PERSPECTIVE

Exploring and exploiting unique properties of the hippocampal dentate gyrus for post-stroke therapy: astrocytes link ischemic resistance with neurogenic potential

Acute cerebral ischemia can occur secondary to embolism, cardiac arrest, hemorrhage, traumatic brain injury, edema, vascular compression, or any physiologic condition resulting in low cardiac output state. Survivors of cerebral ischemic events frequently suffer from profound disability, accounting for > $70 billion in 2010 for treatment of embolic stroke in the US alone. More strikingly, the cost to treat stroke in the US is anticipated to soar to > $180 billion by 2030, according to estimates from the American Heart Association (Ovbiagele et al., 2013). Despite hundreds of promising pre-clinical studies in animal models, effective clinical treatments for the most common forms of cerebral ischemia remain limited: standard treatment for embolic stroke is restricted to thrombolytic therapy or clot retrieval during a narrow therapeutic window, while post-resuscitation treatment following cardiac arrest remains the (controversial) application of mild hypothermia. The massive failure to translate experimental findings to the development of any successful application of mild hypothermia. The massive failure to translate experimental findings to the development of any successful application of mild hypothermia. The massive failure to translate experimental findings to the development of any successful application of mild hypothermia. The massive failure to translate experimental findings to the development of any successful application of mild hypothermia.

Acute cerebral ischemia, neurogenic potential in the adult mammalian brain that maintain active neurogenesis, and cerebral ischemia has been shown to stimulate adult neurogenic activity in both of these regions (Li et al., 2010). Appropriately, targeting the neurogenic niche has been a recent focus for the development of novel post-injury interventions to augment post-injury neurogenesis and improve long-term neurobehavioral outcome following cerebral ischemia (Marlier et al., 2015). Interestingly, neurons in the hippocampal DG subregion are also known to have a relatively higher ischemic resistance versus neurons in the adjacent cornu ammonis 1 (CA1) subregion. Delineating the mechanisms that determine observed differences in both neurogenic potential and sensitivity to ischemia between the CA1 and DG hippocampal subregions might provide new avenues in the development of post-injury therapies to improve long-term neurobehavioral outcome in survivors of all types of cerebral ischemia. Subregional analyses have demonstrated differences in neuronal gene expression between DG and CA1 (Lein et al., 2004), including differences in signal transduction pathways, transcription factors, calcium binding proteins, and genes maintaining a unique extracellular milieu. Most recently (Stary et al., 2016), we have observed that astrocytes, the most numerous type of cell in the brain and critical regulators of neuronal homeostasis, may also account for some of these observed subregional differences.

Astrocytes play many critical roles in supporting normal neuronal functioning, including maintaining ionic balance and modulating neurotransmission (Clarke and Barres, 2013), and astrocyte homeostasis is tightly coupled to neuronal cell fate. Astrocytes provide protection to neurons by a number of mitochondrial-associated mechanisms, including buffering excessive reactive oxygen species (ROS), maintaining calcium (Ca²⁺) homeostasis, and providing metabolic substrate and ATP to neurons. Astrocytes may also regulate neuronal homeostasis and the neuronal bioenergetic response to injury by direct transfer of mitochondria from astrocytes to neurons. Following cerebral ischemia, astrocytes become activated and have the capacity to promote neurogenesis.

Figure 1 Astrocytes regulate neuronal survival and neurogenesis. Astrocytes provide protection to neurons by a number of mitochondrial-associated mechanisms, including buffering excessive reactive oxygen species (ROS), maintaining calcium (Ca²⁺) homeostasis, and providing metabolic substrate and ATP to neurons. Astrocytes may also regulate neuronal homeostasis and the neuronal bioenergetic response to injury by direct transfer of mitochondria from astrocytes to neurons. Following cerebral ischemia, astrocytes become activated and have the capacity to promote neurogenesis.

Figure 2 miR-29a targets voltage dependent anion channel-1 (VDAC1). (A) miR-29a biogenesis begins in the nucleus with genomic transcription of pri-miR-29a (1). Drosha-mediated cleavage results in pre-miR-29a, which is then exported to the cytosol by Exportin-5 and processed to the final mature miR-29a form by Dicer (2). In the cytosol, miR-29a is then free to interact with the 3’ untranslated region of VDAC1 messenger RNA (3). miR-29a/VDAC1 complexes are then targeted by the RNA-induced silencing complex (RISC) for either miR-29a mRNA degradation or translational silencing, depending on the degree of miR/mRNA binding complementarity. (B) A decrease in expression of miR-29a in response to injury in astrocytes results in an increase in VDAC1, inducing astrocyte dysfunction, which can contribute to neuronal cell death and ultimately impair long-term, post-injury neurogenesis. Astrocytes in the cornu ammonis-1 (CA1) hippocampal subregion have a greater decrease in miR-29a than astrocytes from the dentate gyrus (DG), in part explaining the select resistance of DG neurons to cerebral ischemia.
following ischemia-reperfusion (Figure 1) by buffering reactive oxygen species, maintaining Ca\(^{2+}\) homeostasis and ensuring adequate neuronal energy stores (Nedergaard and Dirnagl, 2005; Ouyang et al., 2014). Intriguingly, Hawakawa et al. (2016) recently demonstrated that astrocytes are also capable of direct transfer of functional mitochondria to neurons, and that suppressing this process worsens injury following cerebral ischemia. Astrocyte “activation” and glial scar formation post-injury have traditionally been considered detrimental to stroke recovery, but reactive astrocytes have more recently been demonstrated to have the capacity to promote neuroplasticity (Figure 1) via secretion of neurotrophic factors, cholesterol and thrombospondins (Liu and Chopp, 2015). Previous observations (Ouyang et al., 2007) suggest that astrocytes within the hippocampus CA1 are more sensitive to ischemic injury, and with a higher degree of mitochondrial dysfunction compared to DG astrocytes, and that disruption of mitochondrial homeostasis in local astrocytes following cerebral ischemia contributes to CA1 neuronal cell death (Xu et al., 2010; Ouyang et al., 2013). Together these observations demonstrate a potential role for mitochondria in neuronal-astrocyte communication, and position astrocytes as central for maintenance of neuronal metabolism and bioenergetics in response to cell stress and recovery from injury. Therefore, long-term post-injury gene therapy strategies aimed at maintaining astrocyte mitochondrial function may provide a novel approach to accelerate and magnify recovery from ischemic brain injury.

MicroRNAs (miRs) are an endogenous class of small, non-coding RNAs that modulate gene expression by binding to the 3’ untranslated region (UTR) of target genes and destabilizing or inhibiting their translation (Figure 2A). microRNAs are an attractive therapeutic candidate for long-term modulation of neuronal and glial mitochondrial function as endogenous microRNA levels can be easily altered with chemically modified non-coding RNAs that modulate gene expression by binding to the 3' untranslated region (UTR) of target genes and destabilizing or inhibiting their translation. These microRNAs regulate cell survival in astrocytes from cornu ammonis-1 and dentate granule cell survival. Most recently (Stary et al., 2016), we have demonstrated that astrocytes selectively cultured from the hippocampal DG subregion had a greater resistance to injury versus astrocytes cultured from the CA1, recapitulating in vivo observations of selective vulnerability of CA1 neurons to ischemia. This difference was due, at least in part, to a differential expression pattern in miR-29a/VDAC1 between subregions (Figure 2B), and was eliminated by blocking the interaction of miR-29a with the VDAC1 3’UTR. These findings suggest that microRNA biogenesis in astrocytes may provide a critical nexus between neuronal survival and post-injury neurogenic potential, and explain in part the unique properties of the DG subregion. Further exploring the differences between these two hippocampal subregions in baseline astrocyte physiology, and function in response to injury, may give rise to effective novel therapies for long-term recovery and repair of the injured brain following ischemic injury.