INVESTIGATING THE RELATIONSHIP BETWEEN ANATOMIC AND METABOLIC CHANGES IN THE MOTOR CORTEX AND UPPER-EXTREMITY HEMIPARESIS IN INDIVIDUALS WITH CHRONIC SUBCORTICAL STROKE

by

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ABSTRACT

Hemiparesis is one of the most prevalent chronic disabilities after stroke, particularly in subcortical stroke. Neuroimaging has provided important morphological insight in to the mechanisms associated with hemiparesis in individuals with stroke. Assessing morphological changes within the primary motor cortex may provide valuable information of the neural events that underlie upper-extremity (UE) hemiparesis in chronic stroke. The purposes of this study were to 1) evaluate anatomical and metabolic changes in the motor cortex, and 2) examine the relationship between anatomical and metabolic changes and hemiparetic arm use in individuals in the chronic stage of stroke recovery. Seventeen individuals with chronic (>6 months) subcortical ischemic stroke and eleven neurologically healthy controls were recruited. Single voxel proton magnetic resonance spectroscopy (H^1MRS) was performed to measure metabolite concentrations of total N-acetylaspartate (tNAA) and glutamate+glutamine (Glx). FreeSurfer software package (http://surfer.nmr.mgh.havard.edu) was used to quantify cortical thickness of the precentral gyrus. Upper-extremity motor performance was assessed using the Wolf Motor Function Test (WMFT) and the Motor Activity Log Quality of Movement scale (MAL-QOM) and upper-extremity motor activity was assessed using activity counts from wrist-mounted accelerometers. Results demonstrated a significant decrease in tNAA and Glx concentration in the hand area of the primary motor cortex in the stroke group, particularly within the ipsilesional hemisphere. Precentral gyrus cortical thickness was also decreased in the ipsilesional hemisphere of the stroke group. Parametric correlation analysis revealed a significant positive correlation between precentral gyrus thickness and tNAA concentration bilaterally. Multivariable regression analyses revealed that, after accounting for age and post-stroke duration, the combination of ipsilesional metabolite concentration (tNAA and Glx) and ipsilesional cortical thickness was associated with hemiparetic UE motor performance, but not UE motor activity in individuals in

the chronic stage of stroke recovery. The observed link between structural and neurochemical changes in the stroke-affected brain and hemiparetic UE motor performance during the chronic phase of recovery may improve the understanding of the underlying neural mechanisms that support motor impairment after stroke.

PREFACE

This thesis contains a research experiment that was conducted by the candidate, Paul W. Jones under the supervision of Dr. Lara A. Boyd with guidance from Drs Janice Eng, Alex MacKay and Martin McKeown. The collection, analysis and writing of the experiment were principally the work of the candidate. This thesis will be submitted for publication as a multi-authored manuscript in peer-reviewed journals. Members of the committee and contributing authors provided support and feedback. Ethical review and approval for this thesis was performed by the UBC Clinical Research Ethics Board.

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LIST OF ABBREVIATIONS

ADL	Activities of Daily Living
ANOVA	Analysis of Variance
Cho	Choline
Cre	Creatine
CSI	Chemical Shift Imaging
FM	Fugl-Meyer assessment of motor recovery after stroke
Glu	Glutamate
Glx	Glutamate+glutamine
H^1 MRS	Proton magnetic resonance spectroscopy
M1	Primary motor cortex
MAL AOU	Motor Activity Log Amount of Use scale
MAL QOM	Motor Activity Log Quality of Movement scale
mI	Myo-inositol
mM	Millimolar concentration
MRI	Magnetic Resonance Imaging
NAA	N-acetylaspartate
Ppm	Parts-per-million
SVS	Single Voxel Spectroscopy
tNAA	N-acetylaspartate + N-acetylaspartylglutamic acid
UE	Upper Extremity
WMFT	Wolf Motor Function Test

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1. INTRODUCTION TO THE PROBLEM

1.1 Stroke Classification

A stroke is a neurological deficit of cerebrovascular cause that persists beyond 24 hours or is interrupted by death within 24 hours. The condition is caused by a rapid loss of blood supply to the brain. This may be due to a blockage of blood vessels (ischemic stroke) or a hemorrhage. Ischemic strokes result from a blockage of blood supply, which may be caused by the narrowing of blood vessels (thrombotic stroke), or a clot, which has traveled from another area of the body (embolic stroke). Hemorrhagic strokes are the result of ruptured blood vessels or abnormal vascular structure. With an absence of blood supply, metabolic demands of brain tissue are not met resulting in permanent damage and neurological dysfunction if not corrected promptly (Frizzell, 2005). According to the Heart and Stroke foundation of Canada, ischemic events account for approximately 80% of strokes.

1.1 Stroke Epidemiology

According to the World Health Organization, stroke is the second leading cause of death worldwide with an annual burden of approximately 15 million individuals. Many strokes result in death or permanent disability, which requires long-term rehabilitation. In Canada alone, approximately 50,000 individuals suffer a stroke each year, which is equivalent to one every 10 minutes. As many as 80% of individuals who suffer a stroke report restrictions in their daily activities (lifeafterstroke.ca). While improved medical management has caused stroke mortality to decline, the number of survivors living with permanent disability is steadily increasing, necessitating a greater focus on neurorehabilitation research. The disability adjusted life years for stroke, which measures the overall disease burden, was approximately 38 million in 1990, and is projected to rise to roughly 61 million by 2020 (World Health Organization). The economic

impact from stroke is extremely high, with lifetime costs to society in Canada climbing as high as $240 million (data from 2005) (Mittmann et al., 2012). This amounts to an annual economic burden of approximately $110,000 per person (Mittmann et al., 2012). Individuals with a disabling stroke have a greater than two-fold increase in average cost compared to those without disability.

1.2 Background of Study

The prevalence of post stroke disability has been well documented in literature (Gresham et al., 1979, Hong et al., 2010). Recovery from stroke and the success of rehabilitation seems to be highly variable, most likely due to the heterogeneity of the disease. The symptoms associated with stroke are highly dependent on the area of the brain affected. While most survivors will exhibit some degree of spontaneous recovery, many are left with permanent neurological and physical impairments that require ongoing rehabilitation. One of the most common chronic disabilities after stroke is impaired motor function affecting one side of the body, otherwise known as hemiparesis. Hemiparesis of the upper-extremity (UE) is particularly common after stroke, with up to 70% of individuals experiencing mild to severe UE dysfunction (Nakayama et al., 1994). Unfortunately, only 15% of all acute stroke patients who enter rehabilitation for UE weakness will regain full use of their limbs (Sunderland et al., 1992), and as little as 50% of individuals that present with significant arm paresis will recover any useful function (Wade et al., 1983, Sunderland et al., 1989). This creates enormous physical and psychological burdens as the individual with stroke may no longer be able to perform simple daily tasks like eating or cleaning. Upper extremity motor deficits have been associated with longer stay in acute care facilities, increased mortality within acute hospital care (Persson et al., 2012), worse performance in activities of daily living (Harris and Eng, 2007), low levels of physical activity

and overall diminished quality of life (Morris et al., 2013). Since the prevalence of UE dysfunction after stroke is so great, understanding the brain pathologies associated with UE impairment after stroke is of great clinical importance, and a key component of the rehabilitation process. In order to optimize the potential for recovery of stroke survivors and implement successful rehabilitation, it is essential to understand both the underlying mechanisms of injury and processes associated with recovery. The advancement of neuroimaging technology has made this possible.

Neuroimaging has significant potential for tracking structural and functional changes during rehabilitation and may provide critical information in determining recovery from stroke. Advances in neuroimaging techniques over the past few decades have allowed the exploration of the pathophysiology of stroke and subsequent recovery mechanisms in greater detail. After stroke, the brain undergoes extensive remodeling. Morphological and structural changes are triggered spontaneously at the onset of infarction and include mechanisms such as dendritic remodeling, synaptogenesis, and axonal reorganization; each of these changes may persist into the chronic phase post stroke (Dimyan and Cohen, 2011). Additionally, structural changes may be caused by secondary symptoms of stroke, such as cerebral edema, or atrophy which cause both local and distant damage to brain tissue. In individuals with hemiparetic stroke, it has been suggested that the extent of structural damage to cortical areas likely underlies the potential for post-stroke recovery (Schaechter et al., 2006). Therefore, assessing morphological changes within the motor cortex may provide valuable information of the neural events that underlie upper-extremity hemiparesis. The present study will focus on two MRI-based methods of assessing morphological changes to the cortex in individuals with chronic subcortical stroke and how they relate to UE function; 1) computational measurement of cortical thickness, and 2) spectroscopic measurement of cerebral metabolites. Upper-extremity motor performance and

motor activity will be assessed using laboratory based UE performance measures, and non-laboratory based activity measures.

2. REVIEW OF LITERATURE

2.1 Introduction

Although the brain shows a remarkable ability to adapt to ischemic cell death through neuroplastic changes, there are often residual impairments and disability caused by stroke. Neuroimaging developments have been invaluable for investigating the progress of ischemic brain damage. Magnetic resonance imaging (MRI) is useful for highlighting morphological changes in stroke and is routinely implemented as a method of confirming diagnosis, optimizing acute stroke care, investigating post stroke repair, or tracking rehabilitation. Since the first magnetic resonance images were published in 1973, MRI has evolved to become one of the preferred imaging methods to investigate the central nervous system. Recent advances have allowed clinicians and researchers to study, in detail, the pathological processes of stroke. Investigating the neural events that underlie post stroke morphological changes may improve our understanding of the dynamic changes that occur after stroke and may aid in the implementation and success of proper treatment programs following stroke.

2.2 Morphological changes after stroke

The human cerebral cortex is estimated to contain approximately 10 billion neurons, which are structurally organized into a network of local and long-range projection pathways (Hagmann et al., 2008). The cerebral cortex, the highly folded outermost layer of the cerebrum, has an average overall thickness of approximately 2.5mm (Fischl and Dale, 2000), however regional variations in thickness can be quite large. Among the thinnest area of cortex is the postcentral gyrus (primary sensory cortex), which has an average thickness of under 2mm. In contrast, the precentral gyrus (primary motor cortex) is some of the thickest region of cortex and often measures between 2mm-3.5mm thick (Fischl and Dale, 2000, Wang et al., 2010). Cortical

thickness and cerebral volume measures are valuable indicators of the structural integrity of a given brain region, and have been used as a method of tracking morphological changes in both healthy and pathological cases. For instance, longitudinal studies of healthy individuals suggest that loss of brain tissue might be a consequence of normal aging (Resnick et al., 2003, Thambisetty et al., 2010). Rates of cerebral tissue loss during aging have been estimated to be as large as 5.4 cm^3 per year (Resnick et al., 2003). Normal aging has also been associated with widespread declines in cortical thickness. Greater rates of decline have been observed in the frontal and parietal regions, compared with temporal and occipital areas (Resnick et al., 2003, Thambisetty et al., 2010). Cortical morphology may also be a useful tool in identifying pathological cases. Systematic structural changes in the human cerebral cortex have been reported in a variety of pathological states, including epilepsy (Widjaja et al., 2011), multiple sclerosis (Sailer et al., 2003), schizophrenia (Kuperberg et al., 2003) and stroke (Schaechter et al., 2006, Brodtmann et al., 2012, Gauthier et al., 2012). Studying morphological changes in neuropathological conditions has proved useful in identifying areas vulnerable to deterioration as well as evaluating disease progression. In stroke, a decrease in cortical thickness has been documented in animal studies of middle cerebral artery occlusion (Karl et al., 2010); the authors showed that decreases in cortical thickness were present in both ipsilesional and contralesional hemispheres. These results became more pronounced in chronic cases with up to 18.5% loss of ipsilesional cortical tissue, and approximately 7.5% loss contralesional cortical tissue reported.

In humans, structural changes within cortical areas have been documented *in vivo* by measuring grey and white matter volume derived from T1-weighted structural MRI scans. Focal thinning of cortical areas in individuals with subcortical infarct has been reported in patients with cerebral autosomal dominant arteriopathy and leukoencephalopathy, a hereditary small vessel disease. In a study of 9 individuals with single incident infarct, Duering et al. (2012) reported a

strong correlation between mean focal cortical thinning and probability of connectivity to the subcortical lesion. Structural changes in regional cortical thickness have also been observed in individuals in the sub-acute phase (3 months recovery) after subcortical ischemic stroke (Brodtmann et al., 2012), and have been linked to functional activation changes in individuals with chronic stroke (Schaechter et al., 2006). In individuals with sub-acute stroke, Brodtmann et al. (2012) reported decreases in ipsilesional thalamic volume, as well as increases in contralesional paracentral, superior frontal and insular regions, which may reflect compensatory mechanisms involved in motor recovery. Supporting these results, Schaechter et al. (2006) reported increases in cortical thickness in individuals in the chronic stage of stroke recovery, particularly within the ipsilesional postcentral gyrus, which was associated with increased activation response from this brain region during tactile stimulation. However, it remains unexplained how documented changes in precentral gyrus cortical thickness may relate to functional impairment or activity of the stroke-affected UE.

2.3 Proton magnetic resonance spectroscopy in stroke

Proton magnetic resonance spectroscopy H^1MRS is a non-invasive method of obtaining metabolic information from a section of tissue *in vivo*. The technique has been used to assess metabolic changes in a number of pathological conditions such as schizophrenia (Kraguljac et al., 2012), Alzheimer's disease (Roman and Pascual, 2012), brain tumors (Kinoshita and Yokota, 1997), multiple sclerosis (Kirov et al., 2013) and stroke (Fenstermacher and Narayana, 1990, Duijn et al., 1992, Federico et al., 1998, Cirstea et al., 2011). *In vivo* H^1MRS has allowed clinicians and researchers the ability to investigate the pathophysiology of stroke in greater detail by facilitating measurement of certain metabolites that can act as markers for neuronal integrity, cellular metabolism and energy availability within neurons. Metabolic changes have been noted

in individuals with both acute and chronic cerebral infarction (Duijn et al., 1992, Gideon et al., 1992, Houkin et al., 1993, Gideon et al., 1994, Federico et al., 1998, Sztriha et al., 2012). For instance, H^1MRS has been applied to the investigation of acute ischemic stroke to inform of neuronal viability and potential mechanisms of cellular repair (Federico et al., 1998, Sztriha et al., 2012). In acute stroke, the presence of a measurable lactate peak demonstrates a shift from aerobic to anaerobic metabolism in the brain. Previous studies have shown this to be only a temporary effect, and that levels return to almost undetectable levels within 3 weeks (Henriksen et al., 1992). Metabolic changes have also been observed chronically within ipsilesional normal appearing grey matter (Munoz Maniega et al., 2008b) and have been associated with morphological changes in stroke (Demougeot et al., 2004). One metabolite in particular is N-acetylaspartate (NAA), a chemical found within neurons that is believed to reflect neuronal structural and functional integrity (Demougeot et al., 2004, Johansen-Berg, 2012). MRS has shown utility as a prognostic tool when employed in the study of individuals in the acute or sub-acute phase post-stroke. Significantly reduced levels of NAA, choline (Cho; a component of cellular membranes) and creatine (Cre; an energy rich compound that is used as a marker for brain energy reserve) have been reported within the ischemic lesion, of which NAA displays a significant positive correlation with prognostic tests such as the Scandinavian stroke scale (Federico et al., 1998). In individuals with chronic stroke, lower NAA and higher myo-inositol (mI; an astrocyte marker) concentrations have been reported within ipsilesional and contralesional primary motor cortices (Cirstea et al., 2011). A positive correlation has been observed between both NAA and glutamate (Glu; the principle excitatory neurotransmitter in the human central nervous system) and upper extremity function in a chronic stroke population (Cirstea et al., 2011, Craciunas et al., 2013).

2.4 The magnetic resonance spectrum

Any nucleus with an odd atomic number (such as hydrogen, H^1) will spin (aka resonate). When exposed to a static magnetic field, the net magnetization will align in the direction of the field and produce a secondary spin, or wobble, at a certain frequency, known as the Larmor frequency. Because atomic nuclei are surrounded by electrons that slightly shield the nucleus from any external magnetic field, the Lamor frequency is slightly different for different nuclei or compounds. This frequency difference between nuclei or compounds (ie metabolites) is known as chemical shift, and is generally measured in parts-per-million (ppm). The chemical shift of a compound or metabolite is proportional to the external magnetic field applied, but since the shift is reported in ppm, the value is independent of the external magnetic field strength. Each metabolite therefore has a typical signature, or resonance peak that can be found at a known location corresponding to its known chemical shift. The resulting proton magnetic resonance spectrum is displayed as a set of peaks (resonances) along the x-axis, labeled as ppm, and the magnitude of peak intensity is displayed on the y-axis. An example of a spectrum of metabolites can be seen in Figure 3. The spectral data is then processed using external software packages such as LCModel which fits the spectral data to a model spectrum and estimates metabolite intensities. The resonance intensity is proportional to the concentration of the metabolite within the voxel of interest, and is typically reported in molarity units (millimolar; mM).

2.5 Observable peaks

The most common nucleus used for *in vivo* MRS is hydrogen (H^1), because it is abundant in the body, and provides a much higher signal-to-noise ratio compared to other available atoms such as carbon (C^{13}) or phosphorous (P^{31}). One of the advantages in using hydrogen (H^1) in MRS studies is that no additional hardware is required; meaning that the same hardware used for

standard magnetic resonance imaging (MRI) is applicable to H^1 MRS. One particular problem that arises when using H^1 MRS is that of water suppression. The concentration of water in the brain exceeds the concentration of other metabolites by a factor of 10,000 or more. Since the spectrum of observable metabolites in the brain is quite small, the result is an overwhelming resonance peak of water masking any other metabolites of interest. In order to solve this problem, a series of magnetic pulses are implemented to selectively excite just the water resonance, which serves to eliminate the signal from the spectrum (Prost, 2008). Another source of signal contamination may come from lipid signals in the skull. This may be easily corrected by placing the voxel of interest in an area of the brain that avoids overlapping excitation from the scalp or skull. After adjusting for these issues, observable peaks typically reported in H^1MRS studies include N-acetylaspartate, glutamate/glutamine, creatine, choline, and myo-inositol.

N-acetylaspartate (NAA)- NAA is one of most abundant metabolites in the human brain. Levels found in certain regions of the brain can reach as high as 10mM, which makes it one of the most concentrated molecules in the central nervous system (Moffett et al., 2007). Although its precise functional significance remains somewhat controversial, there is some evidence suggesting that it may act as a molecular water pump in myelinated neurons (Baslow, 2002). The interest in studying NAA began largely due to its abundant concentration, making it an ideal and highly reliable molecule to study using spectroscopy. Since it is found almost exclusively in intact neurons, the signal from NAA is a useful marker for neuronal integrity. This is supported by immunocytochemical localization of NAA in neurons (Simmons et al., 1991). In individuals with acute stroke, decreased NAA levels have been reported within the ischemic core, likely reflecting neuronal death due to ischemia (Pereira et al., 1999). In individuals in the sub-acute and chronic phase post-stroke, there is evidence to suggest NAA (reported as a ratio to

choline in the study) may be predictive of neurological outcome (as assessed by the Canadian Neurological Scale, Barthel Index, and Rankin Scale) (Parsons et al., 2000). Total NAA (tNAA) concentration, which includes NAA and the neuropeptide N-acetylaspartylglutamate (NAAG), is commonly reported in spectroscopy studies, and is a known marker of neuronal damage (Pouwels and Frahm, 1997).

Glutamate (Glu)- Glutamate is the principle excitatory neurotransmitter in the human central nervous system. As such, it is generally regarded as one of the most important neurotransmitters, and is important during neuronal development as well as pathological processes in the brain (Meldrum, 2000). Glutamate is synthesized locally from precursors in neurons, and does not cross the blood brain barrier. One of the most prevalent biological precursors to glutamate is glutamine, an amino acid that resides within synaptic terminals. Within presynaptic terminals, the mitochondrial enzyme glutaminase metabolizes glutamine into glutamate (Purves et al., 2001).

$$\text{Glutamine} \rightarrow \text{Glutamate} + NH_3$$

In MRS studies, the peak resonance is typically reported as a mixture concentration of glutamate, as well as the concentration of its readily available precursor glutamine. In stroke, levels of glutamate+glutamine (Glx) in the ipsilesional primary motor cortex have been correlated with arm motor impairment (Cirstea et al., 2011).

Creatine (Cre)- Creatine, and its high-energy phosphorylated form phosphocreatine (PCr) serve as a rapidly available energy reserve in the brain. Its capacity for energy storage is

obtained through reaction with the energy rich compound adenosine triphosphate (ATP). Creatine has historically been used as a reference peak when measuring metabolic changes in the brain, as it was believed to exhibit little to moderate change. This idea has been challenged in recent years, as it appears creatine is susceptible to significant change, and as such should not be used as a reference molecule, particularly when studying pathological cases (Munoz Maniega et al., 2008a). Indeed, a number of studies have reported a significant decrease in Cre concentration after stroke (Fenstermacher and Narayana, 1990, Federico et al., 1998).

Choline (Cho)- Choline is an essential nutrient for humans, and must be obtained in proper quantity from diet. It is an important component of cellular membranes as well as the synthesis of acetocholine in the body (Michel et al., 2006). In MRS of a healthy brain, the choline peak is thought to consist of signals from glycerophosphocholine and phosphocholine, both molecules that are involved in normal membrane synthesis and degradation (Saunders, 2000). In pathological conditions such as stroke that cause accelerated membrane breakdown, decreased signal intensity from Cho would be expected. Indeed a study by Munoz Maniega et al. (2008a) showed that choline levels are significantly decreased in ischemic brain regions two weeks after stroke. As Munoz Maniega et al. (2008a) noted, there has been a reported discrepancy between studies as to the direction of change of choline after stroke. For instance, some have noted an increase in choline levels after stroke (Graham et al., 1992), while most, including Munoz Maniega et al. (2008a), report decreased levels. A possible explanation for this may be individual variances based on small sample sizes, or the time-point of the scan in relation to stroke recovery.

Myo-inositol (mI)- One of the most abundant metabolites in the human brain, mI is found mainly in glia cells and functions as an osmolyte, maintaining cell volume and fluid balance (Haris et al., 2011). Myo-inositol is of interest in clinical investigation because its concentration is altered in many neurological disorders involving osmotic stress to the brain. For example, elevated levels of mI have been reported in Alzheimer's disease (Miller et al., 1993), multiple sclerosis (Kapeller et al., 2005) and stroke (Cirstea et al., 2011). In contrast, decreased concentrations of mI have been observed in certain brain tumors (Kinoshita and Yokota, 1997). A summary of each metabolite and their role in stroke can be found in Table 2.

2.6 Assessing UE motor impairment after stroke

Rehabilitation programs after stroke are focused on restoring functional use and enhancing skills needed to perform daily activities. Several measures have been developed that attempt to capture upper extremity impairment or disability. One of the most commonly used in stroke is the Wolf Motor Function Test (WMFT) (Wolf et al., 1989). The WMFT is a quantitative index of upper extremity motor ability. The original test developed by Wolf et al. (1989) was designed to examine the effects of the constraint-induced movement therapy (CIMT) trial in stroke and traumatic brain injury. The current version of the test is presented as a series of 15 timed functional tasks, which are sequenced according to joints involved and level of difficulty. The tasks are designed to reflect functional tasks that may be encountered on a daily basis, such as lifting a can to drink, turning a key in a lock, or folding a towel. Each of the tasks is timed to completion, up to a maximum of 120 seconds. The test has been validated for use in stroke, and exhibits high inter-rater reliability, construct validity, and criterion validity (Wolf et al., 2001, Wu et al., 2011).

While laboratory based functional assessments such as the Wolf motor function test are generally able to describe residual functional abilities in the paretic limb of stroke affected individuals, there seems to be a disparity between functional impairment (as measured by the WMFT) and upper-extremity use outside the laboratory (Uswatte et al., 2005a). This would suggest that WMFT scores should not be used as proxies for everyday upper-extremity use outside of the laboratory (Dromerick et al., 2006). In order to comprehensively evaluate the affected upper-extremity, two key factors should be taken in to account: 1) identifying impairments that limit movement of the UE and 2) how impairments limit activity and restrict activities of daily living (Lang et al., 2013). Currently, only 2 assessment tools have been validated to measure use of the affected UE outside of the laboratory in everyday life, the Motor Activity Log (MAL) and accelerometry.

The Motor Activity Log is a structured interview designed to assess real world upper extremity function. It was initially developed by Taub et al. (1993) to assess the more-impaired arm outside of a laboratory setting in individuals who were undergoing constrain-induced movement therapy. The MAL consists of 14 activities of daily living (ADL), such as picking up a glass or brushing teeth, and asked individuals how they used their more-impaired arm for each task within a given time period (i.e. within the last week). Individuals are asked to rate how often (Amount of Use [AOU] scale), and how well (Quality of Movement [QOM] scale) they perceive they are able to perform each ADL compared to before their stroke. Responses are recorded on a 6-point scale, with options between 0 (never used their more impaired arm for the task within the given time period) and 5 (they were able to use their arm the same as before the stroke). Studies assessing the MAL's clinimetric properties have found it to be internally consistent and relatively stable in chronic stroke patients (van der Lee et al., 2004); however, only the validity

of QOM scale was supported (Uswatte et al., 2005b). A high correlation has been found between the QOM scale and accelerometry readings of UE activity (Uswatte et al., 2005b).

Accelerometers are devises that measure the intensity of acceleration and are used as a measure of physical activity. Accelerometry recordings have shown to be reliable and valid as a tool to objectively measure the amount of daily physical activity of individuals with stroke (Uswatte et al., 2000, Rand et al., 2009, Rand and Eng, 2010) and are useful as a measure of real-world upper extremity movement (Uswatte et al., 2000). Associations between accelerometry measures and upper extremity impairment have been observed in individuals after stroke (Gebruers et al., 2010).

3. RATIONALE

Magnetic resonance imaging is one of the most important imaging modalities for the diagnosis of acute and chronic ischemic lesions; however, its use as a prognostic or predictive tool for recovery is relatively new. Advances in magnetic resonance imaging (MRI) have vastly improved our understanding of the spontaneous events that accompany post stroke plasticity and functional recovery. This has permitted a more comprehensive analysis of the sequelae of changes that occur both local to and distant from primary infarction. To our knowledge, no study has investigated how changes in regional cortical thickness may relate to metabolic changes in individuals with subcortical stroke in the chronic stage of recovery, or how changes in precentral gyrus cortical thickness or the concentration of tNAA or Glx may be used to assess UE functional impairment and activity outside of the laboratory setting. This study will assess how metabolic and volumetric changes in the motor cortex relate to upper-extremity motor impairment in individuals with chronic subcortical ischemic stroke.

3.1 Specific Aims

1) To investigate anatomical and metabolic changes within normal appearing grey matter of the primary motor cortex in individuals with subcortical ischemic stroke in the chronic stage of recovery, and neurologically healthy controls. Our hypothesis is that the stroke group will exhibit decreased precentral gyrus cortical thickness and metabolite concentration. Further, in the stroke group we expect a significant decrease in ipsilesional precentral gyrus thickness, and tNAA and Glx concentration compared to contralesional hemisphere.

2) To examine the relationship between precentral gyrus cortical thickness and metabolite measures. Our hypothesis is that cortical thickness measures will be associated with both tNAA and Glx concentration.
3) To investigate the relationship between imaging data and UE performance and activity in individuals in the chronic stage post-stroke. We hypothesize that measures of ipsilesional cortical thickness as well as tNAA and Glx concentration will be linked to upper-extremity motor performance and activity.

4. INTRODUCTION TO THE CURRENT STUDY

Hemiparesis is one of the most common and persistent side effects of stroke. The condition is characterized by weakness on one side of the body and may affect an individual's ability to perform everyday activities such as eating, drinking, or cleaning. Upper extremity weakness in particular is common after stroke and presents a serious obstacle to normal daily activities (Sveen et al., 1999) and quality of life (Edwards et al., 2010, Morris et al., 2013). Neuroimaging developments have provided valuable insight in to the underlying mechanisms contributing to upper extremity dysfunction after stroke. Dynamic structural reorganization occurs as neuronal connections and cortical maps spontaneously remodel in response to brain lesions (Calautti and Baron, 2003, Brodtmann et al., 2012). Survival of peri-infarct tissue is critical during this period and is an important component of recovery (Cramer et al., 2006). Evidence suggests that areas distant from the primary infarction may also be critical to recovery. For instance, in individuals with subcortical stroke, recovery of arm function has been linked with greater functional activity within cortical areas remote from the lesion (Weiller et al., 1993, Cramer et al., 1997, Cramer, 2008). Further, functional imaging of individuals with chronic stroke has shown that activation of the ipsilesional hemisphere is associated with improved motor outcome (Ward et al., 2003, Calautti et al., 2010) and structural integrity of overlying cortical areas (Schaechter et al., 2006). Taken together, it is evident that morphological changes and structural integrity of surviving cortical areas remote from the lesion may contribute to upper-extremity motor deficits in chronic stroke.

Morphological changes in the cortex after stroke have previously been investigated using biological markers of membrane integrity and neuronal damage, such as the neuronal metabolite N-acetylaspartate (NAA). Magnetic resonance spectroscopic (MRS) studies of acute stroke have linked decreases in NAA concentration to poor functional recovery of the hemiparetic upper-

extremity (Federico et al., 1998). Similar results have been reported in individuals with chronic stroke, as decreases in NAA within cortical motor areas remote from the lesion have been associated with increased arm impairment (Cirstea et al., 2011). While MRS has provided important information about neuronal integrity and metabolism, it remains unclear how regional structural changes such as in cortical thickness may be associated with metabolic assessment of neuronal integrity and motor function after stroke. Voxel-based morphometric investigation in individuals with chronic subcortical stroke has revealed grey matter atrophy in motor areas, which may be associated with arm impairment (Gauthier et al., 2012). Increases in cortical thickness have also been reported in the contralesional hemisphere of individuals with sub-acute (Brodtmann et al., 2012) and chronic stroke (Schaechter et al., 2006) within sensorimotor cortical areas, but it is not known how these changes may relate to functional activity of the hemiparetic upper-extremity. In the current study, we performed metabolic and structural assessment of the primary motor cortex using H^1 magnetic resonance spectroscopy and cortical thickness measurement to investigate whether morphological changes are associated with upper extremity motor performance and activity.

4.1 Materials and Methods

4.2 Participants

Seventeen hemiparetic individuals with subcortical ischemic stroke in the chronic phase (>6 months) of recovery, and 11 neurologically healthy controls were recruited. The research ethics board at the University of British Columbia approved all aspects of this study, and consent was obtained according to the Declaration of Helsinki. Individuals were excluded if they: 1) were in the acute (0-3 months) or sub-acute (3-6 months) phase of recovery, 2) had contraindications to MRI, 3) had previous history of stroke, epilepsy, neurodegenerative disorder

or head trauma, or 4) showed a visible lesion extending in to the precentral gyrus grey matter. Upon enrollment, each participant underwent neuroimaging assessment in a Phillips Achieva 3.0T MRI scanner at the University of British Columbia, which was followed by functional assessment in the Brain Behaviour Lab. A subset of 10 participants from the stroke group were administered the MAL questionnaire and fitted with wrist mounted accelerometers to assess motor activity outside of the laboratory.

4.3 Imaging

As part of a larger imaging study, several MRI scans were completed. For the purposes of the current thesis, a high-resolution T_1-weighted scan (TR/TE: 7425/3.64ms, FOV=256x256mm, 160 sagittal slices, flip angle 6°, voxel size=1mm^3) was performed on each enrolled participant. In addition, single voxel ^{1}H-MRS (TR/TE=2000/35ms, sampling frequency: 2000Hz, data points=1024, signal averages=128) was performed to measure absolute metabolite concentrations. A single 30mm x 22mm x 15mm voxel was centered over the hand area of the primary motor cortex in both ipsilesional and contralesional hemispheres. Localization of hand representation for each individual and corresponding voxel placement was determined using known anatomical coordinates (Yousry et al., 1997) (Figure 4).

4.4 Upper-extremity motor assessment in the laboratory

UE motor performance was assessed using the WMFT, which measures how rapidly individuals are able to perform a series of 15 timed functional tasks in the laboratory. A trained physiotherapist carried out all components of the assessment. Since it was developed for use in individuals involved in CIMT, most of whom exhibited mild to moderate stroke, the WMFT is most sensitive for use in this population of stroke survivors (Morris et al., 2001). Several

problems exist with the WMFT when applied to severely impaired subjects. Namely, there is a significant floor effect, which occurs when a task is not completed within the allotted time, thus preventing an accurate representation of task performance. Recently, the test has been validated for use in severely affected subjects, by adjusting the method by which the scores are calculated (Hodics et al., 2012). Instead of reporting median scores for each task, Hodics et al. (2012) used the task time scores and reciprocally transformed them in to task rate data

$$\text{Task Rate} = 60\ (\text{s}) / \text{Performance Time (s)}$$

This method of calculation has shown to be more sensitive in assessing the functional abilities of the severely affected upper extremity of stroke patients. Thus, participants in the current study were assessed using the standard 15-task WMFT and scores were calculated into task-rate data as described by Hodics et al. (2012).

4.5 Upper-extremity motor assessment outside of the laboratory

In the current study multidirection piezoelectric accelerometers (Actical™, Mini Mitter Co) were used to quantify daily upper extremity use in both arms over 11 consecutive days as previously described by Rand and Eng (2010). The Actical accelerometer is a small (28X27X10 mm), lightweight (17g) device which has a frequency range of 0.3 to 3 Hz, is sensitive to 0.05 to 2.0 G force, and samples at 32 Hz. Acceleration data is rectified and integrated over 15-second epochs and stored as activity counts. The device detects acceleration in all 3 planes although they are more sensitive to detection in the vertical plane. The Actical™ system was selected as it was found to have higher intra-instrument and inter-instrument reliability as compared to 2 other commonly used accelerometers; Actigraph and RT3 (Esliger and Tremblay, 2006). Further, daily activity counts from wrist mounted Actical accelerometers have shown to significantly correlate with hand dexterity and grip strength (Rand and Eng, 2010). Participants wore wrist-mounted

accelerometers on each wrist consecutively over an 11-day period. Each individual was instructed to wear the accelerometers outside of the laboratory during waking hours and advised to go about their daily activities as normal. Upon completion of the 11-day period, participants were asked about their experience and if there were any extraordinary circumstances during the time period that may show up in the data. The MAL was also used to assess how stroke survivors use their more impaired arm outside the laboratory. Interviews were conducted under standardized procedures according to the protocol explained by Taub E (1996).

4.6 Neuroimaging

Cortical reconstruction and volumetric segmentation was performed with the Freesurfer image analysis suite, which is documented and freely available for download online (http://surfer.nmr.mgh.havard.edu). The technical details of these procedures are described in prior publications (Dale et al., 1999, Fischl and Dale, 2000). A number of steps for processing are required, and include: 1) motion correction and averaging (Reuter et al., 2010) of multiple volumetric T1 weighted images (when more than one is available), 2) removal of non-brain tissue using a hybrid watershed/surface deformation procedure (Segonne et al., 2004), 3) automated Talairach transformation, 4) segmentation of the subcortical white matter and deep gray matter volumetric structures (including hippocampus, amygdala, caudate, putamen, ventricles) (Fischl et al., 2002, Fischl et al., 2004) 5) intensity normalization (Sled et al., 1998), 6) tessellation of the gray matter white matter boundary, and 7) automated topology correction (Fischl et al., 2001, Segonne et al., 2007), and surface deformation following intensity gradients. This final step is done to optimally place the gray/white and gray/cerebrospinal fluid borders at the location where the greatest shift in intensity defines the transition to the other tissue class (Dale and Sereno, 1993, Dale et al., 1999, Fischl and Dale, 2000). MRI-derived measures of

human cerebral cortical thickness have shown to be highly reliable (Han et al., 2006), and have been described in a number of pathological conditions such as Huntington's disease (Rosas et al., 2002), schizophrenia (Kuperberg et al., 2003) and multiple sclerosis (Sailer et al., 2003). Recently the technique has been used to investigate regional changes in cortical thickness in a sub-acute stroke population (Brodtmann et al., 2012). We compared average cortical thickness in the precentral gyrus in both stroke (ipsilesional/contralesional) and healthy (non-dominant/dominant) groups. Identical group divisions were used for metabolic analysis. Metabolic measures were quantified using LCModel package (with water scaling), which provides automatic quantification of *in vivo* proton magnetic resonance spectra (Provencher, 1993). An example of a spectrum from one participant is presented in Figure 5.

4.7 Statistical tests

To examine differences in tNAA and Glx concentration and precentral gyrus thickness between the stroke and healthy groups, multivariate analysis of variance (MANOVA) was conducted. Univariate analysis of variance (ANOVAs) for each dependent variable was conducted as follow-up tests to the MANOVA. Parametric correlation analysis using Pearson's r was conducted to examine the relationship between tNAA and Glx and precentral gyrus thickness in both hemispheres. Multiple linear regression analyses were performed to assess the amount of variance in UE motor performance (WMFT score and MAL-QOM score) and motor activity (accelerometer activity counts) explained by demographic information (age and post-stroke duration), metabolite concentration (tNAA and Glx), and cortical thickness. In the stroke group, age and post stroke duration were entered first to account for post-stroke (Graham et al., 1993) and age related changes in metabolites (Angelie et al., 2001, Kaiser et al., 2005) and cerebral tissue (Resnick et al., 2003). Concentration of ipsilesional cerebral metabolites tNAA

and Glx were the next predictor entered, based on previous reports suggesting their association with UE motor impairment (Cirstea et al., 2011, Cirstea et al., 2012). Ipsilesional cortical thickness was the final predictor entered in to the model.

5. RESULTS

Participants

Seventeen individuals (12 male, 5 female; age 68 +/- 9.6 years) with first-time subcortical ischemic stroke who were in the chronic stage (>6months) of recovery, and 11 neurologically healthy controls (4 male, 7 female; age 60.4 +/- 6.0 years) enrolled. Participant group summaries are listed below in Table 1, and individual information from participants in the stroke group is presented in Table 3. The location of each lesion was determined from T1-weighted MRI (Figure 6).

Table 1. Participant characteristics

	Stroke Group	**Healthy Control Group**
N (gender)	12 Male, 5 Female	4 Male, 7 Female
Age (years)	68 (+/- 9.6)	60.4 (+/- 6.0)
Post-stroke duration (months)	51 (+/- 36.6)	-

Aim 1: To investigate anatomical and metabolic changes within normal appearing grey matter of the primary motor cortex in individuals with chronic subcortical ischemic stroke and neurologically healthy controls.

Concentrations of tNAA ($F = 6.47$; $p = 0.017$), Glx ($F = 10.37$; $p = 0.001$), and cortical thickness ($F = 6.12$; $p = 0.021$) were all significantly lower bilaterally in the stroke group compared to the healthy control group. Within the stroke group, a significant decrease in ipsilesional tNAA ($F = 4.14$; $p = 0.05$), Glx ($F = 11.72$; $p = 0.002$) and cortical thickness ($F = 6.37$; $p = 0.017$) was observed compared to the contralesional hemisphere (Figure 1). No

significant difference between hemispheres was found in the healthy control group for any of the variables.

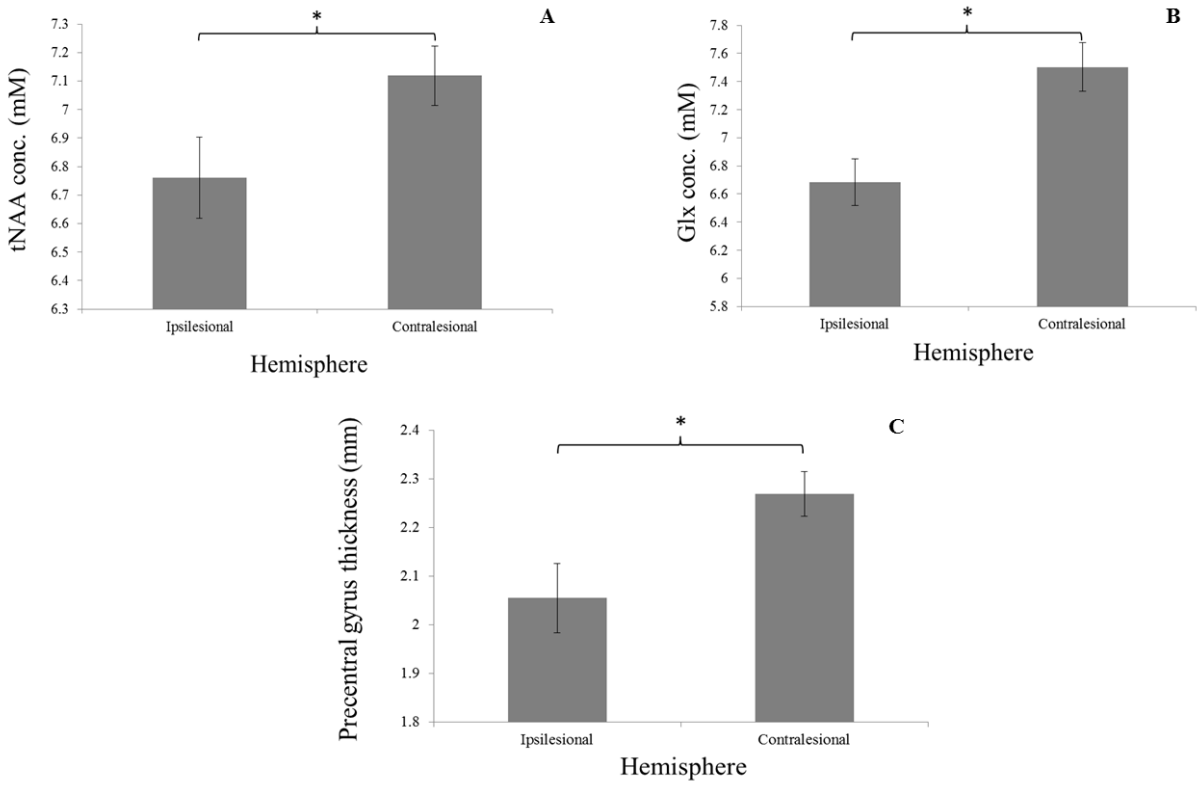

Figure 1. Mean values for metabolite concentrations and cortical thickness measurement from the stroke group. A) tNAA and B) Glx both exhibited significantly lower concentration in the ipsilesional hemisphere. C) Ipsilesional precentral gyrus thickness was also significantly lower in the ipsilesional hemisphere. Significance ($p<0.05$) is indicated with a star (*).

Aim 2: To examine the relationship between precentral gyrus cortical thickness and metabolite measures in chronic subcortical ischemic stroke.

Correlation analyses are summarized in Table 4. A significant positive correlation was found between tNAA and precentral gyrus thickness in both ipsilesional ($r = 0.782$; $p < 0.001$) and contralesional ($r = 0.517$; $p = 0.033$) hemispheres of the stroke group (Figure 2). No correlations between Glx concentration and precentral gyrus thickness was observed.

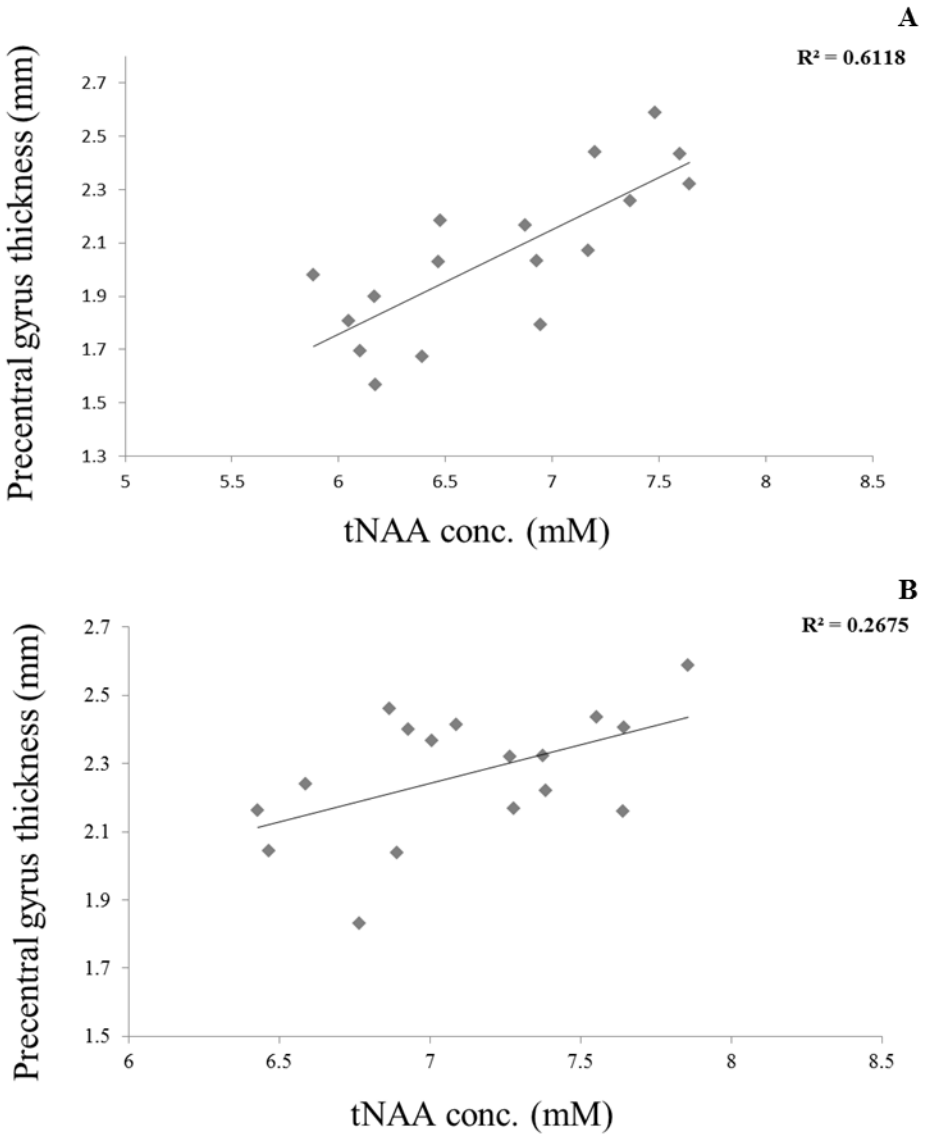

Figure 2. Scatterplot depicting the bivariate relationship between precentral gyrus thickness and tNAA concentration. Parametric correlation analysis using Pearson's r showed a significant positive correlation between precentral gyrus thickness and tNAA concentration in A) ipsilesional and B) contralesional hemispheres.

Aim 3: To investigate the relationship between imaging data and UE performance and activity in individuals in the chronic stage post-stroke.

Wolf Motor Function Test

All regression analyses are summarized in Tables 5-7 and Figures 7-9. In the stroke group, after adjusting for age and post-stroke duration, ipsilesional metabolite concentrations (tNAA and Glx) accounted for 40% of the total variance of hemiparetic UE WMFT score (R^2 = 0.535, p = 0.043). Note that tNAA concentration explained a larger amount of variance in WMFT motor performance. Adding ipsilesional precentral gyrus cortical thickness to the model did not explain an additional amount of unique variance in WMFT score, and the overall model failed to reach significance (R^2 = 0.540, p = 0.088).

Motor Activity Log and Accelerometry

Ten participants from the original group of 17 were administered the MAL and fitted with wrist mounted accelerometers to assess activity of the hemiparetic upper-extremity outside of the laboratory. Linear regression analysis following identical steps to the previous section was performed (summarized in Tables 5-7; Figures 7-9). After adjusting for age and PSD, ipsilesional tNAA and Glx concentration was able to predict 60% of the total variance in MAL-QOM score, although the model failed to reach significance (R^2 = 0.765, p = 0.078). The addition of ipsilesional cortical thickness improved the model, accounting for 10% of the variance in MAL-QOM scores (R^2 = 0.864, p = 0.07). For the accelerometry data, neither the addition of ipsilesional metabolite concentration (R^2 = 0.652, p = 0.188), or ipsilesional cortical thickness (R^2 = 0.780, p = 0.167) to the model was able to significantly predict the amount of variance in accelerometry activity counts from participants' hemiparetic UE.

6. DISCUSSION

The aim of the current study was to investigate the relationship between changes in cortical thickness and metabolites in the motor cortex and how they relate to upper-extremity performance and use in individuals in the chronic phase of stroke recovery. We observed decreases in precentral gyrus cortical thickness, as well as tNAA and Glx concentration in individuals with stroke, particularly in the ipsilesional hemisphere. The decrease in tNAA concentration was associated with a decrease in cortical thickness bilaterally in the stroke group. Our results suggest that ipsilesional precentral gyrus thickness and tNAA concentration were associated with motor performance in laboratory (WMFT) and in everyday life (MAL-QOM) but not activity outside of the laboratory.

Metabolite Concentrations

In the current study, we assessed the concentrations of tNAA and Glx in the hand area of the motor cortex in individuals in the chronic stage of stroke recovery. tNAA and Glx were selected *a priori* for a couple of reasons; 1) changes in both these metabolites have been reported in individuals with chronic stroke, and have been associated with upper-extremity impairment (Cirstea et al., 2011, Cirstea et al., 2012), but it is unclear how each may be associated with upper-extremity activity outside of the laboratory 2) an observed decrease in cortical thickness would most likely be explained by neuronal death, which can be assessed using tNAA as a marker of neuronal integrity.

To our knowledge, this is the first study to report decreased Glx concentration in motor cortical areas of individuals in the chronic stage post-stroke. We also observed a decrease in the cerebral metabolite tNAA which is consistent with previous findings in stroke (Cirstea et al., 2011, Craciunas et al., 2013) and other pathological cases such as multiple sclerosis (Kapeller et

al., 2001, Chard et al., 2002) and schizophrenia (Lim et al., 1998). Because these metabolic changes were observed in brain regions remote from the infarct, it is possible that diaschisis could explain these results. Diaschisis is a common dysfunction after stroke caused by the impairment of function in intact brain regions connect to, but distant from the area of infarct (Seitz et al., 1999). Diaschisis is typically accompanied by a decrease in cerebral blood flow, which may have distant effects on neurometabolic activity resulting in neuronal loss. This has been observed in animal models of focal stroke, where areas adjacent to the infarct, particularly within ipsilesional cortical areas, exhibit sustained hypometabolism (decreased blood flow) (Carmichael et al., 2004) or decreased physiological activation in response to peripheral stimulation (Ginsberg et al., 1989). Decreased NAA concentration has been observed in brain regions that show reduced cerebral blood flow but incomplete ischemia (Handa et al., 1997). Further, reduced regional cerebral blood flow in cortical areas has been associated with neuronal metabolite variations (specifically Cr/NAA) in stroke (Chu et al., 2002), so it seems reasonable to believe that chronic hypometabolism in the ipsilesional cortex may contribute to the decreased tNAA concentrations we observed.

Another possible mechanism that may explain our finding of Glx and tNAA concentration is neuronal death. Because NAA and NAAG are believed to exist almost exclusively in intact neurons, a measurable decrease suggests neuronal death (Pereira et al., 1999). Neuronal death following ischemic stroke is often attributed to improper homeostatic balance of glutamate. The release of glutamate following ischemia results in a cascade of cellular events such as intracellular calcium increase and mitochondrial dysfunction that ultimately leads to cellular death (Hazell, 2007). Acutely after stroke, the increase of glutamate concentration increases for approximately 6 hours after the onset of infarction (Davalos et al., 1997), however the permanent damage caused during this period of time may offer a mechanism for distant

neuronal death. Because ipsilesional tNAA and Glx decrease was accompanied by decreased thickness of the percentral gyrus, it is likely that neuronal death can, at least in part, explain our results. Supporting this idea, age-related brain shrinkage has been associated with decreased NAA and Glx concentration in the motor cortex (Kaiser et al., 2005). Further, in the current study we observed a significant positive correlation between tNAA concentration and precentral gyrus thickness.

Cortical thickness

Precentral gyrus thickness was significantly decreased in the stroke group compared to the healthy controls. Ipsilesional thickness in particular was decreased in the stroke group and was significantly associated with UE motor performance. These results are consistent with previous reports suggesting significant bilateral diffuse tissue loss is common in individuals with chronic stroke (Kraemer et al., 2004, Gauthier et al., 2012), and decreases in grey matter density in non-infarcted motor regions in individuals with chronic stroke is correlated with upper-extremity motor deficit (Gauthier et al., 2012). Interestingly, these results differed from previous studies that reported increases in contralesional sensorimotor cortical thickness in individuals with chronic stroke (Schaechter et al., 2006), and increases in contralesional paracentral and superior frontal regions in individuals with sub-acute stroke (Brodtmann et al., 2012). It's possible that these differences could be explained by compensatory mechanisms within the contralesional hemisphere, such as structural plasticity within cortical regions in response to brain injury, or possibly the activation of neural growth promoting elements. Functional reorganization of contralesional motor cortical areas has described previously in animal models of stroke undergoing rehabilitation training (Nudo et al., 1996), and has been linked with improved hand function. There is evidence to suggest that contralesional compensatory changes

are dependent upon increased use of the less-impaired limb (Nudo et al., 2001). Further investigation should be done to determine the effect of rehabilitation on structural changes in the cortex of individuals in the chronic stage post-stroke.

Upper extremity motor assessment

Upper-extremity motor performance was assessed using the WMFT and MAL-QOM scale, while UE motor activity was measured using wrist-mounted accelerometers. In the multivariable regression analyses, age and time post stroke alone were not significantly associated with UE motor performance or UE motor activity. When these variables were combined with tNAA and Glx concentration the model was able to explain a significant amount of variance in hemiparetic UE motor performance. tNAA was able to explain the greatest amount of variance in WMFT score and MAL-QOM score. The addition of precentral gyrus thickness to the model only resulted in a small increase in R^2. These results suggest that tNAA concentration, a marker for neuronal integrity, in spared ipsilesional motor cortical areas may contribute to UE motor performance in individuals with UE hemiparesis in the chronic stage post-stroke. The fact that ipsilesional precentral gyrus thickness resulted in only a modest improvement to the model suggests that the two measures (tNAA concentration and cortical thickness), may be redundant. This is supported by the strong correlation we observed between tNAA concentration and precentral gyrus thickness. These data support previous work that found decreased grey matter structural density in individuals with chronic stroke, suggesting a role of grey matter atrophy in arm motor impairment (Gauthier et al., 2012).

Contrary to our hypothesis, while Glx concentration was significantly decreased ipsilesionally in the stroke group, we observed no association between this decrease and upper-extremity motor performance. Our original hypothesis that an ipsilesional Glx decrease would be

associated with UE motor performance was based on reported intracortical excitability changes after stroke (Tarkka et al., 2008, Carmichael, 2012) and because Glx concentration in motor areas has been correlated with arm motor impairment in individuals with chronic stroke (Cirstea et al., 2011). Further, Glx concentration in the motor cortex has been correlated with global motor cortical excitability (assessed by transcranial magnetic stimulation) (Stagg et al., 2011), which has shown to be a predictor of motor recovery after stroke (Jung et al., 2012). Further investigation of the relationship between glutamate and glutamine concentration and motor function in individuals after stroke should be done.

Although upper-extremity performance was associated with ipsilesional tNAA concentration and precentral gyrus thickness, UE motor activity in everyday life showed no association with any of the measures (ipsilesional tNAA and Glx concentration or precentral gyrus thickness). It has been reported that there is a disparity between functional motor recovery after stroke and use of the affected upper extremity during daily activities (Rand and Eng, 2012). There are a number of possible reasons to explain this disparity. For instance, as Rand and Eng (2012) suggested, individuals may avoid using their more affected arm for reasons such as fatigue or learned non-use of the UE. In addition, it has been reported that individuals undergoing rehabilitation will perform most of their physical activity under supervision of a therapist, and remain largely inactive for the remainder of the day (Esmonde et al., 1997). Alternatively, it is possible that individuals in the current study were using their hemiparetic UE almost exclusively during daily activities as an attempt to improve function of their weaker arm. This particular circumstance was encountered in at least one of the participants in our current study. This may explain why we did not find a correlation between accelerometry measures of activity and either metabolite concentration or cortical thickness measures. Finally, because UE

use outside of the lab was assessed in only ten participants, it is likely that the sample size of the group was too small, and we did not have sufficient power to detect an association.

Limitations

There were a few of limitations to this study that warrant mention. The mean age of the stroke group was significantly older than the mean age of the neurologically healthy control group. Because normal aging is associated with cerebral tissue loss (Resnick et al., 2003) and changes in NAA and glutamate concentration (Angelie et al., 2001, Kaiser et al., 2005), it is possible that the group differences observed in these measures can be explained by age differences. As such, we controlled for this variable in our analysis. It's worth mentioning that the age difference between the groups in our current study was relatively small (stroke group age 68 +/- 9.7 years; healthy control group age 60.4 +/- 6.0 years) and therefore differences in metabolite concentration and cerebral tissue volume would not likely be explained by differences in age between groups.

For H^1MRS data collection, we used a technique known as single voxel spectroscopy (SVS), which obtains a signal from a single volume of interest. An alternative technique to SVS, known as chemical shift imaging (CSI) obtains a chemical signal simultaneously from multiple voxels in brain regions of interest. A limitation with SVS is that our voxel size (30x22x15mm) is extends beyond the hand area of the primary motor cortex. However, we selected SVS because it is the preferred method for obtaining consistent, high quality *in vivo* spectra (Drost et al., 2002). Finally, the sample size of our groups was relatively small, which may have reduced statistical power necessary to detect associations between imaging measures and UE motor activity.

7. CONCLUSIONS

Taken together, these findings suggest that persistent motor impairment in individuals with chronic stroke may be, at least in part, due to decreased structural integrity of motor areas remote from infarction. To our knowledge, this is the first study to investigate the relationship between local changes in cortical thickness and metabolites in chronic stroke. Reduced cortical thickness and metabolite concentration support previous work indicating persistent local changes in the stroke-affected brain.

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APPENDICES

Appendix A: The magnetic resonance spectrum

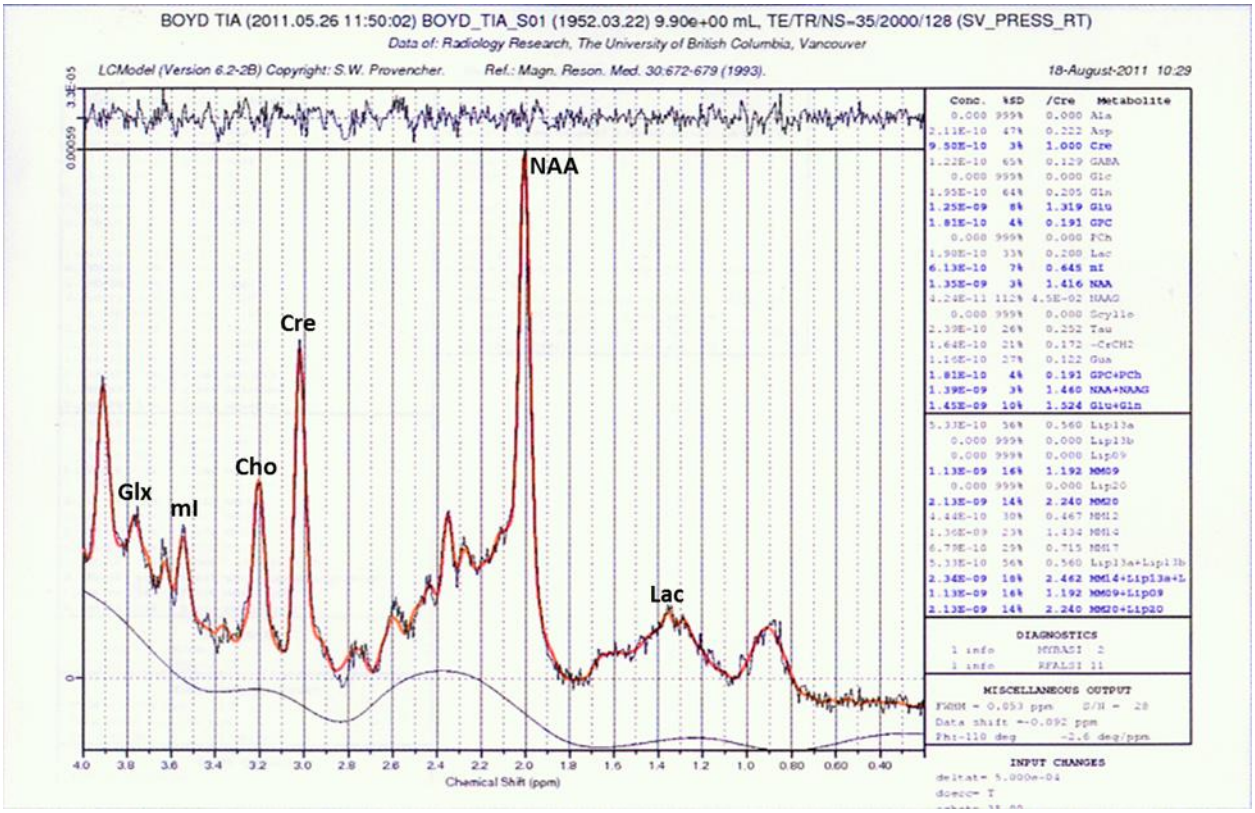

Figure 3. An example of a magnetic resonance spectrum from the human brain. Chemical shift is listed along the x-axis (in ppm). Some common peaks are labeled, including NAA, Glx, Cre, Cho, and mI.

Appendix B: Metabolite summaries

Table 2. List of metabolites

Metabolites	Concentration	Function (s)	Role in stroke
N-acetylaspartate (tNAA)	~ 8-10 mM	- Marker for intact neurons - Possible reservoir of acetyl groups in the brain - Possible role as a molecular water pump	- Levels decreased, possibly due to neuronal death. - Associated with arm motor impairment
Glutamate+glutamine (Glx)	~7-8 mM	- Principle excitatory neurotransmitter	- Associated with global motor cortical excitability and arm motor impairment
Creatine (Cre)	~6-6.5 mM	- Related to cell energy pathways - Reflects energy potential available in brain tissue	- Decreased in stroke
Choline (Cho)	~1-1.5 mM	- Precursor in the synthesis and breakdown of cell membrane phospholipids	- Increased signal in stroke associated with accelerated membrane turnover -Decreased signal has also been reported in stroke
Myo-inositol (mI)	~5-6 mM	- Astrocytic marker and possible marker for intracellular osmotic integrity	- Concentration decreased in stroke

Appendix C: Placement of H^1MRS voxel

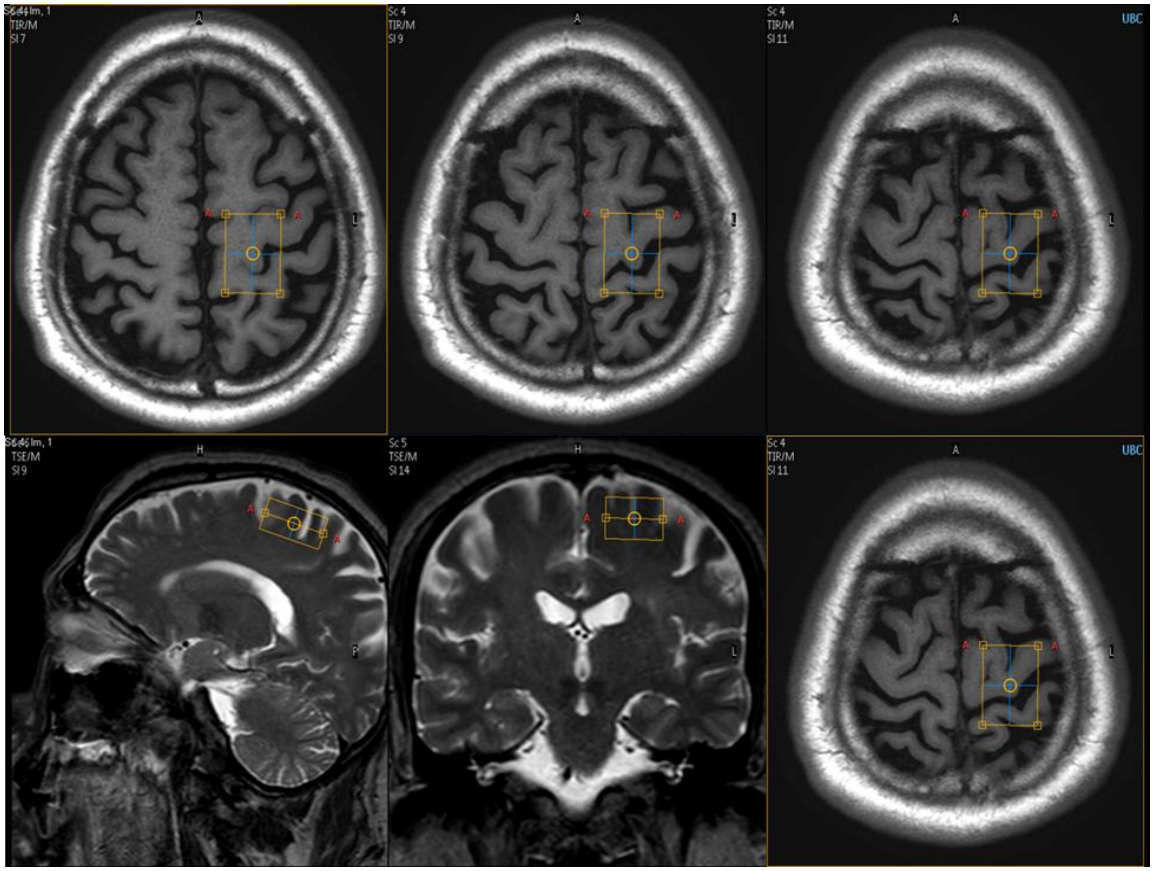

Figure 4. Orientation of the voxel for MRS. A single 30x22x15mm voxel was centered over the hand area of the primary motor cortex in both ipsilesional and contralesional hemispheres.

Appendix D: Sample spectrum of metabolites from H^1MRS imaging

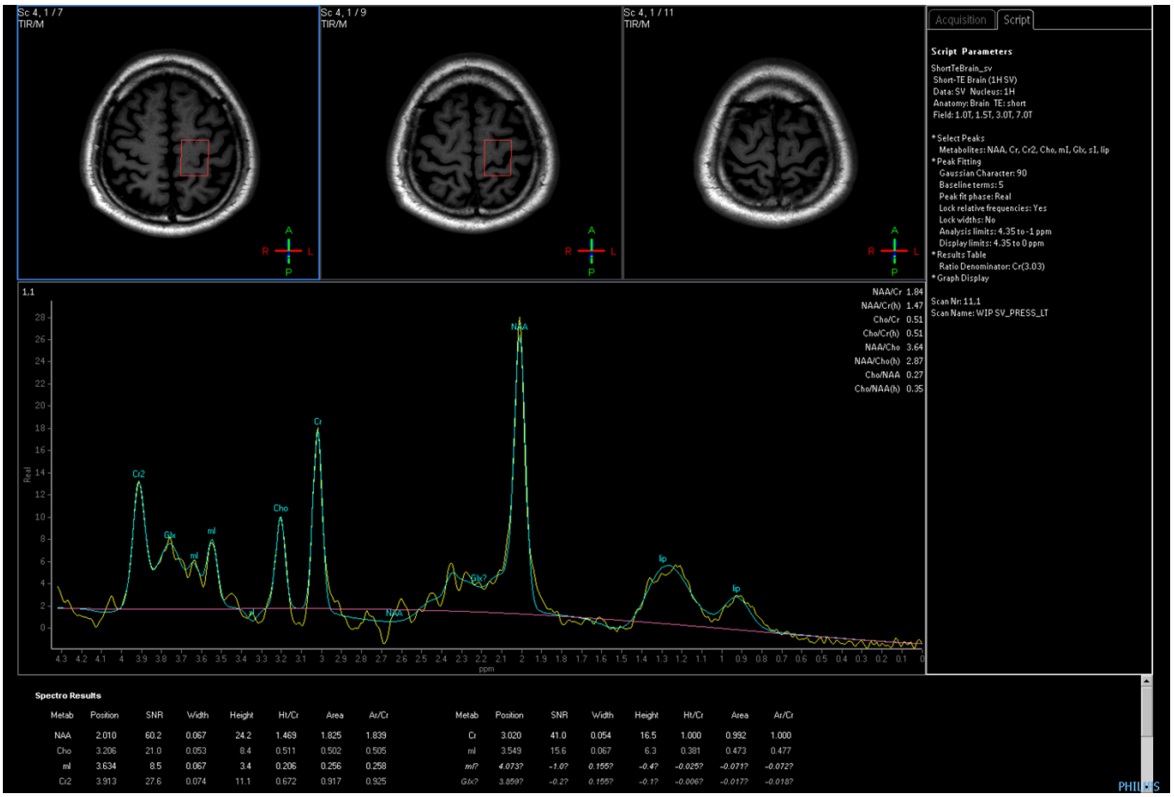

Figure 5. A screen capture of LCModel output shows the placement of the voxel centered over the hand area of the motor cortex, and the corresponding spectrum of metabolite concentrations obtained from the voxel.

Appendix E: Summary of individual participants

Table 3. Participant demographics

Participant number	Age	Gender	Time post stroke	FM	Stroke hem	Ipsilesional NAA conc. (mM)	Contralesional NAA conc. (mM)	Ipsilesional Glx conc. (mM)	Contralesional Glx conc. (mM)
S01	73	M	142	62	L	7.645	7.642	7.247	7.24
S02	71	F	83	60	L	6.927	6.464	7.424	8.099
S03	85	M	35	56	R	6.875	6.429	6.318	6.198
S05	63	M	41	23	R	6.946	7.385	6.783	8.436
S07	50	F	37	63	L	7.597	7.856	5.365	7.422
S08	56	F	27	35	R	7.365	7.266	6.682	6.523
S12	76	M	81	46	L	7.168	6.89	6.489	6.978
S14	71	M	20	58	R	7.482	7.554	6.399	7.756
S15	65	M	67	62	R	6.393	6.764	6.101	7.445
S21	82	M	12	59	R	6.101	6.588	6.989	7.093
S22	61	M	91	16	L	6.169	7.088	6.794	8.096
S23	69	M	15	57	R	6.478	7.005	7.092	6.42
S27	57	M	94	7	L	6.467	6.927	7.715	8.472
S28	79	M	18	61	L	7.202	7.374	7.845	7.274
S30	69	F	50	11	R	6.171	7.277	6.827	8.366
S31	58	M	25	16	L	5.883	6.865	5.821	7.773
S34	65	F	21	15	L	6.05	7.644	5.74	7.961

Appendix E: Summary of individual participants (cont.)

Table 3. Participant demographics (cont.)

Participant number	Ipsilesional precentral gyrus thickness (mm)	Contralesional precentral gyrus thickness (mm)	WMFT rate score (affected arm)	WMFT rate score (unaffected arm)	MAL-QOM score	Accelerometer counts (activity kilocounts)
S01	2.321	2.159	57.6	76.4	4.67	68.49
S02	2.031	2.045	52.0	64.2	2.68	70.6
S03	2.166	2.162	34.0	40.7	4.1	62.27
S05	1.791	2.222	10.3	65.8	-	-
S07	2.435	2.589	58.2	61.5	4.43	350.21
S08	2.259	2.32	38.4	71.4	2.71	230.81
S12	2.07	2.039	64.1	63.6	-	-
S14	2.588	2.435	42.7	61.6	-	-
S15	1.674	1.832	44.4	52.4	4.0	236.82
S21	1.694	2.24	46.4	52.9	3.5	160.27
S22	1.898	2.413	11.3	75.9	0.0	18.75
S23	2.185	2.367	53.0	62.8	-	-
S27	2.028	2.401	9.2	47.5	-	-
S28	2.441	2.324	45.0	54.9	4.64	224.02
S30	1.567	2.168	0.42	40.3	-	-
S31	1.979	2.46	3.48	44.2	0	90.04
S34	1.808	2.406	12.3	44.4	-	-

Appendix F: Participant lesion locations

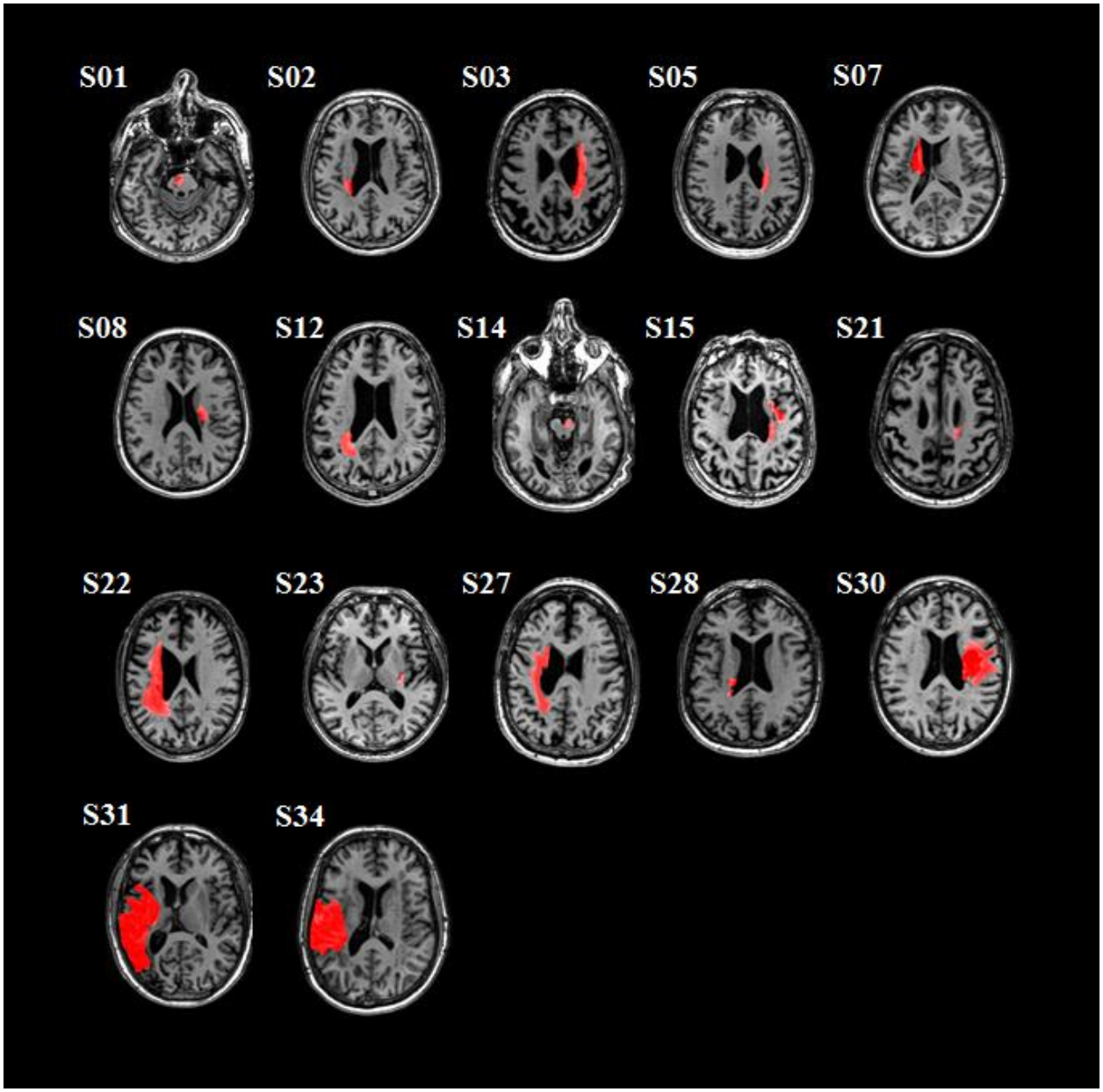

Figure 6. Individual participant lesion locations. Each image shows the section of the brain that contains the largest portion of the lesion. Each participant's T1-weighted anatomical scan was examined to ensure that the lesion did not extend in to the voxel for H^1-MRS. None of the participants contained lesions that extended in to the motor cortical areas.

Appendix G: Correlation analyses

Table 4. Summary of correlation analyses

		tNAA	**Glx**	**Precentral gyrus thickness**
tNAA	Pearson correlation	1	0.092	0.782
	Sig. (2-tailed)		0.725	0.000*
	N	17	17	17
Glx	Pearson correlation	0.092	1	0.063
	Sig. (2-tailed)	0.725		0.809
	N	17	17	17
Precentral gyrus thickness	Pearson correlation	0.782	0.063	1
	Sig. (2-tailed)	0.000*	0.809	
	N	17	17	17

Appendix H: Regression analyses

Table 5. Regression models to examine correlates with upper-extremity Wolf Motor Function Test score

Ipsilesional Hemisphere	**Predictors**	**R^2**	**F statistic**	**Significance**	**βAge (sig)**	**βPSD (sig)**	**βNAA (sig)**	**βGlx (sig)**	**βCortical thickness (sig)**
Model									
1	Age, PSD	0.131	1.051	0.375	0.348 (0.186)	0.125 (0.625)	-	-	-
2	Age, PSD, NAA, Glx	0.535	3.445	0.043	0.369 (0.113)	0.040 (0.857)	0.640 (0.008)	-0.137 (0.556)	-
3	Age, PSD, NAA, Glx, Cortical thickness	0.540	2.580	0.088	0.380 (0.121)	0.072 (0.772)	0.536 (0.165)	-0.150 (0.542)	0.125 (0.731)

Contralesional Hemisphere	**Predictors**	**R^2**	**F statistic**	**Significance**	**βAge (sig)**	**βPSD (sig)**	**βNAA (sig)**	**βGlx (sig)**	**βCortical thickness (sig)**
Model									
1	Age, PSD	0.195	1.698	0.219	-0.152 (0.536)	0.405 (0.114)	-	-	-
2	Age, PSD, NAA, Glx	0.371	1.767	0.200	-0.163 (0.557)	0.515 (0.052)	0.312 (0.243)	-0.376 (0.173)	-
3	Age, PSD, NAA, Glx, Cortical thickness	0.371	1.298	0.333	-0.154 (0.625)	0.526 (0.089)	0.302 (0.322)	-0.377 (0.193)	0.027 (0.938)

Appendix H: Regression analyses (cont.)

Table 6. Regression models to examine correlates with upper-extremity accelerometer activity counts

Ipsilesional Hemisphere	**Predictors**	**R^2**	**F statistic**	**Significance**	**βAge (sig)**	**βPSD (sig)**	**βNAA (sig)**	**βGlx (sig)**	**βCortical thickness (sig)**
Model									
1	Age, PSD	0.391	2.243	0.177	-0.414 (0.203)	-0.483 (0.146)	-	-	-
2	Age, PSD, NAA, Glx	0.652	2.337	0.188	-0.297 (0.431)	-0.585 (0.094)	0.540 (0.111)	-0.126 (0.738)	-
3	Age, PSD, NAA, Glx, Cortical thickness	0.780	2.840	0.167	-0.389 (0.282)	-0.758 (0.052)	1.181 (0.072)	-0.053 (0.876)	-0.732 (0.201)

Contralesional Hemisphere	**Predictors**	**R^2**	**F statistic**	**Significance**	**βAge (sig)**	**βPSD (sig)**	**βNAA (sig)**	**βGlx (sig)**	**βCortical thickness (sig)**
Model									
1	Age, PSD	0.070	0.262	0.777	-0.220 (0.566)	-0.154 (0.686)	-	-	-
2	Age, PSD, NAA, Glx	0.393	0.808	0.570	-0.610 (0.240)	0.149 (0.723)	-0.308 (0.510)	-0.682 (0.168)	-
3	Age, PSD, NAA, Glx, Cortical thickness	0.399	0.532	0.748	-0.622 (0.291)	0.081 (0.889)	-0.235 (0.714)	-0.654 (0.252)	-0.125 (0.845)

Appendix H: Regression analyses (cont.)

Table 7. Regression models to examine correlates with Motor Activity Log- Quality of Movement Scores

Ipsilesional Hemisphere	**Predictors**	**R^2**	**F statistic**	**Significance**	**βAge (sig)**	**βPSD (sig)**	**βNAA (sig)**	**βGlx (sig)**	**βCortical thickness (sig)**
Model									
1	Age, PSD	0.167	0.701	0.528	0.408 (0.275)	0.028 (0.937)	-	-	-
2	Age, PSD, NAA, Glx	0.765	4.078	0.078	0.676 (0.063)	-0.088 (0.722)	0.812 ((0.017)	-0.338 (0.301)	-
3	Age, PSD, NAA, Glx, Cortical thickness	0.864	5.078	0.070	0.596 (0.073)	-0.238 (0.334)	1.372 (0.023)	-0.274 (0.338)	-0.640 (0.164)

Appendix I: Regression plots

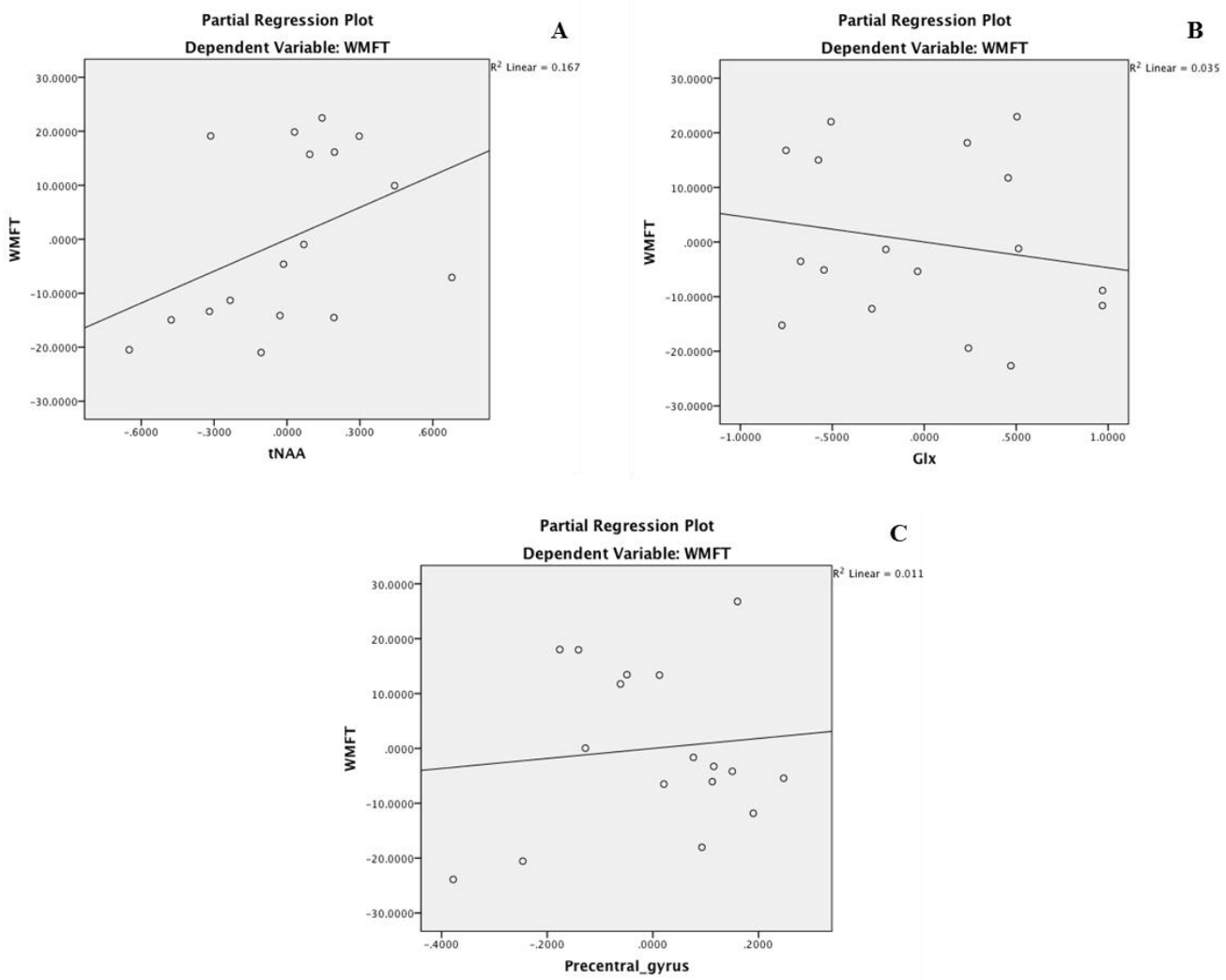

Figure 7. Partial residual plot illustrating the relationship between WMFT score of the hemiparetic UE and ipsilesional: A) tNAA concentration after accounting for age and post-stroke duration B) Glx concentration after accounting for tNAA concentration, age and post-stroke duration and C) precentral gyrus thickness after accounting for tNAA concentration, Glx concentration, age and post-stroke duration.

APPENDIX I: Regression plots (cont.)

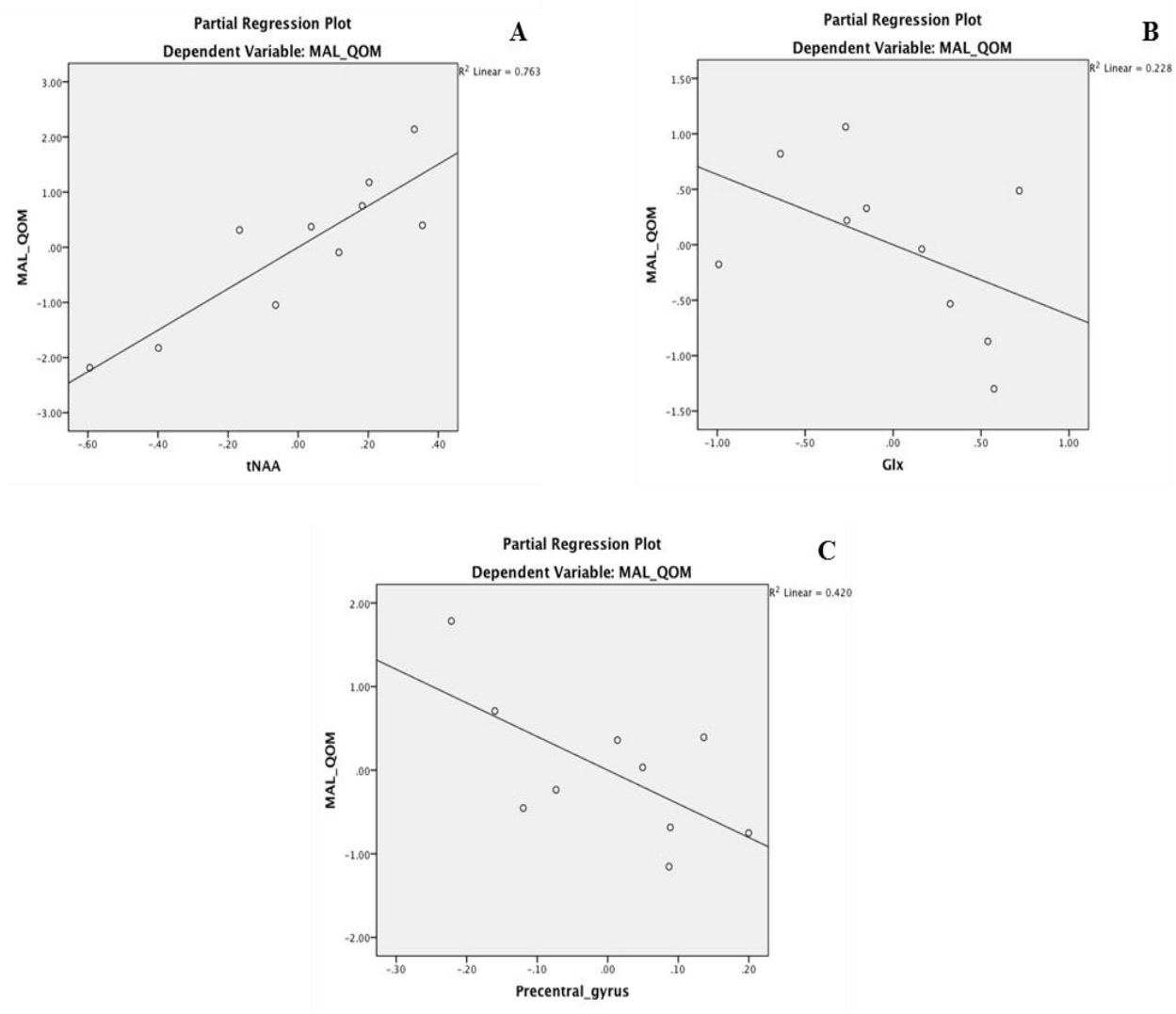

Figure 8. Partial residual plot illustrating the relationship between MAL-QOM score and ipsilesional A) tNAA concentration after accounting for age and post-stroke duration B) Glx concentration after accounting for tNAA concentration, age and post-stroke duration and C) precentral gyrus thickness after accounting for tNAA concentration, Glx concentration, age and post-stroke duration

Appendix I: Regression plots (cont.)

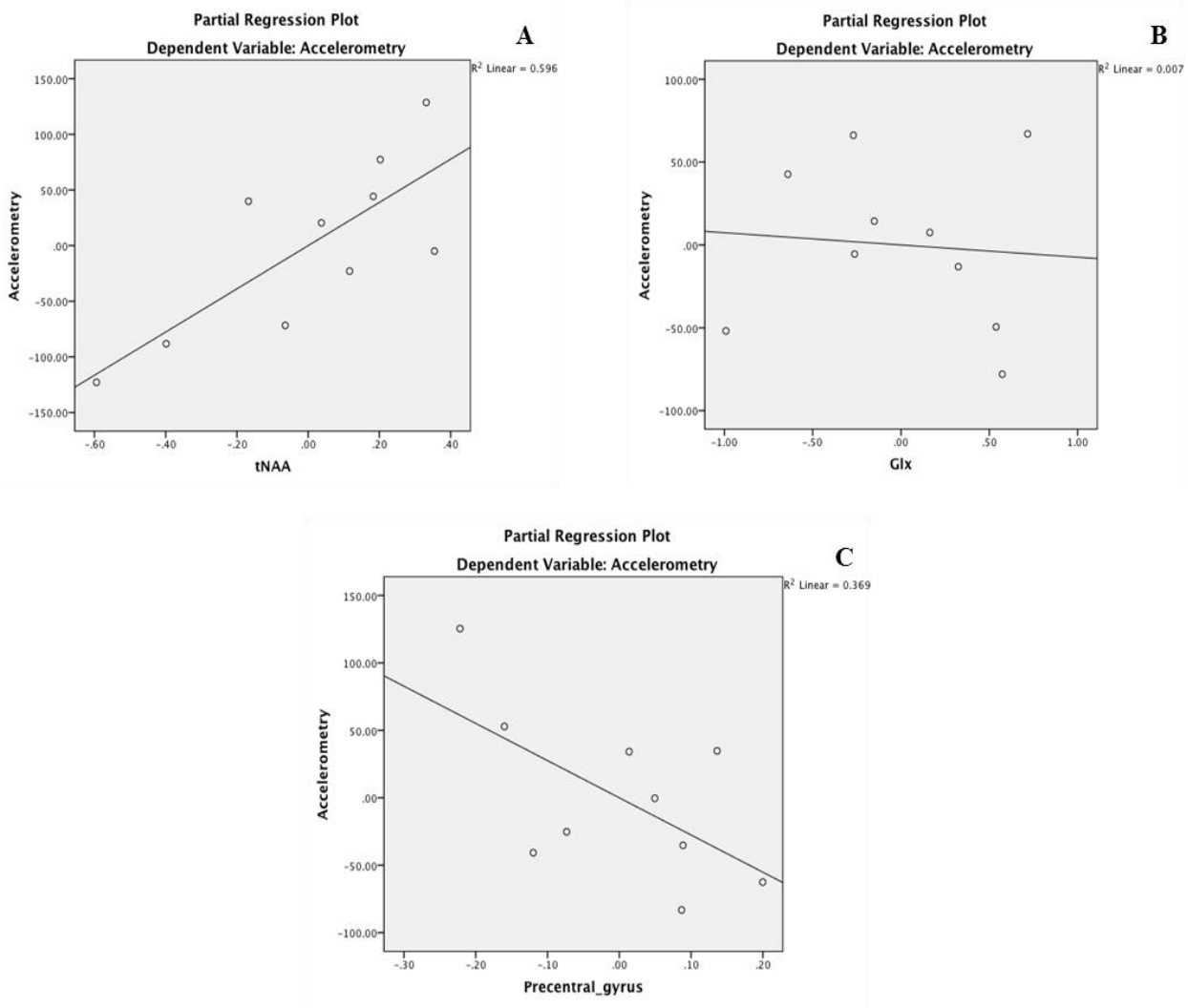

Figure 9. Partial residual plot illustrating the relationship between activity counts of the hemiparetic UE and ipsilesional A) tNAA concentration after accounting for age and post-stroke duration B) Glx concentration after accounting for tNAA concentration, age and post-stroke duration and C) precentral gyrus thickness after accounting for tNAA concentration, Glx concentration, age and post-stroke duration.

Appendix J: The Motor Activity Log

Motor Activity Log 14 (MAL-14)

1. General:

This instrument is a structured interview intended to examine how much and how well the subject uses their **more-affected arm** outside of the laboratory setting. Participants are asked standardized questions about 1) do they use the more-affected arm 2) the amount of use of their **more-affected arm** (Amount Scale or AS) and 3) the **quality of their movement** (How Well Scale or HW) during the functional activities indicated. The scales are printed on separate sheets of paper and are placed in front of the participant during test administration. Participants should be told that they can give half scores (i.e., 0.5, 2.5, 3.5) if this is reflective of their ratings.

2. Rating Scales:

- Both the AS and HW scales are used during all test administrations.
- In all administrations, **begin with the "AS scale" and ask participants to rate all tasks using the AS scale first**.

3. Asking Questions

Step One: Read the MAL instructions to the participant and explain the rating scales. Answer any questions that the subject may have. The tester should remind the participant that the questions on the MAL pertain to what they actually do outside the treatment setting – not what they think they may be able to do.

Instructions for Participant:

- *The purpose of this measure is to examine how you use your more-affected arm at home.*
- *Please note that you can give half ratings if that best describes your performance of the activity in question.*
- *It is important that you realize that these questions are about what you actually do at home and in the community – not what you think you may be able to do with your weaker arm. There are no right or wrong answers; simply select the ratings you believe best describes what you do.*

Step Two: The assessor should inquire about the use of the more-affected arm for each activity using the following questions and then the amount of use for the same activity

Considering your activities during the past week, did you use your weaker arm to... (state the activity)?" If no, then ask why and direct the participant to the list of possible reasons why the arm was not used. For scoring on the recording form, use the codes to categorize the participant's response. It is desirable to have these codes printed on a separate sheet so that it is easy for the participant to make a selection". If answer "Yes", go to step three. *Halfway, at question 7, remind them again "Just a reminder that these questions are about what you actually do at home and in the community – not what you think you may be able to do with your weaker arm".*

Codes for recording "no" responses:

1. I used the unaffected arm entirely. (Assign "0").
2. Someone else did it for me. (Assign "0").
3. I never do that activity, with or without help from someone else because it is impossible or not relevant. For example, combing hair for people who are bald. (Assign "N/A" and drop from list of items). When a task is impossible, such as combing the hair if the person is bald, the "N/A" score should be used. However, "N/A" should be used sparingly since many times a subject might indicate that an activity is impossible, but it is actually not being performed because it is either very difficult for the participant, inconvenient, or requires increased time for completion. It has been found that when subjects progress, they will sometimes begin doing tasks that they may have previously identified as impossible
4. I sometimes do that activity, but did not have the opportunity since the last time I answered these questions. (code as No_Opp).
5. Non-dominant hand hemiparesis and prior to stroke would have only used that hand. (Assign "N/A" and drop from list of items).

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Step Three: Rating the Amount of Use and How Well

a. Amount Rating: *Can you tell me **how much** you do each of the activities with your weaker arm over the last week. For example, you might have used your more-affected hand to pick up a glass and drink only rarely over the last week. If that was the case, what do you think you would have rated the Amount? (it should be 1.5 or 2). Do you understand when I ask you how much you used your weaker arm? Using the **Amount Rating Scale**, tell me how you would rate the amount you used your weaker arm to... (state the activity)."* Once the participant selects a rating, verify their response by repeating the selected rating in the following manner; *" So, you believe that you (read the description of the selected AS rating) – Is that correct?"*

Amount Scale:

0	Did not use my weaker arm (not used).
0.5	
1	Occasionally used my weaker arm but only very rarely (very rarely).
1.5	
2	Sometimes used my weaker arm but did the activity most of the time with my stronger arm (rarely).
2.5	
3	Used my weaker arm about half as much as before the stroke (half pre-stroke).
3.5	
4	Used my weaker arm almost as much as before the stroke (3/4 pre-stroke).
4.5	
5	Used my weaker arm as often as before the stroke (same as pre-stroke).

b. How Well Rating: *Now I want you to rate **how well** you used your weaker arm over the last week.* For example, you may not have used your weaker hand very much last week, but when you did use it, your movements of the hand was really quite good and almost normal. In this case, can you tell me what the rating would have been for how well you use your hand? (It should be 3.5-4). Do you understand what I mean by "how well you used your weaker arm"? Okay, how would you rate **how well** you used your weaker arm over the last week to ...? Once the participant selects a rating, verify the selected rating in the following manner; *" So, you believe that you... (read the selected HW rating scale) - Is that correct?"* Once they agree, record the response in the blank HW space provided for that question during **"Initial"** and **"Baseline"** assessments.

During the **Initial** assessment and after the participant provides an HW rating, ask the subject to demonstrate/pantomime an approximation of the first 3 activities scored above 0 by saying, *"Please show me how you do that activity."* During the "Initial" assessment, demonstration of the performance of an activity should also be requested whenever the tester is unsure of what the participant means by a rating. The demonstration should be carried out after the participant attempts to rate the activity and only when using the HW rating scale. Observation of the pantomime of an activity allows the tester an opportunity to discuss the participant's HW rating in order to set an agreed-upon rating frame of reference. If obvious discrepancies exist between what is observed and the rating provided, the tester should discuss the rating with the participant to develop a common frame of reference (i.e., "You rated that activity a "5". However, you moved your arm very slowly to do the activity. So, for this project that would be more like a "2". Do you agree?"). The final rating is determined by the participant.

On the subsequent assessments, do not request the demonstration/pantomime. If the current score is at least 1 point different from the previous score, say "last time, you scored X on the Amount, do you feel you used your arm/hand less/more than last month?

How Well Scale

0	My weaker arm was not used at all for that activity (not used).
0.5	
1	My weaker arm was moved during that activity but was not helpful (very poor).
1.5	
2	My weaker arm was of some use during that activity but needed some help from the stronger arm, moved very slowly, or with difficulty (poor).
2.5	
3	My weaker arm was used for that activity but the movements were slow or were made only with some effort (fair).
3.5	
4	The movements made by my weaker arm for that activity were almost normal but not quite as fast or accurate as normal (almost normal).
4.5	
5	The ability to use my weaker arm for that activity was as good as before the stroke (normal).

4. **Scoring**.

Motor Activity Log-14 (UE MAL) Score Sheet

	Items	Amount	How well	If "No", why? (Use code)
1	Hold a book			
2	Use a towel to dry self			
3	Pick up a glass (doesn't include drinking)			
4	Brush teeth (doesn't include preparing toothbrush)			
5	Shave/Put on Make-up (or lotion)			
6	Open door with a key			
7	Write/type			
8	Steady self			
9	Put arm through clothing			
10	Carry object			
11	Grasp fork/spoon			
12	Comb hair			
13	Pick up a cup by a handle			
14	Button clothes			

Codes for recording "no" responses:

1. "I used the unaffected arm entirely." (Assign "0").
2. "Someone else did it for me." (Assign "0").
3. "I never do that activity, with or without help from someone else because it is impossible." For example, combing hair for people who are bald. (Assign "N/A" and drop from list of items).
4. "I sometimes do that activity, but did not have the opportunity since the last time I answered these questions." (code No_Opp).
5. Non-dominant hand hemiparesis. (Assign "N/A" and drop from list of items).

Appendix K: The Wolf Motor Function Test

Compensatory Brain Activation After Stroke

WOLF MOTOR FUNCTION TEST

Patient ID#:_____

Date:___ ___ / ___ ___ / ___ ___ Time: _____

Examiner:_____

Testing Instructions and Scoring

The WMFT consists of 17 tasks performed without the subject's awareness of how the components are defined or scored. All tasks will use both arms, starting with the unaffected arm first. Each task is timed and rated by Functional Ability (0 – 5 Ordinal Scale). Subjects are given a maximum time limit of 120 seconds to complete the assigned task.

Score – Functional Ability

0	Does not attempt with upper extremity (UE) being tested
1	UE being tested does not participate functionally; however, attempt is made to use the UE
2	Does, but requires assistance of the UE not being tested for minor readjustments or changes of position, or requires more than two attempts to complete, or accomplishes very slowly
3	Does, but movement is influenced to some degree by synergy or is performed slowly or with effort
4	Does, movement is close to normal*, but slightly slower; may lack precision, fine coordination or fluidity
5	Does; movement appears to be normal

*Normal – the less involved UE can be utilized as an available index comparison, with pre-morbid UE dominance taken into consideration

Read the follow statement aloud to the patient:

Today we are going to take a look at how you are able to use your arm. Let me tell you how we are going to do this. First, I will give you instructions on how to do the task, and then I will show you how to do it. I will describe and demonstrate each task 2 times. Do not practice the task while I'm describing and demonstrating it. However, I will be happy to clarify any confusing points. Then I will say, "Ready, set, go", and you will do the task. It is important that you do not start until I say "go", otherwise, we will need to repeat the entire task. Each of the activities you will be asked to do should be carried out as rapidly as possible. You can work on each task for up to two minutes. We ask that you attempt each part of the test even if you do not think you can do it. If you are unable to carry out a task, then we will go on to the next one. Again, try to do each task as rapidly as possible. Do you have any questions?

UNAFFECTED HAND

Task	Time	Functional Ability	Comment
1. Forearm to table (side)	_____	0 1 2 3 4 5	_____
2. Forearm to box (side)	_____	0 1 2 3 4 5	_____
3. Extend elbow (side)	_____	0 1 2 3 4 5	_____
4. Extend elbow (weight)	_____	0 1 2 3 4 5	_____
5. Hand to table (front)	_____	0 1 2 3 4 5	_____
6. Hand to box (front)	_____	0 1 2 3 4 5	_____
7. Weight to box	_____ lbs.		_____
8. Reach and retrieve	_____	0 1 2 3 4 5	_____
9. Lift can	_____	0 1 2 3 4 5	_____
10. Lift pencil	_____	0 1 2 3 4 5	_____
11. Lift paper clips	_____	0 1 2 3 4 5	_____
12. Stack checkers	_____	0 1 2 3 4 5	_____
13. Flip cards	_____	0 1 2 3 4 5	_____
~~14. Grip strength~~	_____ kgs.		_____
15. Turn key in lock	_____	0 1 2 3 4 5	_____
16. Fold towel	_____	0 1 2 3 4 5	_____
17. Lift basket	_____	0 1 2 3 4 5	_____

AFFECTED HAND

Task	Time	Functional Ability	Comment
1. Forearm to table (side)	_____	0 1 2 3 4 5	_____
2. Forearm to box (side)	_____	0 1 2 3 4 5	_____
3. Extend elbow (side)	_____	0 1 2 3 4 5	_____
4. Extend elbow (weight)	_____	0 1 2 3 4 5	_____
5. Hand to table (front)	_____	0 1 2 3 4 5	_____
6. Hand to box (front)	_____	0 1 2 3 4 5	_____
7. Weight to box	_____lbs.		_____
8. Reach and retrieve	_____	0 1 2 3 4 5	_____
9. Lift can	_____	0 1 2 3 4 5	_____
10. Lift pencil	_____	0 1 2 3 4 5	_____
11. Lift paper clips	_____	0 1 2 3 4 5	_____
12. Stack checkers	_____	0 1 2 3 4 5	_____
13. Flip cards	_____	0 1 2 3 4 5	_____
14. Grip strength	_____kgs.		_____
15. Turn key in lock	_____	0 1 2 3 4 5	_____
16. Fold towel	_____	0 1 2 3 4 5	_____
17. Lift basket	_____	0 1 2 3 4 5	_____

Clinician Signature

Date: ___ ___ / ___ ___ / ___ ___
Month Day Year

Appendix L: Study Consent

THE UNIVERSITY OF BRITISH COLUMBIA

School of Rehabilitation Sciences

Faculty of Medicine

T325-2211 Wesbrook Mall

Vancouver, British Columbia V6T 2B5

Title of Study:

Interhemispheric contributions to neuroplasticity and motor learning after stroke
Consent Form for Individuals with Stroke

Principal Investigator: Lara Boyd, PT, PhD. Department of Physical Therapy, Brain Behaviour Laboratory, Faculty of Medicine, UBC

Co-Investigator: Michael Borich, PhD, Department of Physical Therapy, Brain Behavior Laboratory, Faculty of Medicine, Todd Handy, PhD, Department of Psychology, Faculty of Arts, UBC
Sean Meehan, PhD, Department of Physical Therapy, Brain Behaviour Laboratory, Faculty of Medicine, UBC

Team Members: Jodi Edwards, Katie Wadden, Paul Jones, Tamara Koren, Cameron Mang, Kate Brown, Sonia Brodie**,** Katharine Cheung**,** Brenda Wessel

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Invitation to Participate: You are being invited to participate in a research study that investigates the brain structure and function across the lifespan.

Participation is Voluntary: You do not have to participate in this research study. It is important that before you make a decision to participate, you read the rest of this form. Please read the following form carefully and ask questions if anything is not clear. The consent form will tell you about the study, why the research is being done, and what will be done during the study and the possible risks, benefits, and discomforts.

If you wish to participate, you will be asked to sign this form. If you do decide that you would like to participate, you are still free to withdraw at any time and without giving any reasons for your decision. If you do not wish to participate, you do not have to provide any reason for the decision nor will you lose the benefit of any medical care to which you are entitled or presently receiving.

Please take time to read the following information carefully and to discuss it with your family, friends and doctor before you decide.

Purpose

The purpose of this study is to determine whether pairing brain stimulation with motor learning can enhance rehabilitation that helps people recover the use of their stroke affected arm as compared to rehabilitation alone (i.e. without brain stimulation). These efforts should lead to the development of new rehabilitation approaches that can help restore normal patterns of brain activity after stroke.

Who Can Participate in this Study?

You have been identified because you are between the ages of 30-95, have no neurological disorders, and have the ability to understand English. If you agree to take part in the study, Dr Boyd or her associates will determine if you have any condition that will prevent you from being in the study. Screening should take no more than 5 minutes.

Who Should Not Participate in this Study?

You should not participate in this study if you have a history of seizure, epilepsy, neurodegenerative disorder, head trauma, or a psychiatric diagnosis. If you are pregnant, claustrophobic (have a fear of enclosed or narrow spaces) or have metallic objects in your head or heart you should not participate.

What does the study involve?

If you are eligible and decide to participate in this study, you will come to the Brain Behavior Lab for up to 9 visits. The first visit (Day 1) will be expected to last at least 1 hour and will be scheduled independently of the other 7-8 visits. The second and third days (Days 2 and 3) will be expected to last approximately 3 hours. The fourth, fifth, sixth, seventh, and eighth days (Days 4-8) are anticipated to last 60 minutes. Your final visit will last approximately 3 hours. Days 4-9 will be spaced over a three-week period.

You will be randomly assigned to one of three experimental groups according to a random number generator where you will have an equal chance (like flipping a coin) of being placed into one of the three groups. Each group will conduct the same activities but on Days 4, 5, 6, 7 and 8 will be stimulated in different ways. One group will receive stimulation that reduces brain activity in the part of the brain that responds to touch of the arm. One group will receive stimulation that reduces activity in the part of the brain that sends commands to the arm muscles.

The third group will receive stimulation over the vertex of the head that is the part of the brain that sends commands to the leg muscles. There is an equally probable chance that you will end up in any of the groups. The investigators and team members do not have any influence over group assignment. During the study you will not know which group you are in. At the end of the study we will inform you of your group assignment.

On the first day of the study, you will be asked to come to the Purdy Pavilion of University of British Columbia Hospital to have an anatomical Magnetic Resonance Image (MRI) which will provide us with a picture of your brain. This is not a diagnostic MRI, but rather will help us in guiding the stimulation you will receive on the successive days. For the remaining days you will be asked to come to the Brain Behavior Lab (T142a Koerner Pavilion, University of British Columbia Hospital) where you will receive brain stimulation to assess the excitability of your brain before and immediately after brain stimulation (Days 2, 3, and 9). On Day 4-8 you will practice a motor task after a session of brain stimulation.

On day 1 you will be asked to come to Purdy Pavilion where one of the research staff will meet you to explain the study. Next, the MRI scan will be explained to you before you enter the scanner. You will be asked to leave any metal objects (e.g. watches, bracelets, rings, and metal eyeglasses) at home or in lockers provided in the waiting room of the MRI centre. You will also be asked to remove any articles of clothing with metal inserts or clasps before entering the magnet room. Please ask the study staff about anything you are unsure of. You will be positioned on the table of an MRI scanner, lying on your back, and a magnetic resonance (MR) coil (specially designed loop of insulated wire) will be placed near your head. You will then be slid into the centre of the scanner.

It is possible that you may feel uncomfortably confined once inside the MRI machine. This feeling usually passes within a few minutes as the study staff talk with you and the study begins. However, if this feeling persists, you can tell the investigators over the intercom and you will be removed immediately from the machine. During the scan you will hear banging noises, which are normal. We will ask you to wear headphones or earplugs to ensure that your hearing is not affected by the scan. The scan will take about 1 hour, with set up time included.

On Days 2 and 3 the excitability of your brain, particularly the part that make sense of touch and movement (called somatosensory cortex) and the part of the brain that controls your muscles as well as your ability to perform a number of tasks (called motor cortex) will be assessed immediately before and 10 minutes, 30 minutes and 60 minutes after Transcranial Magnetic Stimulation (TMS). TMS consists of a series of pulses for 40 seconds. TMS will be applied over the outside of your head. Stimulation is largely painless; however, there is a risk of mild pain, such as headache, local pain, neck pain, toothache, or a tugging or tingling feeling on your scalp during this time. On Day 2 the excitability of your somatosensory cortex will be measured using Electroencephalography (EEG). EEG uses a special cap that you wear on your

head and contains little metal discs that sit over your hair and are able to measure the electrical activity of the brain. In addition, you will be asked to perform a continuous motor task where you will be asked to move a computer mouse between targets that appear on a computer screen.

On Day 3 the excitability of your motor cortex will be measured using single independent pulses of TMS. Brain excitability will be measured by delivering single pulse of TMS over the area of your brain that controls the muscles in your forearm. These pulses feel like a light tap on the head. When these pulses are applied they will cause a small reaction in the muscle that will make your arm move. We will use electrodes placed on the skin over the muscle in your arm to measure how much the muscle reacts to the TMS (a technique called "electromyography"). In addition you will also be asked to perform a number of quick tests to look at how well you use your stroke affected limb.

On Days 4 and 8 single pulses of TMS will be applied over the outside of your head to measure the excitability of your motor cortex before and after motor task practice. This TMS will be similar to that on Day 3. TMS will also be applied over the outside of your head in a repetitive manner. On these days a series of TMS pulses will be applied before you perform four blocks (about 20 minutes total duration) of a learning task. The learning task will be identical to the continuous motor task that you performed during Day 2.

On Day 9 we will repeat the EEG measurements from Day 2 while you perform one block of the learning task. In addition we will also repeat the sensory tests from Day 2. We will also look at the excitability of your motor cortex using the same method as described on Day 3 and repeat the exercises that looks at your ability to use your stroke affected limb.

For all cases where TMS is applied (Days 2-9) you will be seated comfortably in a reclining chair. A figure of eight coil (6 inches long) will be fixed to a frame that will hold it in place over your head.

Future studies: We would like to know if you are interested in learning about future studies. If Dr. Boyd thinks you might qualify for another study by her or her colleagues, she will contact you directly by mail or telephone and ask if you are interested. If you choose not to take part in future studies you should tell her. There will be no impact on you if you choose not to take part. You are not giving permission to do any future studies in this consent form.

Are you willing to be contacted in the future about participation in other studies?
_____ YES _____ NO

What Are Possible Harms and Side-Effects of Participation

These procedures will be conducted according to published safety standards. Dr. Boyd or her associates have discussed this research with you and have described them as follows:

MRI: There is very little known risk associated with undergoing an MRI scan. MRI is used routinely in hospitals around the world. A small number of people may find lying still inside the MR scanner uncomfortable and stressful. If this occurs then you will be brought out of the scanner and the study stopped. Some people are also uncomfortable being in small places (i.e., claustrophobia). Because the MRI scanner is a small space you may also be uncomfortable lying inside it. If you do feel this way you will be brought out of the scanner and the study will be halted. The MRI also makes loud noises that you may find uncomfortable. You will not be able to participate in study if you have any metal or surgical implants that may be affected by the strong magnetic fields used in the MRI process or may cause tissue damage associated with dislodging the metal and/or for the objects to become heated during the scan and cause a burn. Most implants are not affected by MRI, but if you have any of the following you will not be able to participate in the MRI study:

- pacemaker
- brain aneurysm clip
- cochlear implant
- recent surgery or tattoos within the past 6 weeks
- possibility of pregnancy
- electrical stimulator for nerves or bones
- implanted infusion pump

If you have any of the following, please let us know as soon as possible and we will get your surgical report and contact MRI technologist to ensure your safety:

- history of any eye injury involving metal fragments
- you have been a metal worker (grinding, machining, or welding)
- artificial heart valve
- orthopedic hardware (artificial joint, plate, screws, rods)
- other metallic prostheses
- coil, catheter of filter in any blood vessel
- ear or eye implant
- shrapnel, bullets, or other metallic fragments
- medication releasing skin patches (nicotine, birth control, nitroglycerine)

TMS: **For single / paired pulse TMS:**

Safety standards for the application of TMS have been developed and will be followed by trained operators during this study to minimize the risk. In accordance with these standards, the TMS machine will always be run at a rate and a frequency that are known to be safe.

A member of the study team has discussed this research with you and has described the risks as follows:

§ There is a potential risk of provoking a seizure in people with a history of seizures (e.g. epilepsy). Though there has never been a report of a seizure associated with the type of TMS you will receive in this study (ie., single and paired-pulse TMS),

you will not be eligible to participate in this study if you have such history.

§ There is a risk of headache, scalp pain, toothache or scalp numbness associated with single and paired-pulse TMS. Each of these side effects is transient (ie., does not last).

§ The clicks associated with single and paired-pulse TMS are loud and could potentially damage your hearing. To minimize this risk you will be asked to wear earplugs throughout the testing session.

Risk statement for repetitive TMS:

Safety standards for the application of TMS have been developed and will be followed by trained operators during this study to minimize the risk. In accordance with these standards, the TMS machine will always be run at a rate and a frequency that are known to be safe.

A member of the study team has discussed this research with you and has described the risks as follows:

§ There is a small but real risk of provoking a seizure in people with a history of seizures (e.g. epilepsy) with repetitive TMS. The risk of seizure is less than 1% in people who have no history of epilepsy. However, you will not be eligible to participate in this study if you have any past history of seizure. Also it is possible that repetitive TMS may change the current in other electrical devices such as pace-makers. Because of this risk you will not be able to participate if you have a pace-maker or other implanted electrical device.

§ There is a risk of headache, scalp pain, toothache or scalp numbness associated with repetitive TMS. Each of these side effects is transient (ie., does not last).

§ The clicks associated with repetitive TMS are loud and can damage your hearing. To minimize this risk you will be asked to wear earplugs throughout the testing session.

§ There is a risk that the repetitive TMS coil may heat up during brain stimulation. To minimize this risk the coil is air cooled and will automatically shut off if it becomes heated.

EEG: Collection of EEG involves application of electrodes over the scalp to measure brain activity. All EEG electrodes are surface electrodes and do not actually contact the skin. A conductive gel provides the contact between the skin and the recording electrodes. In rare instances it is possible that your skin may be sensitive to the conductive gels or rubbing alcohol used for surface recordings. In such cases a skin rash is possible. The conductive gel is water-soluble and washes out quickly with warm water and shampoo.

Sensorimotor Task: There are no known risks associated with performing this short-duration computer based task. If at any point you feel uncomfortable you can tell the researchers and they will stop the testing.

There may be other risks that have not yet been identified, and unexpected side effects that have not been previously observed may occur.

What are the Benefits to You of Participating in the Study

There is direct no benefit to you for participating in this study. It is hoped that additional information gained in this research study may be useful in the treatment of other patients with brain damage. You will be informed if any significant new findings develop during the course of the study that may affect your willingness to participate in this study.

Payments to Subjects

You will receive $35 for each visit for giving your time up and to offset your parking and/or travel expenses incurred to participate in this study. You will not need to provide any receipts to be reimbursed.

In the Event of an Injury

In the event that you experience a serious side effect during this study during normal business hours, you should immediately contact Dr. Boyd. If it is after 5:00 p.m., a holiday or weekend, you should report to an emergency room. Signing this consent form in no way limits your legal rights against the sponsor, investigators, or anyone else. In case of a serious medical event resulting from this study, please report to an emergency room and inform them that you are participating in a research study and Lara Boyd (Principal Investigator) can be contacted for further information.

Confidentiality

Your confidentiality will be respected. No information that discloses your identity will be released or published without your specific consent to the disclosure. However, research records and medical records identifying you may be inspected in the presence of the Investigator or his or her designate by representatives of Health Canada and the UBC Research Ethics Board for the purpose of monitoring the research. However, no records which identify you by name or initials will be allowed to leave the Investigators' offices.

If the results of this study are published or presented in public, information that identifies you will be removed. If you decide not to sign the form, you cannot be in the study.

Your study-related health information such as which group you have been randomized into will be used at UBC only by Dr. Boyd, and members of her research team who are listed on this consent form. This is important to allow members of the research team to communicate about which group you are participating in for this research. Your permission to use and disclose your health information remains in effect until the study is complete and the results are analyzed. After that time, information that personally identifies you will be removed from the study records.

Questions

You have read the information in this form. Dr. Boyd or her associates have answered your question(s) to your satisfaction. You know if you have any more questions after signing this you may contact Dr. Boyd or one of her associates. If you have any questions about your rights as a research subject, you may call the Research Subject Information Line in the University of British Columbia Office of Research Services.

You have a right to change your mind about allowing the research team to have access to your health information. If you want to cancel permission to use your health information, you should either verbally indicate your withdrawal or send a request to Dr. Boyd. If you cancel permission to use your health information, you will be withdrawn from the study. The research team will stop collecting any additional information about you. The research team may use and share information that was gathered before they received your cancellation.

Consent

We, (Dr. Boyd and/or her associates) have given you information about this research study and have explained what will be done and how long it will take. We have explained any inconvenience, discomfort or risks that may be experienced during this study. I freely and voluntarily consent to participate in this research study. I have read and understand the information in this form and have had an opportunity to ask questions and have them answered. **I have been told that I will receive a signed and dated copy of the consent form to keep for my records.**

I have chosen not to receive a copy of this consent form _____ (Initial Here)

_____

Type/Print Subject's Name

_____ _____

Signature of Subject Date

_____

Type/Print Name of Witness

_____ _____

Signature of Witness Date

_____

Type/Print Name of Person Obtaining Consent

_____ _____

Signature of Person Obtaining Consent Date

_____

Type/Print Name of Principal Investigator

_____ _____

Signature of Principal Investigator Date