**Three postdoctoral positions are available immediately; 1 each for the following projects.**

Dr. Lyanne Schlichter is a Professor of Physiology (Faculty of Medicine) at the University of Toronto, and Senior Scientist at the Krembil Research Institute (formerly the Toronto Western Research Institute). My laboratory specializes in the broad field of Neuroinflammation, with the aim of unravelling both the detrimental and beneficial aspects of brain inflammation. I am currently recruiting up to 3 postdoctoral fellows for 3 projects, which are described in the attached file. Funding has been secured to carry out the proposed project and provide salary support; however, successful applicants will also be expected to apply for salary awards.

In brief, my laboratory's research is broad-ranging: from using rodent models of ischemic stroke and intracerebral hemorrhage; studying the role of inflammation in brain damage; studying the many biological functions of the brain's immune cell (microglia); to studying expression, regulation and contributions of several ion channels to microglia functions and brain inflammation. Technical approaches include: stroke surgeries, immunohistochemistry and cytochemistry, cell-based assays (microglia and their interactions with other brain cells), patch-clamp electrophysiology, cellular fluorescence imaging, gene analysis and manipulation (including high-throughput and transgenic models), protein and FACS analysis.

**Seeking:** Postdoctoral fellows with the following traits and experience.
Requirements specific to each project are listed below.
- Fluent in English (spoken and written), with proven experience in writing papers
- Hard-working, adaptable and capable of both independent work and teamwork

Please identify which of the three projects for which you are applying.
***Project 1.*** Relevant experience in diseases of the CNS, brain histology, and related technical training, such as animal surgeries, immunohistochemistry, confocal microscopy
***Project 2.*** Some or most of the following skills: cell culture, immunocytochemistry, confocal imaging, routine molecular biology
***Project 3.*** Some or most of the following skills: cell culture, patch-clamp electrophysiology (mainly whole cell), cellular $Ca^{2+}$ imaging, fluorescence plate-reader assays

**Documents required:**
- Curriculum Vitae
- Cover letter that includes a statement describing your qualifications and research interests
- Confidential letters of recommendation (at least 2)

**Deadline to apply:**
Review of applications will begin immediately and continue until the positions are filled. The successful applicant will be expected to take up the position as soon as possible.

**Contact information: Dr. Lyanne Schlichter;** schlicht@uhnres.utoronto.ca

**Project 1. Inflammation and white-matter injury after ischemic or hemorrhagic stroke**
Ischemic stroke (loss of cerebral blood flow) is a leading cause of long-term disability in North America, and second leading cause of death worldwide. Primary intracerebral hemorrhage (ICH;

is caused by rupture of small brain arteries or arterioles and accounts for 10–15% of strokes in Westerners (more in Asians). When the brain is healthy it is highly vascularized but protected by an intact blood-brain barrier (BBB), which prevents entry of blood-borne immune cells, thereby restricting immune functions to the resident microglial cell. After either type of stroke, microglia rapidly respond and become activated, and circulating 'innate' immune cells (neutrophils, macrophages) enter the brain.

CNS inflammation is an important determinant of the outcome after stroke. However, it is also critically involved in other forms of both acute and chronic CNS damage and disease. While inflammation is the body's protective response that helps to clear damaged cells and invaders, it can also be harmful. Inflammation can kill or compromise brain cells (neurons, glial cells, endogenous stem cells) and transplanted stem cells. For many years, the global effort to identify targets and drugs to reduce stroke damage focused on early neurotoxicity. Unfortunately, treatments that looked promising in animal studies proved ineffective in clinical trials, partly because they targeted early events that occurred before patients could receive medical attention. This failure has prompted my lab and others to re-focus on the secondary injury phase. This phase corresponds with a prominent inflammatory response that is delayed (hours) and prolonged (days to weeks), and is thus temporally amenable to treatment.

This project focuses on the role of inflammation in damage to myelinated axons and myelin-forming oligodendrocytes (white matter), which contributes to neurological deficits after acute CNS injury, including stroke. Stereotaxic surgery is used to induce an ischemic or hemorrhagic stroke. Damage and inflammation are assessed using histology, immunohistochemistry, confocal microscopy, and high-throughput mRNA and protein analyses. Anti-inflammatory treatments will be tested *in vivo*.

**Project 2. Regulation of microglia functions by the cell activation state**

Microglia, the immune cells of the brain, serve a wide variety of functions, from debris clearance to secretion of growth factors to release of potentially toxic molecules. In the healthy adult brain, microglia are in a 'resting' (actually, 'sensing') state, but become activated by 'danger' signals following brain injury. Because activated microglia perform functions that potentially exacerbate damage or conversely, aid in repair; it is essential to conduct mechanistic studies *in vitro*. Our research is focused on defining molecular targets and testing approaches to controlling these cells, with the goal of promoting their beneficial actions and minimizing their harmful functions.

When microglia respond to damage-induced ('danger') signals, their activation is complex but displays several well-defined states. Cellular functions of activated microglia include proliferation, migration, neurotoxicity, phagocytosis, and production of inflammatory mediators (cytokines, chemokines, reactive oxygen and nitrogen species, growth and repair factors) that affect themselves and other brain cells. This project seeks to answer the crucial questions: What are they producing? What are they doing? How can we control them? In this project, we isolate several brain cell types (microglia, neurons, oligodendrocytes, astrocytes), assess all of the cell functions listed above using *in vitro* assays, cytochemistry, and confocal imaging. We will also conduct laser capture and FACS from brain tissue after stroke, and use high-throughput molecular and proteomics approaches.

**Project 3. Ion channel expression, regulation and roles in microglia**

In our search for ways to control microglial activation and its associated functions, we have been particularly interested in ion channels, for several reasons. We and others have already found that specific ion channels contribute to microglia functions; channels are expressed on the cell surface where they can be readily targeted by drugs and modifiers; many selective blockers and activators have been developed; and there has been great success with using ion-channel modulators in the clinic for other tissues, organs and diseases.

The main channels we are currently focusing on are a $Ca^{2+}$-activated $K^+$ channel (KCa3.1; also called *KCNN4*, SK4, IK1), the voltage-gated Kv1.3 channels and the non-selective TRPM channels. Based on numerous *in vitro* studies and *in vivo* work from my lab and others, KCa3.1, Kv1.3 and TRPM7 are considered promising targets for controlling inflammation in several CNS disorders, including stroke.

In this project, we routinely isolate microglia from young and adult rats and mice, and will now also isolate them from rodents and humans after stroke (human work is in collaboration with NIH and others). Technical approaches include patch-clamp electrophysiology (whole cell and single-channels), cellular $Ca^{2+}$ imaging, and fluorescence plate-reader assays to define channel roles in microglial functions.