietic EPO analogues such as carbamylated and asialoerythropoietin derivatives to avoid the dangers of polycythemia, which Mazur et al. identified at higher EPO doses. Recent reviews and meta-analyses provide enthusiastic support for the efficacy of EPO and its analogues for treating ischemic brain injury in humans, including a Phase II trial for subarachnoid hemorrhage. However, the long-awaited publication earlier this year of results of the Phase II/III trial for acute ischemic stroke in adults indicated that EPO was actually not associated with any favorable outcomes, and in fact raised serious safety concerns in the subgroup of patients who also received thrombolytics. Nevertheless, the brain of a baby may ultimately prove to be a less risky target for EPO-based therapeutics, and the findings in the present work by Mazur et al. represent key preclinical advances needed to at least consider such an exciting possibility.

References

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