Title:  *Elucidating Molecular Mechanisms Underlying EPO in the Developing Brain.*

Background:

Hypoxic Ischemic Encephalopathy (HIE), a brain injury caused by a combination of inadequate blood flow and oxygen to the brain, has a high morbidity and mortality leading to death in 15-20% of cases and severe neurological damage in another 25%. While therapeutic hypothermia can somewhat benefit the prognosis of an infant suffering from hypoxic insult, this treatment can only be utilized in term or near term infants and its availability is limited. Recent research has shown that administering exogenous erythropoietin (EPO) may have neuroprotective effects on infants suffering from hypoxic insult, and further that the EPO pathway plays an important role in normal neural development. Little is known regarding the underlying molecular mechanisms downstream of EPO signaling in the brain. We hypothesize that EPO signaling promotes specific patterns of epigenetic modifications, transcription factor binding, and mRNA expression both during development and in response to HIE.

Objective:

The goal of our study is to elucidate the molecular mechanisms underlying the action of EPO and EpoR pathway in the developing brain.

Results:

In order to mimic the developing brain, three neural cell lines were differentiated to neural cells as a model system. Cell morphology and mRNA expression analyses demonstrated that retinoic acid and/or growth factor withdrawal induces neural differentiation in these cell lines. To study the role of EPO in the response to hypoxia, NTERA-2, BE(2)C and Myc-NRP cells were differentiated several days and subsequently exposed to a hypoxic environment in the presence or absence of EPO. Secondary effects of hypoxia such as nutrient deprivation, was also studied +/- EPO. Gene expression analyses of the hypoxia treated cells with EPO suggests that EPO may play a role in the response to hypoxia with an increased expression of the anti-apoptosis genes BclxL and bcl2 as well as its own receptor, EpoR.

Conclusion:

In conclusion, the three cell lines used may be used as a model to study the molecular mechanisms underlying the effect of hypoxia and EPO *in vitro.* In addition, these studies suggest that EPO upregulates anti-apoptotic genes, BclxL and Bcl2, proposing an underlying neuroprotective mechanism.