26367 - ANEMIA INCREASES HIF-1 α PROTEIN EXPRESSION IN RAT CEREBRAL CORTEX

Anya McLaren BSc, CDavid Mazer, MD; Duncan Stewart, MD; Gregory Hare, St. Michael's Hospital, University Of Toronto, Toronto, ONTARIO, Canada

INTRODUCTION: Acute hemodilutional anemia accentuates cerebral injury by currently undefined mechanisms. The current study tests the hypothesis that acute hemodilutional anemia causes cerebral tissue hypoxia by triggering an increase in cerebral hypoxia inducible factor-1 α ; (HIF-1 α ;) protein levels at clinically relevant hemoglobin concentrations.

METHOD: With Animal Care Committee approval, anesthetized rats underwent tail artery and vein cannulation to monitor arterial pressure and for the purpose of performing hemodilution. Hemodilutional anemia was achieved by exchanging 30 ml.kg⁻¹ of blood with pentastarch over 10 minutes. Hemoglobin (Hb) concentrations were assessed by co-oximetry. Control animals underwent blood exchange. Brain harvesting was done at 0 and 18 hours after hemodilution/blood exchange. Animals exposed to hypoxia (10% oxygen for 3 hours) served as positive controls. Quantitative RT-PCR was carried out on cerebral cortical tissue using standard techniques. Cerebral cortical HIF-1 α ; protein levels were assessed by western blot (polyclonal anti HIF-1 α ;, Novus Biologicals). Band density was quantified digitally and reported as pixels.mg protein⁻¹. HIF-1 α ; protein localization in the cerebral cortex was assessed by immunohistochemistry (monoclonal anti HIF-1 α ;, Novus Biologicals). A two-way ANOVA and post hoc Tukey test were used to assess data where relevant. Significance was assigned at $p < 0.05$ (Mean $\pm$ SD).

RESULTS: The hemoglobin concentration in anemic rats decreased to 58 ± 5.9 g.L⁻¹ following hemodilution ($p < 0.05$). There was no increase in the cerebral cortical HIF-1 α ; mRNA at 18 hours after hemodilution. However, there was a significant increase in cerebral cortical HIF-1 α ; protein in both anemic (1.5-fold) and hypoxic rats (2-fold) relative to controls ($p < 0.02$). The HIF-1 α ; increase appears to be localized to the vicinity of the endothelium and subendothelium of the cerebral cortex.

DISCUSSION: Acute hemodilutional anemia caused a 50% increase in cerebral cortical HIF-1 α ; protein with no changes in HIF-1 α ; mRNA levels. This increase in cerebral cortical HIF-1 α ; protein levels may activate neuroprotective mechanisms within the cerebral cortex. The stimulus for increased cerebral cortical HIF-1 α ; may be induced by cerebral tissue hypoxia or non hypoxic mediators (CAS, PSI Support).