26428 - THE NEUROPROTECTIVE EFFECTS OF ANTI-CD18 THERAPY AFTER TRANSIENT GLOBAL ISCHEMIC BRAIN INJURY IN THE MOUSE

Dermot Doherty FCA RCSI, Ryan Salewski, BSc; Julia Lockwood, Cynthia Hawkins, Jamie Hutchison, FRCPC; The Hospital For Sick Children, Toronto, ON, Canada

Background: Transient global cerebral ischemia is an important mechanism of brain injury following cardio-respiratory arrest, traumatic brain injury, surgery for repair of congenital heart lesions and neurosurgery. We have previously shown that global cerebral ischemia leads to an increase in leukocyte-endothelial cell interactions in the pial microcirculation of the mouse using intravital microscopy through a cranial window. We have also shown an increase in cerebral endothelial cell adhesion receptor mRNA and protein expression, for several days following ischemia using laser capture microdissection, PCR and immunohistochemical techniques. Our hypothesis was that blocking leukocyte adhesion to cerebral endothelial cells using a monoclonal antibody to the leukocyte α -2 integrin adhesion receptor (anti-CD18 MAb) would improve neuronal survival and functional outcome following global cerebral ischemia.

Methods: Male C57Bl/6 mice were subject to 10 minutes of bilateral occlusion of the common carotid artery (BCCO) or sham operation followed by reperfusion. Mice were randomized to 4 experimental groups: 1) BCCO ischemia followed by injections of anti-CD18; 2) BCCO ischemia followed by injections of isotype matched non-binding control antibody (NBC); 3) sham surgery followed by injections of anti-CD18; and 4) sham surgery followed by injections of NBC. Anti-CD18 or NBC antibodies were injected i.p. twice a day for three days beginning immediately following sham or BCCO ischemia in all 4 experimental groups. Mice were sacrificed at 72-hours following ischemia or sham and brains were embedded in paraffin and sectioned (4 μ m). Brain sections were stained with H & E and adjacent sections were stained for fibronectin using immunohistochemistry and examined by a neuropathologist for hippocampal damage and breakdown of the blood-brain barrier and peri-vascular staining with fibronectin. In separate experiments, mice were trained to locate a submerged platform in a Morris Water Maze, subject to BCCO or sham and a probe test was performed 7 days later. Mice were then retrained to locate the submerged platform and another probe test was done at 21 days post-ischemia or sham. Mice were then sacrificed at 4 weeks post-ischemia or sham and the brains were examined for hippocampal damage. N=10-11 mice were used in each experimental group.

Results: No neuronal cell death and only minimal perivascular fibronectin staining were seen in sham animals. Cerebral ischemia led to marked damage and perivascular fibronectin staining. Anti-CD18 MAb reduced damage by 65 and 74% respectively assessed at 72 hours and 4 weeks following ischemia and reduced fibronectin staining by 85%. Animals subject to BCCO ischemia also had significant impairment of memory and anti-CD18 MAb led to significant improvement in memory when assessed at 7 and 21 days following the injury.

Conclusion: Anti-CD18 Mab leads to significant improvement in hippocampal cell survival, breakdown of the blood-brain barrier and memory following global cerebral ischemia in the mouse.