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DOES STROKE CAUSE EXPANSION OF TRANSIENT OLIGODENDROCYTES?

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DOES STROKE CAUSE EXPANSION OF TRANSIENT OLIGODENDROCYTES?

By

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A capstone project submitted for Graduation with University Honors

May 06, 2022

University Honors
University of California, Riverside

APPROVED

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ABSTRACT

Stroke is the fifth leading cause of death and the leading cause of adult disability in the United States. Our lab and others have shown that exogenous Neuregulin-1 (NRG-1) decreases acute neuronal death and neuroinflammation after ischemic injury. Clinically, NRG-1 showed significant efficacy for improving cardiac function in patients with heart failure.

Through our studies, we hope to further characterize neuro-regeneration by NRG-1 using an innovative mouse model of ischemic stroke. Photo-thrombosis is a model which targets the middle cerebral artery in rodents, produces a fixed thrombus, and initiates a focused cortical stroke. This non-invasive model creates an ideal basis for stroke studies through its minimally invasive means and in vivo design.

Oligodendrocytes play an important role in traumatic brain injury regeneration as their myelination around neurons are characteristic for important neuronal processes. Oligodendrocytes have been studied extensively in Multiple Sclerosis models but not yet characterized in a photothrombotic stroke model. In studying oligodendrocytes, we can better understand the regenerative processes that might occur after stroke.

Our research question looks specifically at a transient population of oligodendrocytes which are differentiated by BCAS1 positivity through histology. We anticipate that there will be a greater amount of BCAS1 positive cells after photothrombotic stroke. In characterizing the proliferation of these transient oligodendrocytes before and after stroke injury we can understand where proliferation occurs. Understanding the expansion of these transient populations of oligodendrocytes will serve as the basis to understand the role that NRG-1 might play in regeneration and oligodendrocyte maturation.

ACKNOWLEDGEMENTS

I would like to begin by thanking my amazing mentor, teacher, and Principal Investigator, Dr. Byron Ford, who has immensely helped me grow as an undergraduate researcher throughout my 4 years at UC Riverside. Dr. Ford's dedication towards helping me achieve my goals has greatly impacted my undergraduate research experience for the better. I attribute much of my accomplishments to the mentorship, wisdom, and learning that Dr. Ford has given me throughout my time in his lab.

I would like to additionally thank my graduate student mentor Jessica Noll who taught me much of the foundation that I needed to develop my own project and be a successful researcher. Her time towards my research growth has truly influenced my ability to achieve all that I could in the lab.

The continued support of my family and friends also does not go unnoticed as they have been my main support towards my academic achievements. I am grateful that I have been given such an amazing support network which had motivated me to complete my scholarly endeavors.

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INTRODUCTION

Stroke remains a devastating disease in modern healthcare as nearly 800,000 people in the United States experience stroke yearly. Of all the strokes that occur in the United States about 600,000 of them are new cases experienced by patients who have never previously had a stroke (Centers for Disease Control and Prevention 2018). Stroke is categorized into two major classifications: Hemorrhagic and Ischemic. Hemorrhagic stroke results from a rupture of an artery within the brain and results in subsequent bleeding into the surrounding tissue. Ischemic stroke is when there is an occlusion or blockage present in the artery of the brain and prevents the transport of essential oxygen and nutrients to the surrounding tissue area. A 2017 American Heart Association Report indicates that there were 11.6 million events of ischemic stroke and 5.3 million events of hemorrhagic stroke worldwide in 2013 (Benjamin 2017). The American Heart Association further reports that 80% of strokes are ischemic with a global mortality of 3.3 million in 2013 (Benjamin 2017). Ischemic strokes can be detrimental and lead to further hemorrhage in the brain causing greater damage and devastation to the individual experiencing the stroke.

During ischemic injury, a significant and prolonged inflammatory response is initiated which begins at the location of the arterial blockage defined as the core. Beginning at this core damage, the early stages of cell death trigger an inflammatory response which promotes neuronal damage by apoptotic mechanisms (C. Iadecola et al. 2001). The core damage progression defines a new area known as the peri-infarct region where there is a significant amount of penumbral damage, but the tissue can still be salvaged (Dirnagl et al. 1999). This is because blood supply is restricted but not completely interrupted creating an area of compromised but still surviving cells (Dirnagl et al. 1999). Several mechanisms and regenerative components within the brain, such as oligodendrocyte microglia in this study, play an important role in the regeneration of the

salvageable peri-infarct area. It is important to study these regenerative processes and the specific cells which aid in this region as they might yield significant results towards tissue salvageability.

When looking at regeneration within the brain following stroke injury and potential therapeutics to aid in the regenerative processes, NRG-1 is a potential therapeutic that has shown significant results towards contributing to regeneration and minimizing ischemic stroke damage shortly after the onset of a stroke. Neuregulin's are a family of structurally related proteins that have diverse functions in the nervous system and have shown promise in early pre-clinical studies of reducing ischemia. NRG-1 belongs to a family of neuroprotective and anti-inflammatory growth factors (Falls et al. 1993). Considering Neuregulin's are endogenously produced, we know that they are a safe compound that could be exogenously introduced without devastating consequences. NRG-1 has therefor been studied in exogenous administration shortly after stroke injury. NRG-1 acts through erbB4 tyrosine kinase receptors in which it causes a multitude of downstream signaling pathways which ultimately contribute to the anti-inflammatory response produced in injury (Meyer et al. 1997). Working with these erbB4 tyrosine kinase receptors, NRG-1 has been shown to cross both the blood-spinal cord barrier and the Blood Brain Barrier via receptor mediated transport (Pan et al. 2004). This is significant as it is important towards treating patients clinically through exogenous treatment of NRG-1. When looking at exogenous neuroprotection by NRG-1, our lab and others have shown that NRG-1 was able to reduce ischemia by upwards of 90% with a therapeutic window of greater than 12 hours (Noll et al. 2019). This can be

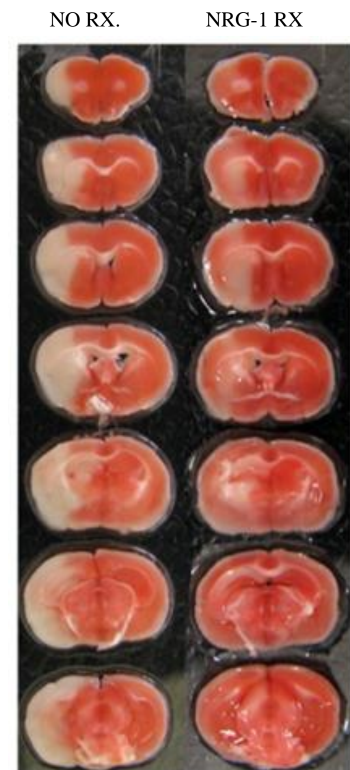

Figure 1

Mice on the right panel were treated with NRG-1 directly following Middle Cerebral Artery Occlusion (MCAO). MCAO was followed by 23 hour reperfusion. NRG-1 reduced total infarct volume by 67%. (Noll et al. 2019)

visualized in Figure 1 which shows a 2,3,5-Triphenyltetrazolium Chloride (TTC) stain of metabolically active/inactive tissue with and without NRG-1 treatment in a mouse injury stroke model. TTC stains metabolically active tissue red and metabolically inactive tissue white. It is evident that exogenous NRG-1 treatment (on the right panel) minimizes the total ischemic area after a stroke takes place. When quantified it can be concluded that exogenous treatment of NRG-1 significantly reduces cerebral ischemia and therefore should be further characterized to understand the role that it could play in stroke treatment and regeneration. (Noll et al. 2019)

This study focuses on a specific set of microglial cells in the context of regeneration and NRG-1 treatment before and after stroke injury. When studying regenerative processes after a stroke, there are several cell types that aid in the pathways towards recovery of damaged tissue in the brain within the peri-infarct region. Oligodendrocytes are an important one of these cells. Oligodendrocytes are an important microglial cell which serves to support insulation to axons in the central nervous system and therefor promote regenerative processes within the brain. Oligodendrocytes wrap their plasma membrane around axons to generate myelin and provide electrical insultation to increase the speed of nerve conduction (Snaidero et al. 2019). Figure 2 outlines the typical morphology of an oligodendrocyte cell representing the way in which the plasma membrane of the oligodendrocyte is interconnected with the axon of neurons to promote neuronal function and electrical insulation. Considering that the two cell types are greatly interconnected and conduct important regenerative processes, further characterizing these cells in a stroke model is important as it can lead to the development of understanding NRG-1 as a therapeutic for patients who experience a stroke.

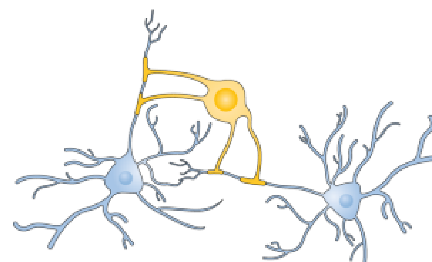

Figure 2

Outlined in yellow marks a mature oligodendrocyte cell wrapping its plasma membrane around the axon of a mature adult Neuron in Blue. Image Credit: Philip Patenall/Springer Nature Limited

Damage to oligodendrocyte cells could have serious implications on Central Nervous System function and nerve conduction.

Regenerative processes of oligodendrocytes have been studied extensively in Multiple Sclerosis models. Oligodendrocyte precursor cells and mature oligodendrocytes both respond to injury associated signals to induce oligodendrocyte expansion, differentiation, axonal contact, and myelin regeneration (Chamberlain et al. 2016). These studies explore regions and cells of interest through studying demyelinating disorders in the Central nervous System such as Multiple Sclerosis. These signaling pathways of myelin regeneration have yet to be characterized in an adequate mouse model for stroke. Our study aims to look at a transient population (after oligodendrocyte precursors and before total oligodendrocyte maturation) of these regenerative cells and characterize them in a stroke model.

Oligodendrocyte cell populations are comprised of three different stages within the developmental process of oligodendrocytes: oligodendrocyte progenitor cells, myelinating oligodendrocytes, and mature cells. Our study looks specifically at the transient population of myelinating oligodendrocytes that are intermediary between pre-mature oligodendrocytes and mature oligodendrocytes. This transient population is undergoing adaptive myelination towards maturation and can be differentiated from the other oligodendrocytes through expression of breast carcinoma amplified sequence 1 (BCAS1). BCAS1+ cells represent a population of oligodendrocytes that segregate from mature oligodendrocytes and their progenitors in humans (Fard et al. 2017). BCAS1+ cells will be the main cell type that will be characterized for this study within an ischemic stroke model. BCAS1+ cells are of interest for this study as the expansive properties of these oligodendrocytes have not yet been characterized and there is unfamiliarity in

the field in the way this transient population of oligodendrocytes behave. Figure 3 outlines the differentiation process from oligodendrocyte Precursor Cells (OPCs) to mature oligodendrocytes (mOLGs). Specific markers unique to each type of cells are identified below the cell type. NG2 glial markers are characteristic of OPCs, BCAS1 markers identify the transient population of pre-mature OLGs, and polyposis coli clone 1 (CC1) are characteristic of pre-mature OLGs and mature oligodendrocytes. In studying these markers through Immunohistochemistry in a stroke model and analyzing their expansive properties, we can better conclude what is happening to this population of oligodendrocytes after a stroke. We hope to understand how expansion is happening whether that be through proliferation on the OPCs which then turn into mOLGs or if the BCAS+ pre-mOLGs cells themselves are proliferating.

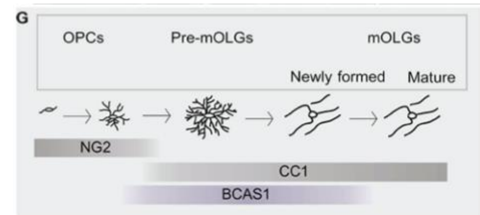

Figure 1

Differentiation of oligodendrocytes as they progress from oligodendrocyte progenitor cells to mature oligodendrocytes. Diagram below shows the histology staining that would be used to visualize each population of oligodendrocytes through their developmental stages. (Fard et al. 2017)

In characterizing this transient populations of oligodendrocyte cells that are identified through BCAS positivity, the role that oligodendrocyte microglia play in neuro-regeneration is stroke can become familiarized. This will set the foundation to understand the ways in which NRG-1 administration into the system can possibly promote regenerative pathways, influence the survival, and promote expansion of this transient population of oligodendrocytes.

IMPORTANCE

Stroke is the 5th leading cause of death in the United States and the 1st leading cause of adult disability (Lloyd-Jones et al. 2010). After a stroke several physical, memory, and personality changes might develop long term consequences in a patient depending on the severity of a stroke. Considering the frequency and severity of a stroke, its ability to cause long term brain damage and disability, it is important to identify new approaches to minimizing risk of stroke after one has occurred. Our study is an important early model that supports the foundation of research in Oligodendrocyte microglia characterization in a mouse stroke model. This pre-clinical research holds translational value towards developing a therapeutic such as NRG-1 that can minimize the detriment that stroke brings about.

NRG-1 has showed significant efficacy for improving cardiac function in chronic heart failure (Gao et al. 2020). NRG1 is currently in phase III human clinical trials for heart failure in China and entering clinical trials in the United States (ClinicalTrials.gov). NRG1 is proving to be a treatment option that is safe in patients that is neuroprotective, anti-inflammatory, and neuro-regenerative for multiple different cell types. It is for this reason that exploring these specific cell types that NRG1 can act on is important. This study looks at Oligodendrocyte cells in the model and will lead to characterization of a transient population of Oligodendrocytes in stroke.

Importance of this study can further be established as there are currently only two viable treatment options for stroke today: Tissue Plasminogen Activator (t-PA) and Endovascular Thrombectomy. Both current treatment options have a multitude of downfalls when considering clinical administration.

Tissue Plasminogen Activator (t-PA) is the main approved drug treatment for ischemic stroke. t-PA is a clot dissolving medicine that can enter the circulatory system intravenously and work towards dissolving and/or minimizing the blockage in the brain (Lapchak et al. 2013). Unfortunately, t-PA has a limited time window for clinical use in patients where only about 3-5% of stroke patients who arrive to a hospital will qualify for treatment by t-PA (Fisher et al. 2009). Additionally, tPA does not show any neuroprotective or neuro-regenerative value as its sole function is to dissolve the occlusion in the brain. After a four-hour time window, t-PA becomes toxic and is therefore no longer a viable treatment option. This means that if a patient experiences a stroke while they are asleep and wake up after the therapeutic time window for this drug, they are no longer eligible for treatment. If t-PA is not administered in a timely manner, the severity of a stroke will only worsen with time. The stroke that a patient is experiencing will continue to expand unless another intervention or treatment option is provided.

The second, and last, treatment option for ischemic stroke is a surgical technique titled endovascular thrombectomy. This procedure mechanically removes the arterial blood clot or blockage by using a probe that traps the clot in a stent which is then used to pull the clot out of the brain (Feng et al. 2021). Endovascular thrombectomy is a relatively new and expensive medical technology that is highly inaccessible in rural areas and hospitals. Considering lack of healthcare professionals who can perform the surgery and the inequities in healthcare access across the nation, endovascular thrombectomy stroke treatment is limited to a very small population of patients who undergo a stroke. Additionally, through the means of this invasive procedure, there is greater risk for mechanical error and further hemorrhage/bleeding in the brain. It is for these reasons that this procedure might not be the preferred means for removal of an arterial blockage within the brain following ischemia.

Considering the downfalls of both t-PA and endovascular thrombectomy, the significance of studying new viable treatment options for stroke can be made clear. This is where studying neuro-regeneration and neuroprotection in a stroke model with new therapeutics such as NRG-1 proves importance. Studying NRG-1 treatment and the mechanisms by which neuro-regeneration can occur after a stroke can lead us to new discoveries exploring downstream pathways involved in recovery of compromised tissue area. Therefore, oligodendrocytes are important to study and characterize as they have not yet been explored in an ischemic stroke model system. This study serves as the foundation of microglia research that will build upon the evidence that NRG-1 is a viable treatment option for stroke patients with protective and regenerative value.

METHODS

All animals in these studies were treated humanely and were approved by International Animal Care and Use Committee (IACUC) of University of California, Riverside prior to initiation of these experiments.

Overview

When looking at an ischemic stroke model to mimic brain injury in rodents, there are a multitude of factors that are associated with development of a good model. These factors include minimal invasiveness, proper treatment and care of animals, and as closely resembling a true stroke as possible. Our lab utilizes two major surgeries (which include these factors) to create an occlusion in the rodent brain: Transient Middle Cerebral Artery Occlusion (MCAO) and Photothrombotic Middle Cerebral Artery Occlusion (PtMCAO). Through both surgeries, a disruption of blood flow is created in the Middle Cerebral Artery for a fixed period and then removed allowing for reperfusion of the blood. Traditional MCAO surgery is an invasive procedure in which there is significant room for further hemorrhage and mechanical error. It is for this reason that our lab has shifted our primary ischemia model to PtMCAO. PtMCAO utilizes a photoreactive dye that is injected into the rodent intra-peritoneally. The rodent skull is then exposed, and a laser is positioned to the point where the occlusion would form. Upon reaction of the photo-reactive dye and the laser, an occlusion is formed, and a fixed thrombus can be observed in the Middle Cerebral Artery. Considering that this procedure is much less invasive and provides successful results in creating a stroke model, it is the primary ischemia model that is utilized by our lab and throughout this study.

Both MCAO and ptMCAO are surgeries that closely mimic ischemic stroke damage as there is an occlusion in the brain which is then removed. Following surgery, the rodent brain was removed and fixed in paraformaldehyde for studies. Brain tissues were then sectioned and stained for Histology. Immunohistochemistry experiments were performed to visualize localization of cells of interest and quantify signal of identifiable markers. Results were then analyzed, and oligodendrocyte cell (transient BCAS+ cells) expansion could be characterized before/after stroke within this model of ischemia.

Tissues utilized for this experiment were used in parallel to surgeries conducted by Dr. Jessica. The following protocols were adapted from Dr. Noll's Graduate Dissertation, "Spatiotemporal Neuroprotection and Early Regeneration after Ischemic Stroke with Neuregulin-1 Treatment" (Noll 2021):

<u>Photothrombotic Middle Cerebral Artery Occlusion</u>

Animals were subjected to left photothrombotic MCA occlusion (MCAO). Mice were anesthetized with 2% isoflurane and circulating air and maintained anesthetized during the procedure via a modified gas tubing nose connection. Eye lubricant was applied to protect the eyes and body temperature was maintained via a heating pad placed underneath the mice during surgery at 37°C. MCAO was performed in an adapted accordance to Zhong et al. (Zhong 2010). Rose Bengal was injected intraperitoneally at 10 mL/g and allowed to incubate for 8 minutes. The animal was stabilized in a stereotaxic instrument and the scalp hair was removed. The scalp was disinfected with iodine and ethanol, then followed by a midline skin incision to expose the skull above the left sensorimotor cortex. A 2-mm diameter focal green laser (520nm) was directed at 2-

mm lateral left. Laser irradiation occurred for 20 minutes at 10 mW at location Bregma 0 (Figure 4). After irradiation, the midline incision was sealed with Vetbond glue followed by triple antibiotic ointment. Mice were then placed into a 37°C incubation chamber for 20-30 minutes to recover. Sham control animals included two groups: 1) Rose Bengal injection without laser irradiation and 2) laser irradiation without Rose Bengal injection. Mice were sacrificed at 3 days post-ischemia (dpi).

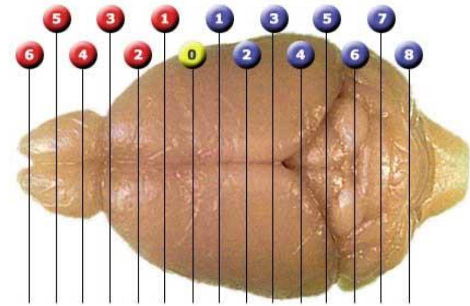

Figure 4

Outline showing Bregma 0 localization where the coronal and sagittal sutures meet. Occlusions was initiated at the left sensorimotor cortex at Bregma 0. Immunohistochemistry stains were conducted on sections from tissue at Bregma +0.20mm.

Histology and Immunohistochemistry

After MCAO, mice were deeply anesthetized with 2% isoflurane and perfused transcardially with saline followed by cold 4% paraformaldehyde (PFA) solution. Brains were quickly removed and maintained in 4% PFA for 24 hours. Brains in preparation for histological and immunohistochemical analysis were quickly removed after transcardial perfusion and maintained in 4% PFA for 24 hours before being cryoprotected in 30% sucrose. The brains were then flash frozen and stored at – 80°C until sectioning. Coronal sections of 12–15 µm thickness were cryosectioned and mounted on slides which were then stored at −80°C until further processed. Sections near bregma +0.20 mm (Figure 4) localized at the left sensorimotor cortex were utilized for this study. This region of study is near the infarct and should have adequate peri-infarct tissue surrounding the core lesion for analysis.

For immunohistochemical studies, sections were dried at room temperature for 30 minutes. After rinsing with 0.01M PBS, sections were blocked in PBS containing 5% normal donkey serum 23 and 0.1% Triton X-100 for 1-2 hours at room temperature, rinsed with PBS/0.2% Tween-20 (PBST) and then incubated overnight at 4°C with primary antibodies of monoclonal mouse anti-NaBC1 (BCA1) (1:300, Cat#SC-136342, Santa Cruz Biotech) and polyclonal rabbit anti-Olig2 (1:700, Cat#AB9610, EMD Millipore Corp.). Sections were washed 3 times with PBST, incubated with respective AlexaFluor488-conjugated donkey anti-mouse IgG antibody (1:400, Cat#715-545-150, Jackson ImmunoResearch Laboratory) or AlexaFluor594-conjugated donkey anti-rabbit IgG antibody (1:400, Cat#711-585- 152, Jackson ImmunoResearch Laboratory) for 1 hour at room temperature. Sections were then rinsed 4 times with PBST before mounting with DAPI-Fluoromount-G (Cat#OB010020, Fisher Scientific).

Immunohistochemical Quantification

Immunohistology images of Olig2 and BCAS1 (NaBC1) were captured at approximately bregma +0.20 mm at four different regions of interest within the coronal section: M1, S1ULp, S2 and PiR. The same four regions of interest were captured on the contralateral cortex with corresponding tissue sections. These regions were chosen as they show a representative sample of the damaged core and the surrounding per-infarct tissue area. The contralateral hemisphere was comprised of undamaged tissue area that would be representative of tissue prior to a stroke that has taken place. The contralateral hemisphere was used for comparative analysis to the ipsilateral damage. In quantifying the signal present within the same regions on both the ipsilateral damage and respective contralateral tissue area, a comparison can be made to mimic damage before and

after stroke injury. All picture properties of immunohistochemistry imaging were obtained and ensured for consistency. Fluorescent signal was maintained consistent with defined image exposure for both cell and nucleus imaging. Cell images and corresponding nuclei images were overlayed in "WaterMarker" image editing software for visualization. Olig2 and BCAS1 count was conducted randomly and blindly by a third party with consideration for specific characteristics of what should be included in the final count. All cells that show a blue DAPI nucleus and green/red surrounding fluorescence (Alexa488 or Alexa594) should be counted as a cell. Additionally, all cells on image borders were included in the final cell count. Final counts were collected, and peri-infarct cell data was analyzed with respect to the contralateral.

DISCUSSION AND PREDICTIONS

Oligodendrocyte Immunohistochemical staining of BCAS1+ and Olig2+ was performed in Bregma +0.20mm tissue sections. BCAS1+ staining allows for visualization of signal from all cells representing the intermediary transient population of oligodendrocytes. Olig2+ staining allows for visualization of signal from all oligodendrocyte cells regardless of their stage in the maturation process. Understanding localization of BCAS+ and Olig2+ after a stroke takes place can help us conclude how oligodendrocyte cells might respond to ischemia in a photothrombotic stroke model. We hypothesize that there will be an increase in the number of BCAS+ cells in the peri-infarct ipsilateral region of a stroke as there will be several regenerative processes occurring in this region.

Figure 5 and Figure 6 represent the way in which cell counts were conducted following immunohistochemistry and the processes by which cells were included within the final count. These cell counts were conducted in the ipsilateral side of injury and the unaffected contralateral tissue in the M1, S1ULp, S2 and PiR regions of the brain. Immunohistochemical quantification and visualization provides evidence to prove localization, expansion, and characterization of this transient population of oligodendrocytes.

BCAS1+ localization can be best represented in Figure 7 which outlines the rodent model brain, the core lesion of the stroke, and the oligodendrocytes which show BCAS+ signal. This outline shows us not only that there is significantly more BCAS+ cells on the side of injury but also that BCAS+ cells are most prevalent in the peri-infarct region. Greatest quantities of BCAS cells are in the M1 ipsilateral side of injury which is closest to the stroke core. This shows that this transient population of oligodendrocytes are localized to regions near the injury. When quantifying the cells on the Ipsilateral side of injury to the contralateral unaffected tissue, there is a significantly

greater number of BCAS+ cells overall on the side of injury, as outlined in Figure 9. This can be a result from the increased regenerative processes occurring in this region due to the ischemic damage present nearby. BCAS1+ oligodendrocytes could be assisting in those regenerative processes and aiding in neuronal repair through myelinating properties.

Olig2+ localization can be best represented in Figure 8 which outlines the rodent model brain, the core lesion of the stroke, and all oligodendrocyte populations which show Olig2+ signal. From this outline it can be visualized that oligodendrocytes after stroke injury are significantly more randomized throughout the rodent brain and not localized in the same way that the BCAS+ cells are. When quantifying the amount of Olig2+ signal throughout the 8 regions of interest it can be seen in Figure 10 that there is no localization to the ipsilateral side of injury or the contralateral tissue. Instead, oligodendrocyte cells counts are uniform throughout the tissue entirely in similar quantities even in different regions of interest.

Through this study we were able to prove BCAS+ localization and expansion after ischemic stroke injury in a photothrombotic stroke model. We were able to successfully show that not all oligodendrocyte cell types are localized to the regions of stroke injury, instead it might only be the BCAS+ cells that are. BCAS+ cells might be actively assisting in the myelination of neurons and aiding in the regenerative processes days after s stroke takes place. This is study serves as the foundation for other studies in oligodendrocyte development and regeneration within a stroke model. We hope to introduce NRG-1 into the system to later understand how NRG-1 might promote further BCAS1 expression in this population of oligodendrocytes. In studying this same model with NRG-1 treatment, oligodendrocyte characterization and regenerative studies can help develop NRG-1 as a new therapeutic for stroke patients.

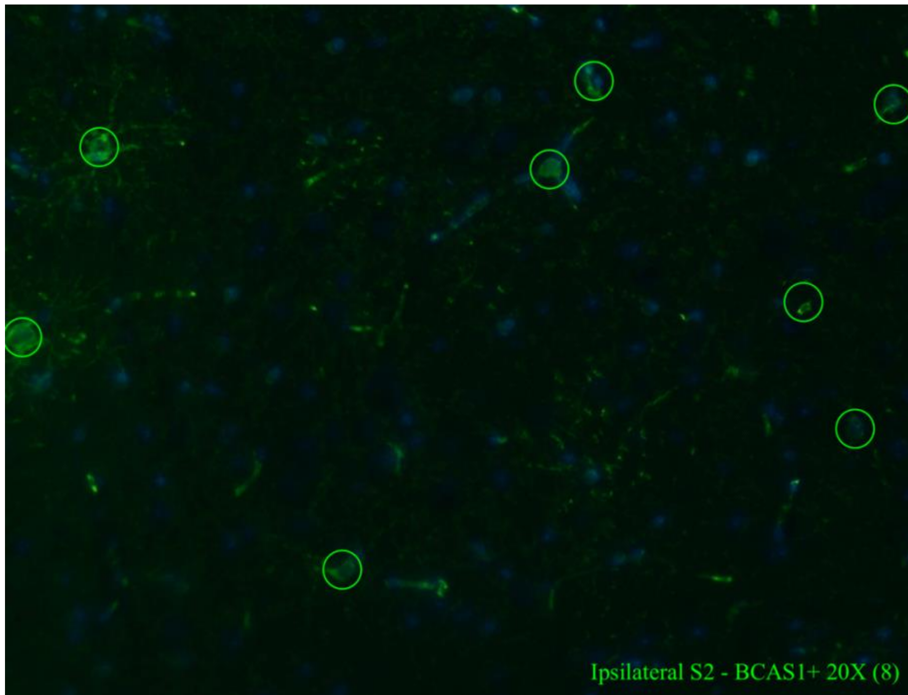

Figure 5

Example Cell count of BCAS1+ cells in tissue area. Cells which show clear oligodendrocyte morphology will be counted towards final cell count. Count also only includes cells with a clear DAPI nucleus signal.

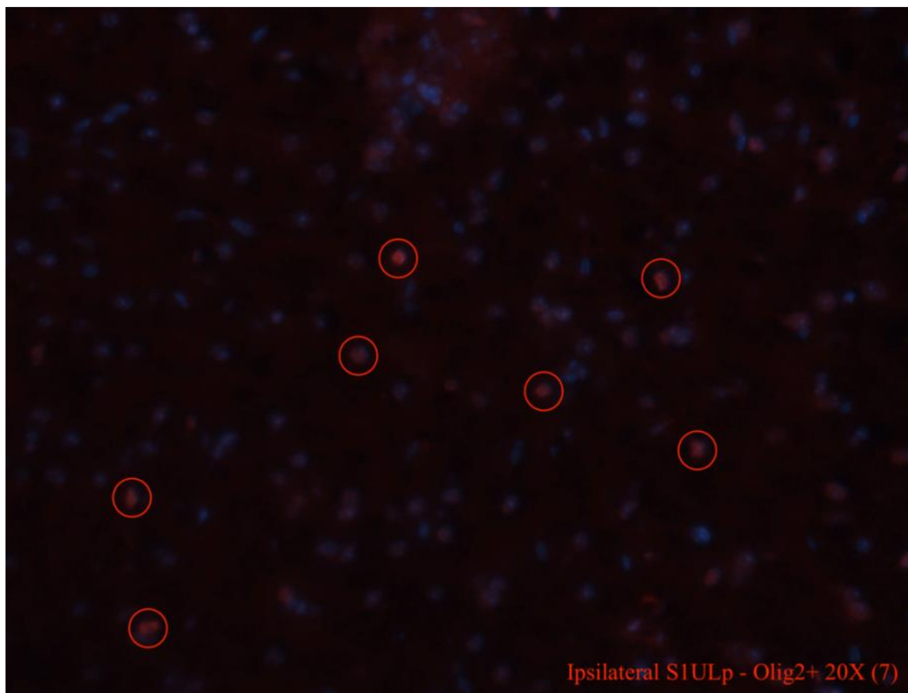

Figure 6

Example Cell count of Olig2+ cells in tissue area. Olig2+ is a nuclear stain (signal present in nucleus of cells) and therefor all significant red fluorescence will be counted towards final cell count.

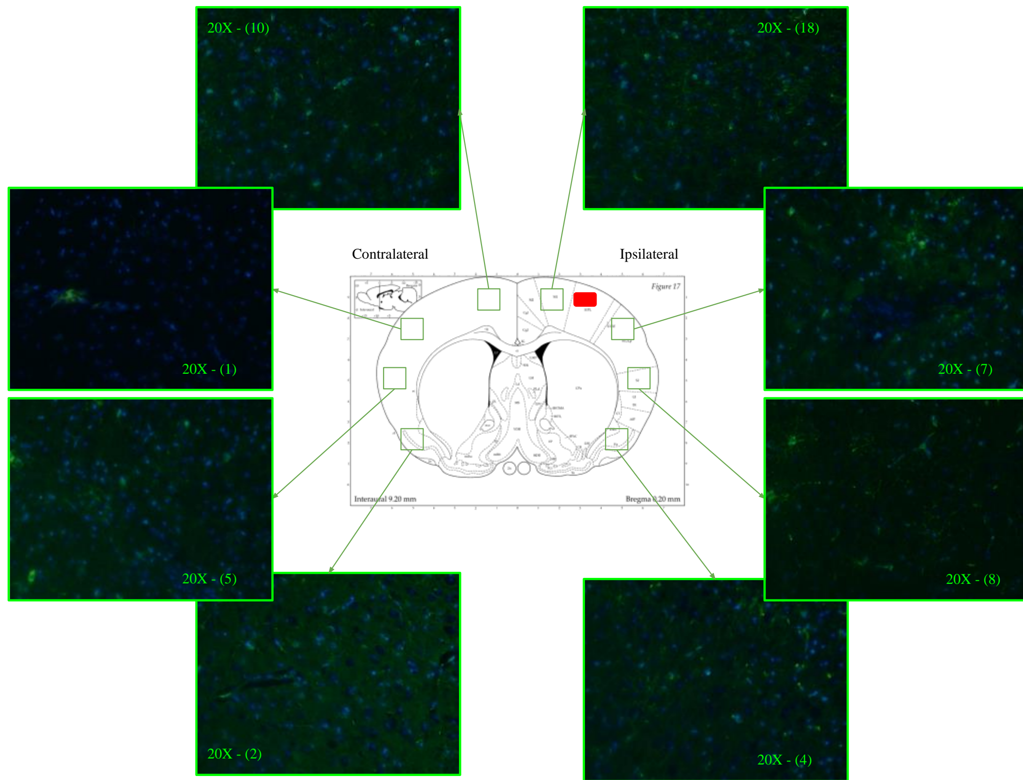

Figure 7 Localization of BCAS+ cells in M1, S1ULp, S2, and PiR tissue area on both sides relative to stroke injury.

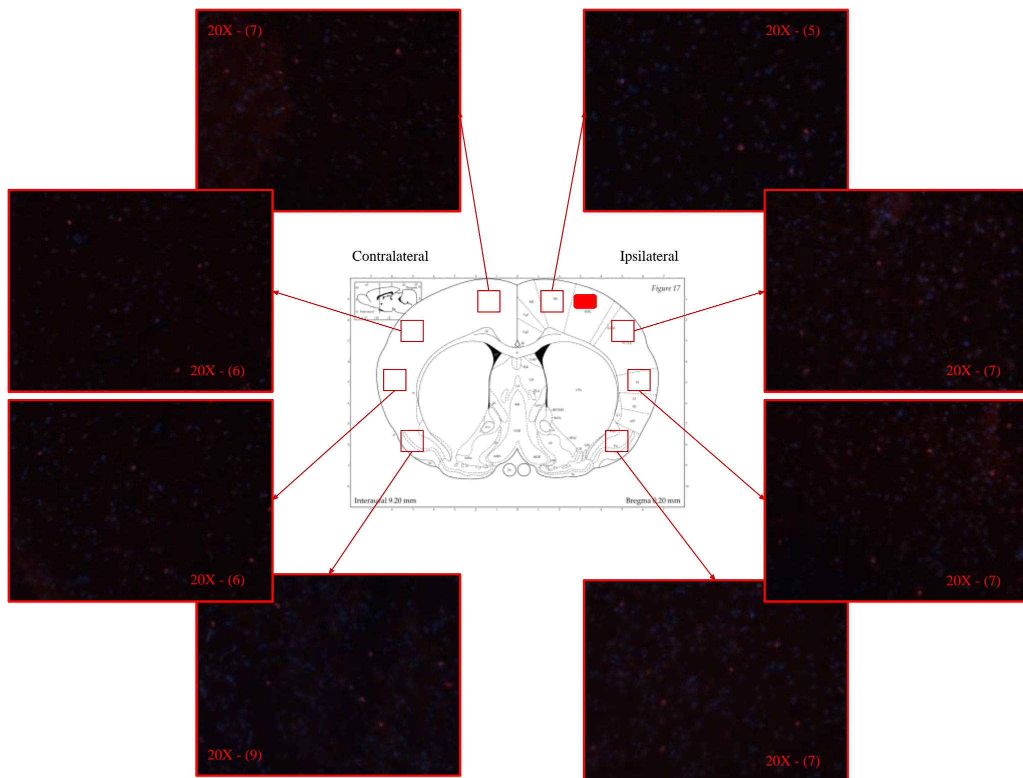

Figure 8 Localization of Olig2+ cells in M1, S1ULp, S2, and PiR tissue area on both sides relative to stroke injury.

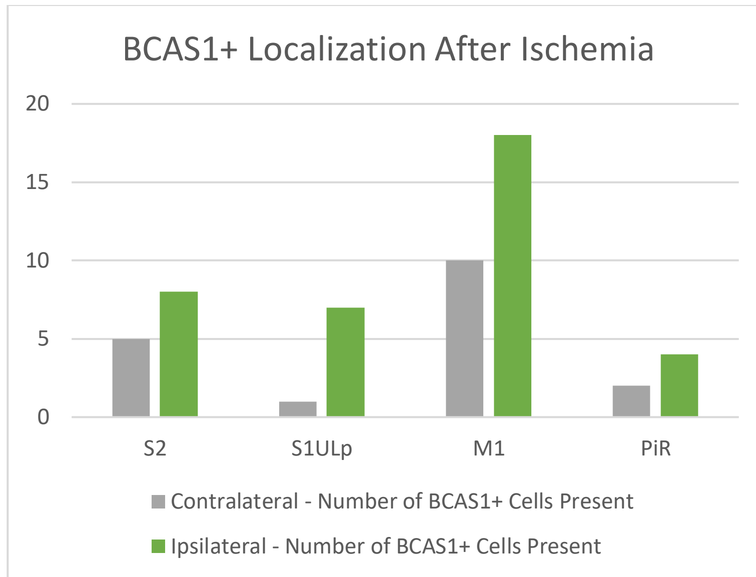

Figure 9

BCAS1+ cells are localized to the Ipsilateral hemisphere (relative to stroke injury) of the brain showing that injury may cause an increase in oligodendrocyte production. Highest quantities of BCAS+ cells are found in the M1 region of the brain which is closest to the core region of the stroke. This shows that BCAS1+ expression is increased on the side of injury when compared to the controlled contralateral cortex.

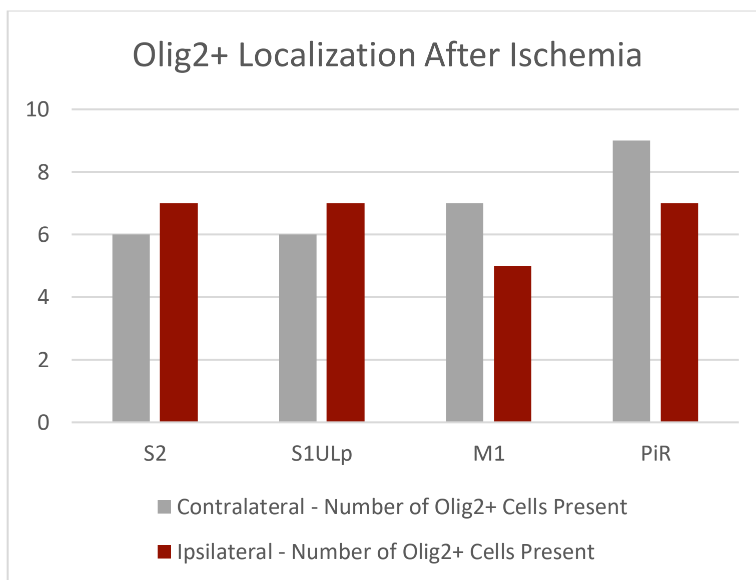

Figure 10

Olig2+ cells are localized in no particularly significant way after stroke injury. Oligodendrocyte quantities are evenly disbursed throughout the brain with no increase or decrease in activation in either the ipsilateral or contralateral regions after a stroke takes place.

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