Notice of the Final Oral Examination
for the Degree of Doctor of Philosophy

of

PATRICK REESON

BSc (University of Calgary, 2005)

“Microvascular plasticity in the healthy and diseased mouse cortex”

Division of Medical Sciences

Friday, June 15, 2018
10:00 A.M.
Medical Sciences Building
Room 160

Supervisory Committee:
Dr. Craig Brown, Division of Medical Sciences, University of Victoria (Supervisor)
Dr. Patrick Nahirney, Division of Medical Sciences, UVic (Member)
Dr. Bob Chow, Department of Biology, UVic (Outside Member)

External Examiner:
Dr. Grant Gordon, Department of Physiology & Pharmacology, University of Calgary

Chair of Oral Examination:
Dr. Wanda Boyer, Department of Educational Psychology and Leadership Studies, UVic

Dr. Stephen Evans, Acting Dean, Faculty of Graduate Studies
Abstract

Brain function relies on a properly functioning vasculature system, to both deliver oxygen and metabolites, remove metabolic waste, and to help maintain brain function. However, as in all biological systems, the cerebral vasculature is sometimes challenged by small or large-scale failures that threaten the function of local vascular systems, and the neuronal networks they support. While often considered two different systems within the brain, the functional and structural interdependencies of the vasculature and nervous system means vascular dysfunction has downstream effects on brain function. Cerebral capillaries are specifically prone to spontaneous obstructions, randomly stopping flow in vessels throughout the cortex. While not surprising given capillaries are narrow, low pressure tubes that pass relatively large and adherent cells and debris, the ultimate outcomes of these obstructions are unknown. The vascular response to these events could have profound effects on brain health, as these random events accumulate over time. Similarly, while much research has studied the neural and vascular responses to large vessel obstructions (ischemic stroke), how common comorbidities which also afflict the vasculature, like diabetes, alters vascular plasticity and in turn neuronal rewiring and functional recovery is not understood. This dissertation furthers our understanding of how microvascular plasticity, in response to either small or larger interruptions in blood flow, effect brain health.

In the first aim I look at the fates of cortical capillaries in the mouse somatosensory cortex to either spontaneous or experimentally induced obstructions. Using in vivo 2 photon imaging of cortical blood flow I found that ~0.12% of cortical capillaries become obstructed each day. Tracking natural or microsphere induced obstructions in anesthetized or awake mice revealed that most capillaries recanalize. Remarkably, 30% of all obstructed capillaries failed to recanalize and were pruned by 21 days. This loss was not compensated for by any angiogenic sprouting in any imaging area. Using this information, I was able to predict capillary loss over time that closely matched experimental estimates. From a mechanistic perspective, VEGF-R2 signaling was a critical factor in dictating capillary re-canalization or pruning. Thus, this work reveals the incidence, mechanism and long-term outcome of capillary obstructions and contributes to our understanding of age related capillary rarefaction.

To provide a better understanding of the intersection between diabetes and stroke, I focused on blood brain barrier (BBB) function. Using a mouse model of type 1 diabetes, I revealed that
ischemic stroke leads to an abnormal and persistent increase in Vascular Endothelial Growth Factor Receptor 2 (VEGF-R2) expression in peri-infarct vascular networks. Correlating with this, BBB permeability was markedly increased in diabetic mice which could not be prevented with insulin treatment after stroke. Imaging of capillary ultrastructure revealed that BBB permeability was associated with an increase in endothelial transcytosis rather than a loss of tight junctions. Pharmacological inhibition or endothelial-specific knockdown of VEGF-R2 after stroke attenuated BBB permeability, loss of synaptic structure in peri-infarct regions, and improved recovery of forepaw function. However, the beneficial effects of VEGF-R2 inhibition on stroke recovery were restricted to diabetic mice and appeared to worsen BBB permeability in non-diabetic mice. These results showed that aberrant VEGF signaling and BBB dysfunction after stroke plays a crucial role in limiting functional recovery in an experimental model of diabetes. Overall this dissertation demonstrates in both small and larger scale vascular insults, in healthy of diseased brains, microvascular plasticity has profound effects in the brain.