FUNCTIONAL AND STRUCTURAL REORGANIZATION IN RELATION TO FUNCTIONAL OUTCOMES AFTER STROKE: INSIGHTS FROM MAGNETIC RESONANCE IMAGING

WONG WAN WA

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Functional and Structural Reorganization in Relation to Functional Outcomes after Stroke: Insights from Magnetic Resonance Imaging

WONG Wan Wa

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CERTIFICATE OF ORIGINALITY

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Abstract

In view of the great impact of stroke, the need and importance of good recovery has been highlighted. Currently, based on the evidences stating the potential for neuroplasticity and its relation to stroke recovery, a restorative and neuroscience-based approach to stroke rehabilitation, such as motor imagery training, has been developed and recommended to the rehabilitation specialists. However, the mechanism of the shaping of neuroplasticity in relation to stroke recovery is still not fully understood. In addition, a number of factors, such as the extent of damage in influencing the structural and functional brain networks, have been identified to have certain impact on the potential for neuroplasticity and the capacity for receiving benefits from rehabilitation. Thus, more understanding of the neural mechanism that underpins an effective and restorative approach to stroke rehabilitation becomes necessary and important. The emerging of neuroimaging techniques can therefore help to study this mechanism of post-stroke recovery. Magnetic resonance imaging (MRI), which is one of these techniques, has been shown having a great potential to give important insights into post-stroke recovery mechanisms by providing rich information on both the structural and functional aspects of the brain.

The overall objective of this study is to find out how the functional outcomes after stroke be related to the changes in functional networks responsible for motor execution (ME) and motor imagery (MI), and the changes in physical structure of the brain. This study is split into two parts to look into the features of brain reorganization and remodeling in patients after stroke from both functional and structural perspectives, and to correlate these features with their functional outcomes. Part I of this study is going to explore neural correlates of motor impairment during ME and MI from a functional perspective using functional magnetic resonance

imaging (fMRI), while part II of this study is going to describe remodeling of structural connectivity and its correlation with motor impairment from a structural perspective using diffusion tensor imaging (DTI).

In study I, ten chronic stroke patients with left subcortical ischemic lesions and right hemiparetic limbs, and ten unimpaired subjects were included. Their cortical processes were studied when they were asked to perform ME and MI unimanually using their unaffected and affected wrists during fMRI. Laterality index (LI) and overlap index (OI) were used to quantify hemispheric asymmetry and the spatial discrepancy of the cerebral activations, respectively. In study II, one of the stroke patients was excluded, since the anatomical image of this patient was contaminated by motion artifacts, which led to failure in subsequent data analysis. One of the unimpaired subjects with similar age was also excluded to match the total number of stroke patients included in this study. Therefore, only nine chronic stroke patients having left subcortical ischemic lesions and nine unimpaired subjects were included to study the post-stroke structural remodeling based on the data collected using DTI. Three connectivity measures (fractional anisotropy FA, connection weight CW, connection strength CS) were used to localize the fiber tracts and areas that were affected by the lesions.

From correlation results in study I, the supplementary motor area (SMA), its activation volume and congruence in functional neuroanatomy associated with ME and MI using affected wrist positively correlated with motor performance. During ME of affected wrist, the precuneus, its activation volume and congruence in functional neuroanatomy between patient and unimpaired groups showed negative correlation, while in non-primary motor areas, the hemispheric balance of premotor cortex and the congruence in functional neuroanatomy of contralesional inferior parietal lobule between patient and unimpaired groups showed positive correlation

with motor performance. From the results in study II, significant positive CS-motor correlations were found in five common regions (ipsilesional precentral gyrus, ipsilesional rostral middle frontal cortex, ipsilesional pallidum, ipsilesional amygdala and contralesional lingual gyrus) before and after controlling for the bilateral CSTs' connectivity property. Meanwhile, the CSs of three of these regions (precentral gyrus, rostral middle frontal cortex and amygdala) were also found significantly higher in unimpaired subjects compared with those in stroke patients, indicating that these three regions exhibited weaker connections to the rest of the network for the stroke patients.

From a functional perspective, the non-primary motor-related areas were revealed to play a critical role in determining motor outcomes after left subcortical stroke, which was demonstrated in our stroke patients. In particular, SMA might be the key neural substrate recruited during MI, which is in association with motor recovery. From a structural perspective, statistically significant alterations of neural connectivity in both the ipsilesional and contralesional hemispheres were shown in our stroke patients compared to the unimpaired subjects, implying structural remodeling occurred in widespread areas in both the ipsilesional and contralesional hemispheres.

Publications

Journal papers

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Conference papers

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Wong WW, Tong KY, Meng F, Tang KW, Fong WC, Gao XR, Gao SK, Chan ST. Motor networks in chronic stroke: Comparison of model-free and model-based analyses on functional MRI signals. 4th WACBE World Congress on Bioengineering, Hong Kong, China, 2009.

Wong WW, Tong KY, Meng F, Tang KW, Gao XR, Gao SK, Chan ST. Exploring effective connectivity during unilateral movement in stroke using structural equation modeling. ISMRM 17th Scientific Meeting, Hawaii, U.S.A., 2009.

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List of Abbreviations

ANOVA	Analysis Of Variance
BA	Brodmann Area
BOLD	Blood Oxygenation Level Dependent
CN-STN	Caudate Nucleus and Subthalamic Nucleus
CS	Connection Strength
CST	Corticospinal Tract
CW	Connection Weight
DALY	Disability-Adjusted Life-Year
DMN	Default Mode Network
DTI	Diffusion Tensor Imaging
DWI	Diffusion-Weighted Imaging
EEG	Electroencephalogram
EMG	Electromyography
FA	Fractional Anisotropy
FMA	Fugl-Meyer Assessment
FMA_SE	FMA Shoulder and Elbow Movements
FMA_WH	FMA Wrist and Hand Movements
fMRI	Functional Magnetic Resonance Imaging
FWHM	Full-Width Half-Maximum
ICC	Intra-Class Correlation Coefficient
IPL	Inferior Parietal Lobule
LI	Laterality Index
M1	Primary Motor Cortex
MANOVA	Multivariate Analysis Of Variance
MCA	Middle Cerebral Artery
ME	Motor Execution
MEG	Magnetoencephalography
ME_L	ME Using Left/Unaffected Wrist
ME_R	ME Using Right/Affected Wrist
MI	Motor Imagery
MI_L	MI Using Left/Unaffected Wrist
MI_R	MI Using Right/Affected Wrist

MRI	Magnetic Resonance Imaging
MSS	Motor Status Scale
MSS_EF	MSS Elbow and Forearm Movements
MSS_SM	MSS Shoulder Movements
OI	Overlap Index
PET	Positron Emission Tomography
PF	Prefrontal Cortex
PI	Perfusion Imaging
PLIC	Posterior Limb of the Internal Capsule
PM	Premotor Cortex
PMd	Dorsal PM
ROI	Region of Interest
S1	Primary Sensory Area
SMA	Supplementary Motor Area
TH-PU	Thalamus and Putamen
TMS	Transcranial Magnetic Stimulation
TTP	Time to Peak
WHO	World Health Organization
YLD	Years of Life Lived With Disability
YLL	Years of Life Lost

Chapter 1 Introduction

1.1 Study Background

1.1.1 Prevalence and Impact of Stroke

Stroke is a leading cause of long-term disability in adult (Paul *et al.*, 2005, Lloyd-Jones *et al.*, 2010). In every year, there are around 15 million people from all over the world suffering from a stroke (Mackay *et al.*, 2004, Lloyd-Jones *et al.*, 2010), in which one in six people have a chance of getting stroke at some time in their lives. According to the self-reported data from population-based surveys conducted in Hong Kong, an increasing trend in the stroke prevalence rates was seen among the elderly (aged 65 and above) between 1998 and 2008. The stroke prevalence rates increased with age for both males and females, and there was a tendency for males to have a higher stroke prevalence rate than females (Department of Health of Hong Kong Special Administrative Region and Department of Community Medicine of the University of Hong Kong, 2005). Besides, a large proportion of stroke sufferers also come from those living in institutions. Based on self-reported estimates, the stroke prevalence rate among the elderly (aged 65 and above) living in institutions was 30.9% in 2008 (Census and Statistics Department of Hong Kong Special Administrative Region, 2009). Looking at the current and future estimates of stroke prevalence, the number of community-living stroke sufferers aged 65 and above could be projected to more than double between 2010 and 2036, increasing from 0.04 million to 0.11 million. The number of institutional stroke sufferers aged 65 and above could also be projected to increase from 0.02 million in 2010 to 0.05 million in 2036 (Yu *et al.*, 2012). Most stroke survivors are however

commonly left with loss or impairment in one or more of their body functions such as movement, sensation, language, thinking, memory and emotion (Carey, 2012). Currently, one-third of these stroke sufferers exhibit persisting and significant long-term disability (Barnes *et al.*, 2005, Roger *et al.*, 2011), and further 20% of them need assistance for their activities of daily living (Bonita *et al.*, 1997). This proportion of stroke sufferers with disability is still kept increasing (Carandang *et al.*, 2006).

Moreover, stroke is a global healthcare problem, and has a continuous impact on quality of life (Langhorne *et al.*, 2011). According to the World Health Organization (WHO) report on the global burden of disease, stroke is the second leading cause of death and remains among the top six causes of burden of disease in the world. There is an estimate of 55%-75% of the stroke survivors remained presenting functional impairments and reduced quality of life after months or even years of the infarct (Levin *et al.*, 2009). These continuing disabilities would in turn significantly decrease their life satisfaction (Ostwald *et al.*, 2009), and therefore the importance of good recovery has been highlighted (Carod-Artal and Egido, 2009).

In view of the importance of recovery, stroke becomes one of the largest impairment categories for rehabilitation (Carey, 2012). In Australia, for instance, stroke is the third largest impairment category, accounting for one in ten rehabilitation cases. The cost has a considerable influence on the healthcare settings, creating a heavy burden of care (Department of Health of the United Kingdom, 2005). In Hong Kong, over HK$ 1,330 million was spent on various stroke services, including hospitalization, out-patient care, rehabilitation service and community allied health service, of which the largest portion of money was spent on hospitalization, occupying over 80% of the direct medical cost (Yu *et al.*, 2012). By 2036, the cost is anticipated to increase to approximate HK$ 3,980 million per year.

At the same time, the cost of institutional care was also found to be significant and was anticipated to increase by 1.8 times to HK$ 4,530 million per year, in parallel with the increase in the prevalence of stroke sufferers living in institutions (Yu *et al.*, 2012).

Although the need of rehabilitation is demanding, the outcomes of rehabilitation are often limited, depended on compensation and interventions that are not always evidence-based (Carey, 2012). Currently, given more evidences stating the potential for neural plastic changes, it is suggested that the time window for restoring capacities and skills through learning-based approaches is open, giving ongoing hope for those stroke survivors (Carey and Seitz, 2007, Richards *et al.*, 2008, Carey *et al.*, 2011). This time window can be longer than the suggested "days to weeks for restoring impairments" and "days to months for task-oriented practice with adaptive learning and compensation strategies" (Langhorne *et al.*, 2011).

1.1.2 Definitions of Recovery and Rehabilitation

Recovery has been used to describe both the restoration of injured structures and behavioral functions, as well as to characterize the clinical improvements (Levin *et al.*, 2009). Terms such as recovery and compensation are frequently used, and each term will be interpreted differently for different level of outcome being investigated (Carey, 2012). It is therefore suggested that they should be interpreted and qualified relative to these levels. In parallel with the International Classification of Functioning, recovery versus compensation has been recommended to be qualified according to the levels of health condition (neuronal), body function/structure (impairment), and activity (disability) (Levin *et al.*, 2009). Besides, other terms are also used, including restitution or restoration (as a process to restore the lost or impaired functionality of damaged neural tissue), substitution (as a process to

reorganize the spare neural pathways for relearning lost functions), and compensation (as a process to minimize the disparity between the impaired skills of the patients and the actual needs of their living environment) (Finger and Stein, 1982, Kwakkel *et al.,* 2004). It is recommended that recovery should be measured across a profile of results from brain reorganization to performance outcomes (Carey, 2012).

As for rehabilitation, the World Health Organization defines it as a process targeted at allowing people with disabilities to achieve and retain their optimal conditions from the physical, sensory, intellectual, psychological and social functional perspectives. Meanwhile, rehabilitation assists them to attain independence and self-determination by providing them appropriate tools. Therefore, the ultimate goal of rehabilitation is "to improve function and/or prevent deterioration of function, and to bring about the highest possible level of independence – physically, psychologically, socially, and financially – within the limits of the persisting stroke impairment" (Dewey *et al.,* 2007). From a neuropsychological perspective, it is suggested that methods such as retraining existing neural pathways or training new neural pathways to regain or improve the lost or diminished neurocognitive functioning, are typically involved during the rehabilitation of sensory and cognitive function. A related term, "neurorehabilitation", is now emerged and is defined as a clinical subspecialty of neuroscience that is dedicated to the restoration and maximization of the lost functions due to impairments caused by damage to the nervous system (Selzer, 2006).

1.1.3 Neuroplasticity as a Basis for Stroke Rehabilitation

Neuroplasticity can refer to as the capacity of the nervous system to react to inherent or extraneous stimuli through structural, functional and connectional reorganization (Cramer *et al.,* 2011). Further, it can occur at many levels ranging

from cellular to behavioral levels; and “can happen during development, in response to the environment, in support of learning, in response to disease, or in relation to therapy” (Cramer *et al.*, 2011). The extent of the brain self-remodeling come from experience forms the foundation of the brain’s capacity to preserve memories, enhance functions, and perform daily tasks (Bruel-Jungerman *et al.*, 2007). Continuous functional reorganization in the brain that represents a new knowledge acquired through experiences is needed to learn or memorize the new fact or skill. This lifelong ability of the brain to adapt based on new experiences and learning is known as neuroplasticity (Carey, 2012).

There is growing evidence from human and animal studies showing that neural plastic changes are associated with stroke recovery (Selzer, 2006). Experience has been highlighted in animal studies as an important element in these neural plastic changes (Nudo *et al.*, 1996). The nature of the experience and whether it is focused on repeated performance versus skill acquisition was further explored in a series of studies conducted by Nudo and his colleagues (Plautz *et al.*, 2000). In their studies, two behavioral tasks were used to distinguish between the mapping of brain regions associated with motor recovery during the repetition of motor activity and the brain mapping during the acquisition of motor skills. Plastic changes in movement representations in the brain were not induced following repetitive motor activity on the easier task. Rather, skill acquisition on the difficult task could result in neural plastic changes, involving selective expansions of movement representations in the brain. Further improvements in performance and further changes in brain mappings corresponding to the movement were invoked by extensive additional training on the more difficult skill-based task. Based on these findings, neural plasticity can be driven by learning-based behavioral changes through training. In other words, rehabilitation may be viewed as a trigger to facilitate plasticity, and as a means by

which neural plastic changes may be shaped to achieve meaningful outcomes for the individual (Carey, 2012). The mechanism of this shaping of neural plastic changes triggered by rehabilitation is still being explored.

Furthermore, more evidences from human studies show that specific interventions promote neural plasticity and improved motor recovery after stroke (Hodics *et al.*, 2006, Carey and Seitz, 2007, Richards *et al.*, 2008, Stinear *et al.*, 2008). A number of promising interventions have been developed to facilitate the neural plastic changes (Carey, 2012). Some of these approaches can be supported and explained partially by preliminary evidence of changes in the brain (Carey, 2012). For example, motor imagery (MI) training was developed following the evidence of showing it as an effective way of stimulating the brain regions normally engaged in planning and controlling movements of the paralyzed limb (Weiss *et al.*, 1994). The stroke patients who participated in the mental practice protocol were observed to exhibit an increased use of their affected limb, as well as demonstrate an improved quality of movement and higher motor assessment scores obtained after the intervention (Page *et al.*, 2005).

Numerous factors, however, are identified to have an impact on the potential for neural plastic changes and the capacity to get benefits from rehabilitation after stroke (Kolb *et al.*, 2010), such as the stroke nature and severity, post-stroke duration, psychological issues, attention and learning ability, and capacity of viable brain networks for plasticity (Carey, 2012). Other factors such as structural (Riley *et al.*, 2011) and functional (Stinear *et al.*, 2007) integrity of white matter tracts in motor recovery and integrity of functionally connected networks in attention recovery (He *et al.*, 2007) have also shown to be critical in promoting long-term recovery from stroke, implying a concept that neural plastic changes rely on the integrity of these

pathways and networks during the process of recovery and rehabilitation (Carey, 2012).

1.1.4 Insights from Neuroimaging into Stroke Rehabilitation

Experience can change both physical structure (anatomy) and functional organization (physiology) of the brain (Carey, 2012). There are a number of modalities and techniques that have been used to study mechanisms of post-stroke recovery (Carey, 2012). One of the modalities is magnetic resonance imaging (MRI). MRI can provide information on morphological aspects (e.g. brain structure and volume), anatomical aspects (e.g. white matter integrity and fiber tract connections), and functional aspects (e.g. functional brain activation and functional network connectivity) of the brain. Therefore, it has potential to give important insights into the mechanisms of post-stroke recovery (Carey, 2012). For instance, functional brain imaging techniques allow us to visualize patterns of the brain activity during motion, perception, cognition, and emotion (Carey and Seitz, 2007). Mappings of brain activation can be used to identify involved brain regions and networks, to see how they differ between stroke patients and healthy controls, and how they might change over time during the process of recovery or rehabilitation (Carey, 2012). Different patterns of neural plastic change have been reported following stroke recovery (Carey and Seitz, 2007, Johansen-Berg, 2007, Cramer *et al.*, 2011). As an example, change in location of movement representation in the ipsilesional hemisphere, shift in the balance of activity across hemispheres (Tecchio *et al.*, 2007, Grefkes *et al.*, 2008), and involvement of remote locations (Seitz *et al.*, 1999), contralesional regions (Schaechter and Perdue, 2008) and distributed networks (Seitz *et al.*, 1998, Sharma *et al.*, 2009) that would not normally be recruited, were observed from previous studies. Besides, changes in brain structure, such as an increase of cortical

thickness in the neighborhood of activation (Schaechter *et al.*, 2006), and changes in cortical thickness in the areas that show increased activation and clinical improvement have been reported (Schaechter *et al.*, 2006, Gauthier *et al.*, 2008), providing an evidence of outcome-related structural change in the stroke patients.

Moreover, the role of intrahemispheric and interhemispheric changes in recovery of motor and language functions has been investigated (Carter *et al.*, 2010), revealing the importance of interhemispheric adaptations. From these discoveries, the importance of brain networks rather than localized regions has been highlighted in stroke recovery (Carey, 2012). Understanding of brain networks and how they are interrupted following stroke will provide us more information on the behavioral manifestations of impairment and the manipulation of neural plasticity in therapy (Carey, 2012).

1.2 Study Significance and Objectives

In view of the great impact of stroke (ranging from individuals to the society), the need and importance of good recovery has been highlighted. Currently, based on the evidences stating the potential for neuroplasticity and its relation to stroke recovery, a restorative and neuroscience-based approach to stroke rehabilitation, such as motor imagery training, has been developed and recommended to the rehabilitation specialists. However, the mechanism of the shaping of neuroplasticity in relation to stroke recovery is still not fully understood. In addition, a number of factors, such as the extent of damage in influencing the structural and functional brain networks, have been identified to have an impact on the potential for neuroplasticity and the capacity for receiving benefits from rehabilitation. Thus, more understanding of the neural mechanism that underpins an effective and restorative approach to stroke rehabilitation becomes necessary and important. The emerging of neuroimaging

techniques can therefore help to study this mechanism of post-stroke recovery. MRI, which is one of these techniques, has been shown having a great potential to give important insights into post-stroke recovery mechanisms by providing rich information on both the structural and functional aspects of the brain, and is therefore adopted in this study to help investigate the brain reorganization after stroke.

In this study, two imaging modalities of MRI were used: functional MRI (fMRI) and diffusion tensor imaging (DTI). Functional MRI is a functional neuroimaging technique that measures brain activity by detecting associated changes in blood oxygenation and flow that happen in response to neural activity. This technique utilizes the coupling relationship between cerebral blood flow and neuronal activation. Increase in blood flow to a region is observed when that region is more active. Activation maps can be generated using fMRI to show the involved active brain regions during a particular mental process. DTI is an imaging technique based on the anisotropic nature of water movement along the white matter fibers. The movement of water molecules is relatively free along the fibers, but is more restricted in the direction perpendicular to the fibers orientation. This phenomenon thus enables DTI to extract the fibers' orientation on a pixel by pixel basis and to quantify the motional anisotropy using quantitative measurements. Here, a number of quantitative measures, including cluster volume, laterality index and overlap index derived from fMRI data, and fractional anisotropy, connection weight and connection strength derived from DTI data, were used to quantify the activation features and connectivity features respectively.

The overall objective of this study is to find out how the functional outcomes after stroke be related to the changes in functional networks responsible for motor execution and motor imagery, and the changes in physical structure of the brain. This study is split into two parts to look into the features of brain reorganization and

remodeling in patients after stroke from both functional and structural perspectives, and to correlate these features with their functional outcomes. Part I of this study is going to explore neural correlates of motor impairment during motor imagery and motor execution using fMRI, while part II of this study is going to describe remodeling of structural connectivity and its correlation with motor impairment using DTI.

Specific objectives of part I of this study include:

1. To find out the clinical relevancy of cluster volume of activation in stroke during motor execution and motor imagery using affected limb
2. To find out the clinical relevancy of lateralization of activation in stroke
3. To find out the clinical relevancy of congruence in functional neuroanatomy between motor execution and motor imagery tasks, and between stroke and unimpaired groups

Specific objectives of part II of this study include:

1. To look at the correlation patterns of structural connectivity properties with motor impairments for the stroke patients
2. To look at the relationships between the structural connectivity properties and the motor impairments after controlling for the CST's structural properties
3. To look at the differences between unimpaired subjects and stroke patients in connectivity features for those regions with significant correlations

Chapter 2 Literature Review

2.1 Stroke Epidemiology

2.1.1 Stroke Incidence and Death

Globally, there was an estimate of 16 million people suffering from a first-ever stroke in 2005, with an estimated 62 million people survived after stroke (Strong *et al.*, 2007). If without any interventions provided to these people, this number is expected to increase to an estimated 23 million people with a first-ever stroke, in association with 7.8 million deaths, by 2030 (Strong *et al.*, 2007). Currently, stroke becomes the second leading cause of mortality worldwide, accounting for 5.5 million deaths annually (equivalent to around 9.7% of all deaths worldwide) (Strong *et al.*, 2007).

In developed countries, the stroke incidence remains relatively high. For instance, in the United States, 0.8 million individuals suffer a stroke annually, with 75% of them exhibiting first-time attacks (Lloyd-Jones *et al.*, 2010), in which around 269 in 100,000 people would have a stroke in the United States (Williams, 2001). This rate mirrors the rate in Europe, where the range of annual stroke incidence is between 94.6 per 100,000 people for women and 141.3 per 100,000 people for men (Heuschmann *et al.*, 2009).

In China, compared with the white populations, the mean age of stroke onset was even younger in Chinese populations (age range: 66-70 years) than the white populations (age range: 72-76 years) (Tsai *et al.*, 2013). The age-standardized annual first-ever stroke incidence was also found higher among the Chinese than white populations for the age category of 45-74 years (incidence range 205-584 vs.

170-335 per 100,000 for Chinese and white populations respectively), based on the findings from community-based studies (Tsai *et al.*, 2013).

As for Hong Kong, from the hospital admission data, the annual age-adjusted incidence rates of first-ever stroke among the elderly aged 65 and above decreased slightly from 1410 per 100,000 people in 2000-2001 to 1050 per 100,000 people in 2006-2007 (Yu *et al.*, 2012). Nevertheless, a higher stroke incidence rate was found to be associated with increasing age, which can be reflected in the reported stroke incidence rates shown in Hong Kong, as well as in other countries, including the developed (e.g. the United States, the United Kingdom) and developing (e.g. China) countries (Yu *et al.*, 2012). Moreover, stroke is the fourth major cause of death in Hong Kong, in association with around 3,400 deaths as a result of stroke in 2009, corresponding to 8.4% of total deaths (Department of Health of Hong Kong Special Administrative Region, 2011). Particularly, there was a gradual increase in the number of deaths from stroke observed among the elderly aged 65 and above between 2001 and 2009 (Department of Health of Hong Kong Special Administrative Region, 2011), probably owing to the ageing population.

2.1.2 Stroke-related Disability

In order to better comprehend the long-term impact of the chronic and morbid conditions upon populations, disability-adjusted life-year (DALY), which is a measure used for reflecting years of healthy life lost due to living with disability and years of life lost due to premature mortality, has been developed (Murray *et al.*, 1996). The DALY, which integrates the measures of years of life lost (YLL) attributable to premature death and years of life lived with disability (YLD), has been widely used in epidemiological analyses for assessing the overall disease burden. Lost DALY is regarded as a valuable index of relative functionality for the

post-stroke survivors. Currently, stroke is associated with 43.7 million lost DALYs worldwide in each year, accounting for about 3.2% of all worldwide lost DALYs. Therefore, stroke ranks as the seventh-major cause of lost DALYs globally among people of all ages (Strong *et al.*, 2007).

By studying the global stroke burden, remarkable patterns have come up across various countries and regions. During the past decade, chronic diseases, including stroke, have accounted for around 85% of entire disease burden in high-income countries. Therefore, there is a great impact of stroke upon lost DALYs to those high-income countries, in which stroke is associated with up to 4.8% of all lost DALYs (Lopez and Mathers, 2006). Furthermore, there is up to tenfold difference in the lost DALYs between those countries that are most and least influenced by stroke. For example, some European countries (e.g. Switzerland and France) only had around 200 per 100,000 lost DALYs in 2002, in contrast with approximately 2000 per 100,000 lost DALYs found in some Asian countries (e.g. Mongolia and Kyrgyzstan) in the same year (Figure 2.1) (Johnston *et al.*, 2009).

In Hong Kong, almost 0.12 million lost DALYs were found as a result of stroke among older people in 2006. The majority of the burden was owing to disability, of which around 90% of DALYs was attributed to the 0.106 million YLDs. The remaining 10% of the DALYs was attributed to the 0.013 million YLLs from stroke (Yu *et al.*, 2012).

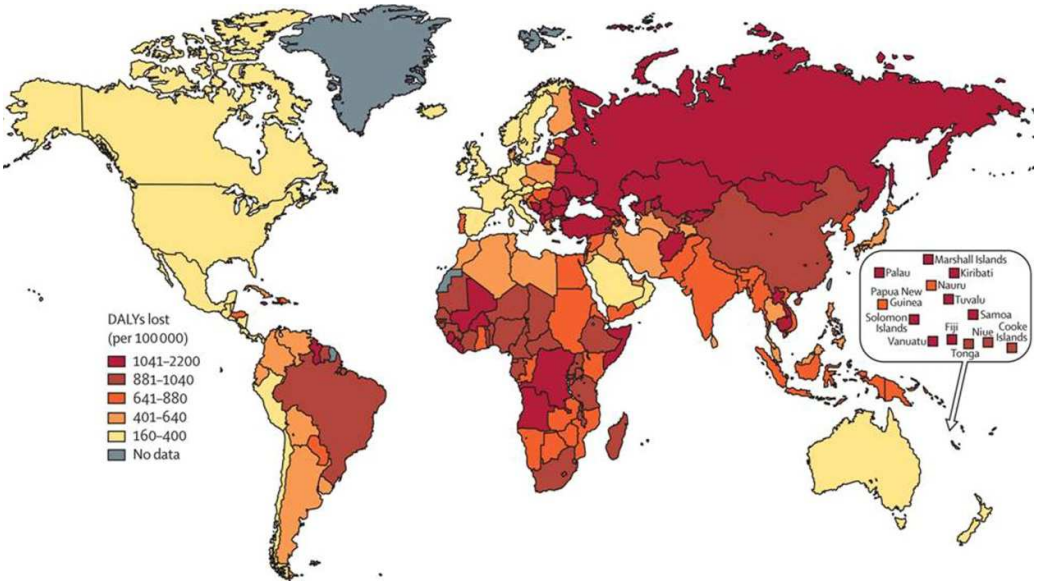


Figure 2.1 Rates of age-adjusted and gender-adjusted lost DALY due to stroke. The highest rates are found in North Asia, Eastern Europe, Central Africa, and the South Pacific (Johnston *et al.*, 2009).

2.2 Stroke Pathophysiology and Recovery

2.2.1 Overview of Ischemic Stroke Pathophysiology

The pathophysiological mechanism of stroke is complicated, involving a number of processes, such as glutamate excitotoxicity, oxidative stress, lipid peroxidation, inflammation, blood brain barrier dysfunction, and leukocyte infiltration (Figure 2.2). These processes are revealed to be involved and closely linked to the incidents in the pathophysiology of ischemic stroke. These events are found interrelated and coordinated, which can result in ischemic necrosis. Necrosis is morphological phenomenon with characteristics of initial presentation of cellular and organelle swelling, subsequent rupture of plasma, nuclear, and organelle membranes, and later disruption of nuclear structure and cytoplasmic organelles with extrusion of cell contents into the extracellular space (Majno and Joris, 1995, Broughton *et al.*, 2009). Following a short period of a cerebral ischemia, the core of brain tissue, which is exposed to the most severe reduction in blood flow, will be severely damaged, and

will undergo necrotic cell death subsequently. An area of less severely affected tissue surrounding this necrotic core is rendered functionally silent by the reduction in blood flow but still keeps metabolically active (Majno and Joris, 1995, Broughton *et al.*, 2009). The region enclosing the infarct core is known as the ischemic penumbra. During the initial phases of ischemia, this penumbra occupies almost half of the total lesion volume. It is characterized as a region where there is room for salvage through post-stroke therapy (Ginsberg, 1997). Relatively mild ischemia, which appears in the penumbra area of a focal ischemic infarct, develops relatively slow, and it relies on specific genes activation and may ultimately lead to apoptosis (Dirnagl *et al.*, 1999, Lipton, 1999, Zheng and Yenari, 2004). Recent studies have found that the neurons in the ischemic penumbra, or in the perilesional area, may experience apoptosis following several hours or even days, and therefore there is room for the neurons in the ischemic penumbra to recover within a period after the stroke onset. Compared with necrosis, apoptosis seems a more sequentially process of energy-dependent programmed cell death for disposal of redundant cells. Cells experiencing apoptosis are disassembled from within in a managed way that reduces the damage to neighboring cells (Broughton *et al.*, 2009). Intrinsic and extrinsic pathways are suggested as the two general paths for the trigger of apoptosis. More information has been provided from recent experimental studies to further characterize the apoptotic processes happening after ischemic stroke.

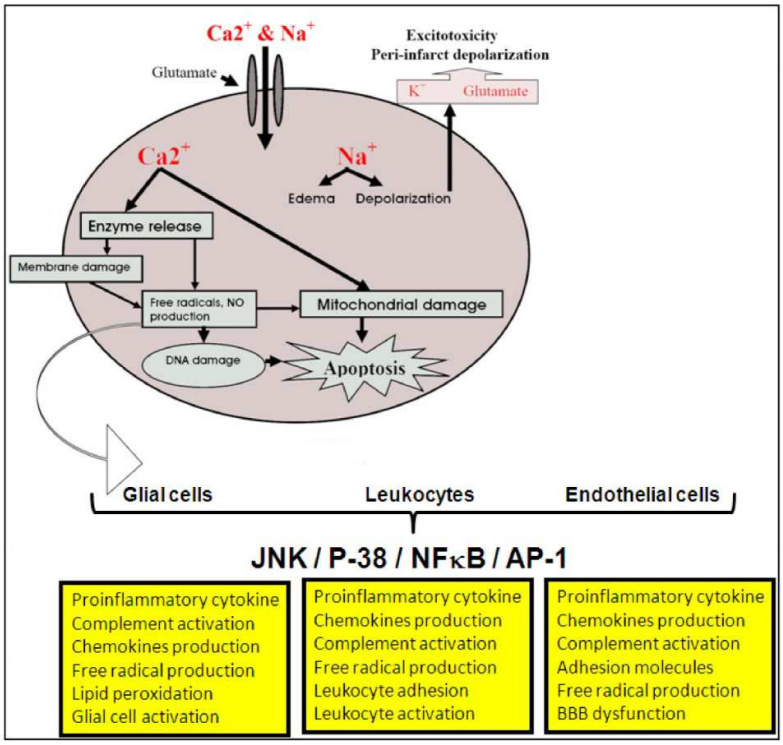


Figure 2.2 Major cellular pathophysiological mechanisms following ischemic stroke. Energy failure induced by brain ischemia results in neuronal depolarization. Intracellular Ca2+ and Na+ are dramatically increased by the activation of specific glutamate receptors, while K+ is discharged into the extracellular space. Edema occurs when water flows into the intracellular space. The increase in the intracellular messenger Ca2+ activates proteases, lipases and endonucleases. Free radicals are then produced, leading to the damage to membranes, mitochondria and DNA, and subsequently driving cell death, and resulting in the formation of inflammatory mediators. This process triggers activation of JNK, p-38, NFκB and AP-1 in glial cells, endothelial cells, and infiltrating leukocytes. This culminates in pro-inflammatory cytokine and chemokine secretion and ultimately results in the irruption of leukocytes through up-regulation of endothelial adhesion molecules (Woodruff *et al.*, 2011).

2.2.2 Overview of Recovery after Ischemic Stroke

Recovery from stroke commences early following the ischemia. Recanalization is one of the important contributors of this spontaneous recovery, which is usually observed together with rapid cerebral reperfusion. It is a determinant of the extent of salvageable brain tissue and the extent of ischemic brain lesion that is under the threat of ischemia (Figure 2.3). Therefore, the neurological deficits often regress significantly and shortly after ischemic stroke, but progress subsequently at a slower

pace for a period up to two years. Beyond the acute time window of around 24 hours, stroke recovery invokes the processes of neural repair and functional reorganization. For instance, secondary changes such as vasogenic edema and inflammatory infiltration retrogress instinctively within up to 2 weeks following stroke (Saleh *et al.*, 2004, Saleh *et al.*, 2007). Although there is a huge variance of instinctive post-stroke recovery over the first three months (Cramer, 2008), the long-term neurological outcome can possibly be predicted by the neurological status as early as at day 4 after stroke onset (Kwakkel *et al.*, 2003, Sprigg *et al.*, 2007). The progressive improvement of performing activities of daily living usually occurs within 26 weeks after the stroke insult (Schepers *et al.*, 2008, Welmer *et al.*, 2008). Since damage to both gray and white matter of the brain is usually regarded as a common consequence following the brain infarcts, tissue repair becomes a critical step which attempts to replace the ischemic tissue debris with non-damaged yet functional brain tissue for each of these compartments. It is suggested that this substitution may come from endogenous sources, given the findings from studies on animal models of stroke. Work in animal studies suggests that proliferation of stem cells in the subventricular germinal area as well as neural progenitor cells is observed in response to focal ischemia (Kokaia *et al.*, 2006, Ohab *et al.*, 2006). They seem to be controlled by numerous factors such as neurotrophic substances and inflammatory mediators (Schabitz *et al.*, 2007, Pluchino *et al.*, 2008). However, it is likely that there is only limited capacity for the regeneration of neuron and lengthy extension of axon within the brain. It seems that the growth of neurons and axons would stop at locations of scars within lesions at central nervous system (Hermanns *et al.*, 2001). In spite of this, there is still promising evidence from animal studies that the nerve fibers may grow with considerable distances from non-damaged nerve cells in the perilesional cortex and may become new synapses at their destination (Biernaskie

and Corbett, 2001, Frost *et al.*, 2003, Dancause *et al.*, 2005). These reorganizational changes might be associated with clinical recovery, which can be improved by dedicated rehabilitative training (Nudo *et al.*, 1996). However, such reorganizational processes are slow and may take months to complete (Figure 2.3).

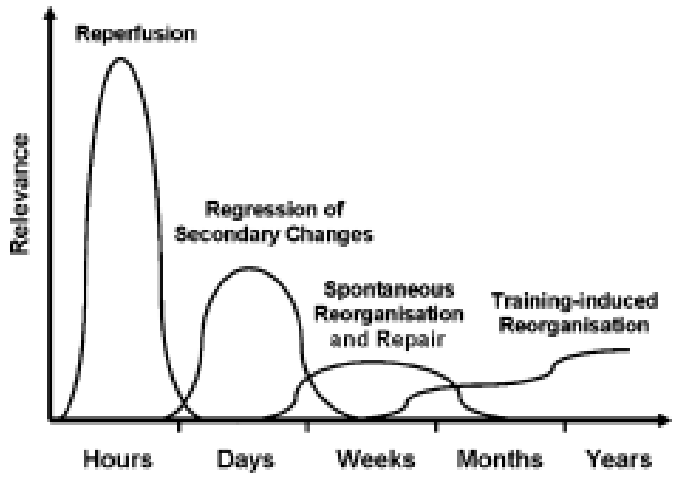


Figure 2.3 A series of events occuring during stroke recovery (Seitz and Donnan, 2010).

2.3 Stroke Neuroimaging

Taking advantages of modern neuroimaging techniques, the human brain structural and disease-related changes can be visualized in vivo and noninvasively. Particularly, magnetic resonance imaging (MRI) is a noninvasive imaging technique that can capture high resolution images of the human brain, and therefore it is widely used as a tool for detecting or investigating the neurological changes induced by stroke. Some specific imaging sequences of MRI, such as perfusion imaging (PI) and diffusion-weighted imaging (DWI), are now commonly used to detect early damages or adaptations in acute stroke. For example, tiny ischemic lesions that are clinically inactive can be detected by MRI, and this detection may act as a predictor of stroke recurrence (Kang *et al.*, 2006). The stroke lesion patterns as revealed by MRI can further be supplemented by the data from diffusion tensor imaging (DTI) which is

able to detect alterations in fiber tracts of central nervous system. Moreover, functional MRI (fMRI), which can detect the physiological responses of the human brain, is also frequently used to study brain activities in response to specific paradigm design. This may allow an investigation of brain adaptation to the existence of a lesion to be made, together with deficit compensation, and in response to relearning.

2.3.1 Functional Imaging in Stroke Recovery

A normal activation pattern can usually be restored progressively in patients during their recovery processes (Marshall *et al.*, 2000, Nhan *et al.*, 2004). Nevertheless, a bilateral activation pattern is still preserved in patients even they have an excellent recovery (Foltys *et al.*, 2003, Butefisch *et al.*, 2005). It is now well established that there are large-scale changes, as revealed by fMRI, which can influence the contralesional cortical and subcortical structures in patients following a focal brain infarct. Additionally, these changes are found to be correlated with the infarct lesion volume. In stroke patients with poorer motor recovery of their affected limbs, as observed from an abnormal electromyographic (EMG) muscle activity, the bilateral activation pattern was revealed and suggested in association with the mirror movements of the unimpaired hand which possibly recruited the undamaged motor cortex in the contralesional hemisphere (Figure 2.4). However, contralesional activations which were not normally recruited in healthy controls were also shown in patients with excellent recovery. These additional activities, which involved activations in premotor cortical areas, are largely evocative of relearning, since the activation patterns they represented are similar to those recruited during procedural relearning and are transient in nature (Butefisch *et al.*, 2005, Saur *et al.*, 2006, Marshall *et al.*, 2009). Furthermore, tiny activations in the area of the motor cortex

(Figure 2.4) were proposed to reflect an active inhibition of the contralesional motor cortex which is more likely to get excited than normally (Butefisch *et al.,* 2005). Indeed, the mirror movements which are frequently observed initially after stroke might result from this enhanced cortical excitability of the contralesional cortex (Nelles *et al.,* 1998). Notably, the damaged functional system can be invoked by specific activation, such as during finger movements in hemiparesis (Nair *et al.*, 2007, Hummel *et al.,* 2009). Other than the atypical local activation patterns, abnormalities in the intra- and inter-hemispheric coupling between cortical areas are also revealed by network analysis of fMRI data.

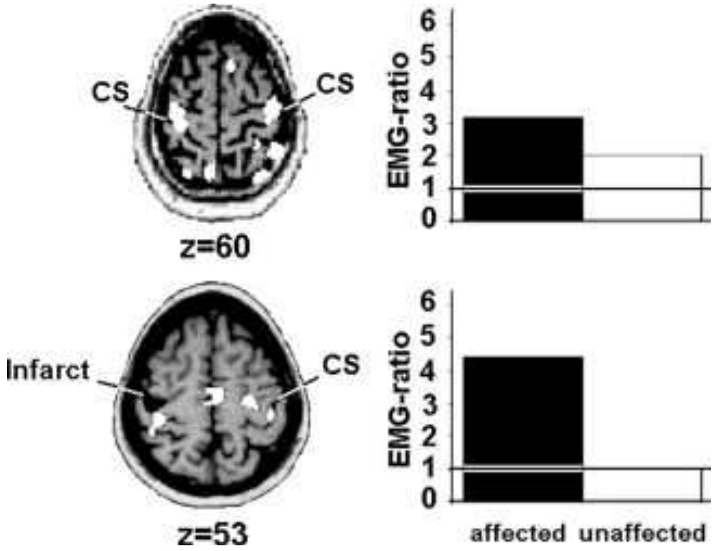


Figure 2.4 Activation pattern during sequential finger movements of the impaired hand in patients who had full clinical recovery following first hemiparetic stroke. Bilateral activation in brain areas was shown in a patient with associated finger movements as observed by enhanced electromyographic (EMG) activity in the unimpaired hand (upper panel of the figure). Another patient with no increased EMG-activity in the unimpaired hand (EMG ratio = 1) also exhibited increased activity in the contralesional motor and premotor cortex (lower panel of the figure). The axial images shown in the figure were obtained at 60 and 53 mm dorsal to the intercommissural line of the brain atlas in stereotactic space (Butefisch *et al.,* 2005).

Network analysis methods have shown that atypical inter-hemispheric interactions between the ipsilesional and contralesional motor cortex as well as between the

ipsilesional SMA and contralesional motor cortex were found in patients with subcortical stroke (Grefkes *et al.*, 2008). During unimanual movements of the affected hand, an inhibitory effect from the contralesional to the ipsilesional motor cortex was shown, and this inhibitory effect was found to be correlated with the motor impairment level. During bimanual movements, a reduction in the interaction between the ipsilesional SMA and the contralesional motor cortex was shown, and this again correlated with impairment level of the bimanual motor performance. This can be explained from the phenomenon that activation in the contralesional motor cortex became less important when the motor task did not rely much on working memory (Kimberley *et al.*, 2008). Besides, the bilateral premotor cortex also became less active when there was no demand of working memory.

2.3.2 Structural Imaging in Stroke Recovery

The involvement of damage to the cerebral cortex is usually highlighted with the occurrence of hemispheric brain infarcts, while the involvement of the white matter damage has been relatively less noted or appreciated. Given the findings from stereotactic lesion mapping of stroke lesions in the middle cerebral artery (MCA) territory, apart from discovering a prominent lesion overlap in the peri-insular cortex and basal ganglia, an obvious lesion overlap is also revealed in the white matter (Figure 2.5). The damage to the white matter is especially remarkable in leading to hemispatial neglect, apraxia, and severe hemiparesis (Karnath *et al.*, 2004, Stoeckel *et al.*, 2007, Pazzaglia *et al.*, 2008, Karnath *et al.*, 2009).

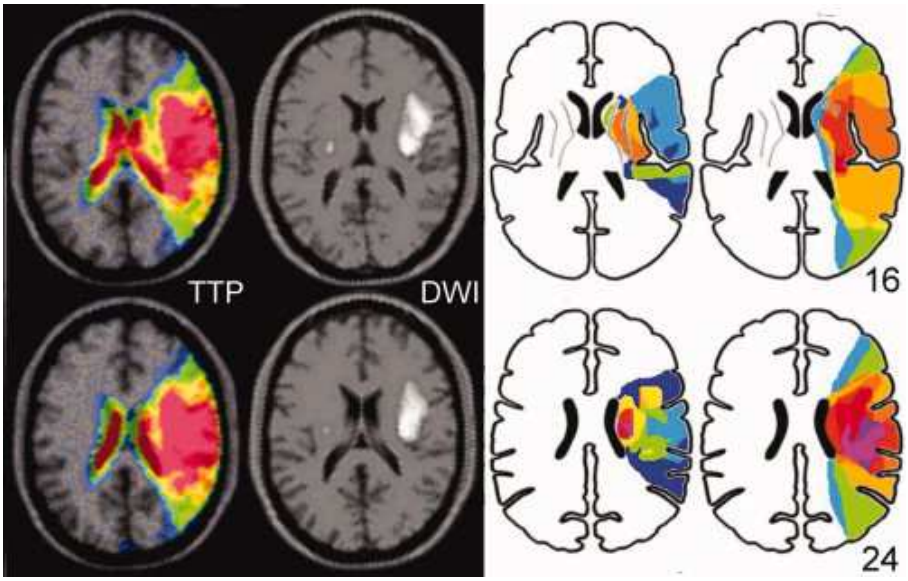
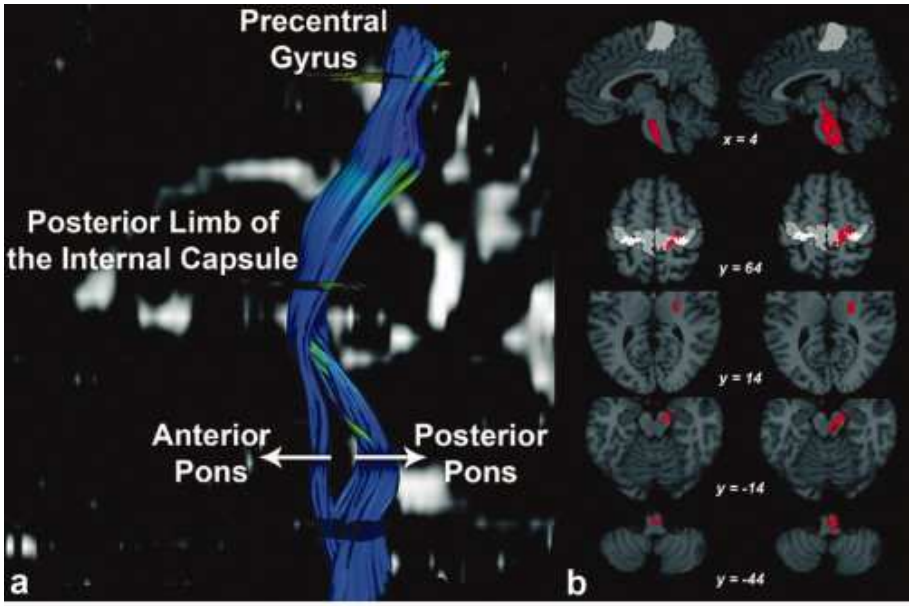


Figure 2.5 Topography of stroke infarct areas in the middle cerebral artery (MCA) territory. The cerebral perfusion disturbance was illustrated as an averaged time to peak (TTP) map of 64 acute stroke patients with lesions in the MCA territory within 3 hours after stroke onset (green represents an averaged TTP-delay >2 seconds, yellow >4 seconds, red >6 seconds) (first column from the left). The affected areas involved both gray and white matter, covering the insular cortex, the basal ganglia, and the perilesional areas. The averaged DWI lesion map showed mainly the involvement in the insular cortex and adjacent white matter (second column) (Stoeckel *et al.*, 2007). Among the patients with rapid recanalization of the MCA after systemic thrombolysis, the resulting infarct lesions had a relatively small overlap area, locating in the periventricular white matter (third column), while the resulting infarct lesions of the patients with poor recanalization had a large overlap area, encompassing the insular cortex, the basal ganglia, and the periventricular white matter (forth column). For these maps, increasing lesion overlap among patients was coded from cold to warm colors. The axial images shown in the figure were obtained at 16 and 24 mm dorsal to the intercommissural line of the brain atlas in stereotactic space (Seitz *et al.*, 2009).

On the other hand, the corticospinal tract (CST) has been suggested to play a critical role in determining the motor recovery. For example, the motor recovery is found to have a close relationship with the integrity of the CST as assessed by DTI (Kwon *et al.*, 2007, Stinear *et al.*, 2007, Vitali *et al.*, 2007, Hamzei *et al.*, 2008, Kim *et al.*, 2008, Schiemanck *et al.*, 2008, Schaechter *et al.*, 2009), as well as the overall atrophy of the cerebral peduncles is also found correlated with the CST integrity. From their findings, the asymmetry of the fractional anisotropy (FA) of the posterior limb of the internal capsule (PLIC) across the ipsilesional and contralesional

hemispheres is revealed to be correlated with the motor recovery, such that the patients with poorer recovery exhibited a greater FA asymmetry found in the PLIC. Besides, the movement-related motor cortex activation can also be influenced by the CST integrity as well (Stinear *et al.*, 2007, Hamzei *et al.*, 2008), such that fMRI activations in the bilateral hemispheres during finger movements were observed in patients with poorer recovery, while a lateralized fMRI activation to the ipsilesional hemisphere was shown in those patients with better recovery who exhibited a lower FA asymmetry in the PLIC. Furthermore, both the ventral and dorsal portion of the projection of the pyramidal tract in the pons are found severely affected in poorly recovered patients, while a good recovery is characterized by the continuity of the projections in the dorsal portion of the tract (Figure 2.6). Not only the ipsilesional motor fiber tract has the effect on the recovery potential, recent study demonstrated that the level of post-stroke motor skill recovery was associated with the microstructural status of both the ipsilesional and contralesional CSTs, since the stroke patients with poorer motor skill exhibited reduced FA in the bilateral CSTs, while the stroke patients with better motor skill showed elevated FA in the bilateral CSTs (Schaechter *et al.*, 2009).



Motor Recovery

Damage of Pyramidal Tract

	Ventral	Dorsal
Good	-	-
Moderate	+	-
Poor	+	+

c

Figure 2.6 (a) Pyramidal tract passing through posterior limb of the internal capsule and splitting up into ventral and supplementary dorsal portions at the pons. (b) The pyramidal tract was hightlighted in red, which originated from the anterior (gray) and posterior (white) portions of the Brodmann area 4 in the precentral gyrus. (c) Relationship between motor recovery and damage of the ventral and posterior portions of the pyramidal tract at the pons (Lindenberg *et al.,* 2010).

In addition, changes are not only revealed in the efferent motor fiber tracts, but also found in the cortico-cortical and cortico-subcortical fiber tract systems. Indeed, enhancement of the post-stroke motor cortical connectivity was revealed by DTI (Pannek *et al.*, 2009), and the functional outcome is found to be correlated with the degree of the orientation uncertainty and the white matter complexity (Zhang *et al.*, 2008, Pannek *et al.*, 2009). It is further suggested that the repair processes can be

driven by functional demands. From these findings, they agree with the evidence from functional neuroimaging suggesting that the coordination in bilateral cerebral hemispheres and in widespread regions is required for good recovery.

2.4 Stroke Neurorehabilitation

There are a number of reports describing different rehabilitative strategies which attempt to improve the neurological deficit following stroke (Cramer, 2008). A widespread involvement of motor representations in the ipsilesional hemisphere, as revealed by fMRI, is induced by these therapeutic approaches (Wittenberg *et al.*, 2003, Boake *et al.*, 2007). Given the evidence from these studies, a number of strategies that are based on pathophysiological mechanisms have been developed in promoting post-stroke motor recovery (Figure 2.7). These include modulations that target for changing the altered excitability in the ipsilesional and contralesional hemispheres, which have been applied as direct cortical stimulation to the brain or as anesthesia applying to the peripheral nerves (Muellbacher *et al.*, 2002, Floel *et al.*, 2004, Fregni *et al.*, 2005, Hummel *et al.*, 2005). These changes are likely to form the neural underpinnings for post-stroke relearning, reestablishing similar underlying physiological learning processes. Motor imagery training, as a relatively new emerging neurorehabilitation approach, is particularly discussed here.

Figure 2.7 Schematic illustration of experimental studies demonstrating the enhancement of neural plasticity via external interventions (such as magnetic or electric stimulation) after stroke. An infarct lesion that weakens functional outcomes within the affected representation area is enclosed by a peri-infarct area with increased excitability. This is accompanied by increased excitability in the contralateral homologous cortical area. The lesion reduces activity in the contralesional hand, while the increased excitability enhances activity in the ipsilesional hand. The changes of excitability might be able to be reverted by electromagnetic stimulation of the central and peripheral nervous system (Carey and Seitz, 2007).

2.4.1 Motor Imagery

A. Rationale and Description

Stroke patients are challenged by many restrictions in managing daily activities, since they are suffering from different level of functional disabilities and impairment of muscle control resulting from stroke. Depending on the patients' own motor ability, active movement therapies sometimes show considerable improvement to the motor function. However, motor execution tasks is often difficult or impossible for many patients with the central nervous system damage, even they have participated early in an active rehabilitation program (Johnson-Frey, 2004). Mental practice has thus been proposed to be used as a therapeutic tool to improve the patients' performance of

motor functions. Motor imagery has raised interest in the area of rehabilitation showing a potential of applying it as an alternative rehabilitation method.

Motor imagery can be defined as a dynamic condition when the representation of an action is internally rehearsed within working memory but without the involvement of any overt movements (Decety *et al.*, 1989, Decety and Jeannerod, 1995, Decety and Grezes, 1999, Solodkin *et al.*, 2004). It is now treated as an engaging new 'backdoor' gateway to approach the motor system and rehabilitation throughout stroke recovery processes (Jackson *et al.*, 2001). It is different from active and passive motor therapeutic treatments, in which motor imagery is not dependent on residual function but still incorporates voluntary drive. Motor imagery may offer an effective way of activating the brain areas normally engaged in motor planning and execution of the paralyzed limb (Weiss *et al.*, 1994). The stroke patients who participated in the mental practice protocol were observed to exhibit increased use of their affected limb, and improved quality of movement, as well as higher motor assessment scores obtained after the intervention (Page *et al.*, 2005).

B. Neuroimaging Findings

Both cross-sectional and longitudinal functional neuroimaging studies have revealed that reorganization of injured brain occurs to compensate for motor deficits after stroke. Increased activity in pre-existing networks involving the disassociated motor cortex after subcortical stroke and the infarct border after cortical stroke has been found as the major mechanism promoting recovery of motor functions. An emerging notion that the greater recruitment of the ipsilesional motor network, the better is the recovery has been proposed. This notion was further enhanced by the increased activity of the ipsilesional primary motor cortex (M1) triggered by motor training and acute pharmacological interventions, in parallel with improved motor

function (Calautti and Baron, 2003).

Primary motor cortex which is a key element associated with improved motor function is also suggested to be involved in motor imagery as proven by direct cellular recordings in the primates (Georgopoulos *et al.*, 1989). Motor imagery may therefore act as a substitute for executed movement in a way to stimulate the motor network in patients after stroke. It is also proposed that motor imagery training can solely enhance motor performance and induce similar cortical plastic alterations, offering a valuable alternative when physical training is not feasible (Jackson *et al.*, 2003). Understanding the brain responses associated with motor imagery in chronic stroke would then be crucial and helpful in developing useful rehabilitation strategies. To date, the brain activation patterns in stroke patients are not fully understood. The involvement of primary motor cortex which is considered as a major target of post-stroke rehabilitation in motor imagery is unresolved. Therefore, the precise mechanism and biological basis for motor recovery in stroke patients after the implementation of motor imagery training are still largely unknown.

Neuroimaging techniques can provide high spatial resolution anatomical and functional information of human brain which is useful for investigation of brain activities in a non-invasive way. Various functional neuroimaging studies involving positron emission tomography (PET) and functional magnetic resonance imaging (fMRI) have been conducted for the investigation of motor imagery in healthy subjects (Boecker *et al.*, 2002, Naito *et al.*, 2002, Solodkin *et al.*, 2004, Lacourse *et al.*, 2005) and stroke patients (Lehericy *et al.*, 2004, Kimberley *et al.*, 2006). Consistent involvement of the non-primary motor areas, but only weak and inconsistent involvement of primary motor cortex during motor imagery was reported by the current literature in healthy subjects and in stroke patients. However, while the functional neuroimaging studies failed to show the consistent activation in

primary motor cortex, studies using other modalities such as magnetoencephalography (MEG) (Schnitzler *et al.*, 1997, Kawamichi *et al.*, 1998), electroencephalogram (EEG) (Pfurtscheller *et al.*, 1999, Caldara *et al.*, 2004), and transcranial magnetic stimulation (TMS) (Fadiga *et al.*, 1999, Hashimoto and Rothwell, 1999, Vargas *et al.*, 2004, Cicinelli *et al.*, 2006), demonstrated consistent involvement of M1 activation.

Although there was seemingly weak and inconsistent activation of M1 during motor imagery as reported by the previous studies using functional neuroimaging, Sharma et al. (2006) suggested that even a weak activation might be sufficient to avoid learned non-use and motivate the motor representations leading to small active movement, bridging the gap across to active movement therapies.

2.5 Basis of Study

There is always a question, arising from stroke clinicians and researchers, that: How can rehabilitation directly influence positive, adaptive changes in the brain in association with functional recovery after stroke? Given the insights from neuroscience and neuroimaging, we now have a chance to look into the brain and to explore its adaptations in response to the damage and in relation to the recovery. In this study, we would like to take advantage of neuroimaging technique, MRI, to look into the features of brain reorganization and remodeling in patients after stroke from both functional and structural perspectives, and to correlate these features with functional outcomes of the stroke patients. Moreover, motor imagery (MI), as a relatively new emerging neurorehabilitation approach, is highlighted in this study. The details and rationale behind this study are discussed below.

Since execution of motor tasks is often difficult or impossible for many persons after stroke (Johnson-Frey, 2004), mental practice such as MI has been used as an

alternative rehabilitation strategy to improve their motor performance (Jackson *et al.*, 2003), before they have full range of motion. MI is defined as a dynamic state when an action representation is internally rehearsed within working memory, but without the involvement of any overt movements (Decety *et al.*, 1989, Decety and Jeannerod, 1995, Decety and Grezes, 1999). It is treated as an intriguing new 'backdoor' gateway to approach the motor system and rehabilitation at all stages of stroke recovery (Jackson *et al.*, 2001). It can incorporate with voluntary drive to activate the brain regions normally engaged in planning and controlling movements of the paralyzed limb (Weiss *et al.*, 1994).

Functional neuroimaging studies, such as using PET and fMRI, have shown similar activation patterns during motor execution (ME) and MI (Tyszka *et al.*, 1994, Lafleur *et al.*, 2002), and therefore suggested that ME and MI are mediated by a common neural substrate. This speculation is in line with observations of congruent movement timing between actual and mental walking performances (Decety *et al.*, 1989), and similar autonomic response modulation, i.e. increase in heart and respiratory rates (Oishi *et al.*, 2000), during executed and imagined movements. This congruent functional neuroanatomy associated with ME and MI might then support the speculation of efficacy of MI training for rehabilitation of movement disorders (Johnson, 2000, Page *et al.*, 2001, Lacourse *et al.*, 2004).

Although these findings provide a relatively comprehensive understanding of the patterns of motor network activation during ME and MI, the question whether the congruent functional neuroanatomy associated with ME and MI can be conserved in stroke patients with different motor recovery levels is still unresolved. Understanding the neural substrates that interact with different levels of functional outcomes is therefore important in enriching the neuroscience-based knowledge to work out a better stroke rehabilitation strategy. In order to achieve this, we need to better

understand how the brain adapts after stroke to coordinate ME and MI.

Furthermore, from a structural perspective, structural remodeling of white matter in the ipsilesional and contralesional sensorimotor cortices has been demonstrated in both animal models of stroke (Brus-Ramer *et al.*, 2007) and stroke patients (Schaechter *et al.*, 2009), and is found to be associated with the level of motor recovery (Brus-Ramer *et al.*, 2007, Schaechter *et al.*, 2009). This motor-related structural remodeling is especially obvious and commonly shown in the corticospinal tract (CST), such that the structural integrity of the CST becomes a major determinant of motor deficit (Lindberg *et al.*, 2007). However, in addition to the brain tissue damage localized at the periphery of the lesion, recent studies pointed out that the brain network far away from the lesion could also be altered (Alstott *et al.*, 2009, Crofts *et al.*, 2011). This alteration is suggested as a secondary white matter degeneration which appears in remote regions interconnected, directly or indirectly, with the primary damaged area (Crofts *et al.*, 2011). Nevertheless, the effect of this remote alteration on motor control/behavior of stroke patients is still under exploration. More understanding to the entire brain adaptation after a stroke might provide a more comprehensive picture of the interactions between structural connectivity remodeling and post-stroke motor impairments. Such information may be of value in redefining potential neural substrates that target post-stroke motor recovery.

DTI which is a noninvasive magnetic resonance technique can measure the random motion of water molecules in brain tissue. It has been used to reveal brain abnormalities in a variety of diseases, including strokes (Nucifora *et al.*, 2007). This technique is based on the extraction and characterization of the changes in diffusion anisotropy in brain tissue (Werring *et al.*, 2000), where the diffusion happens to be unequal in all directions. Combined with fiber tractography which is used to

visualize and quantify the integrity of fiber tracts, it might provide a diverse way to reveal the structural remodeling following a stroke (Crofts *et al.*, 2011).

Chapter 3 Methodology

3.1 Study I: Study on Neural Correlates of Motor Impairment During Motor Imagery and Motor Execution Using fMRI

3.1.1 Overview

In this study, ten chronic stroke patients having left subcortical ischemic lesions and right hemiparetic limbs, and ten unimpaired subjects were included to study their activation patterns during ME and MI. Subcortical stroke was chosen because the ability of performing MI can be preserved if the lesions were not found at parietal or premotor cortex (Johnson *et al.*, 2002). Meanwhile, subcortical stroke also accounts for 20-30% of all cerebrovascular infarcts (Donnan, 2002), and such a constraint can help us to select a relatively homologous stroke population for the study. All subjects were instructed to perform four motor tasks: (1) ME using left/unaffected wrist (ME_L), (2) MI using left/unaffected wrist (MI_L), (3) ME using right/affected wrist (ME_R), (4) MI using right/affected wrist (MI_R). These data were used to find out (1) the clinical relevancy of cluster volume of activation in stroke during ME_R and MI_R using affected wrist, (2) the clinical relevancy of lateralization of activation in stroke, and (3) the clinical relevancy of congruence in functional neuroanatomy between ME and MI tasks, and between stroke and unimpaired groups. Quantitative measures, including laterality and overlap indices, were derived to quantify the activation features. Moreover, 11 regions of interest (ROIs) in bilateral hemispheres were selected to evaluate the functional activity, including the primary motor area (M1), primary sensory area (S1), supplementary motor area (SMA), premotor cortex (PM), prefrontal cortex (PF), inferior parietal lobule (IPL), precuneus, insula, thalamus and putamen (TH-PU), caudate nucleus and subthalamic nucleus (CN-STN) and

cerebellum. These regions were chosen because these were the important areas involved in the motor network of unimpaired subjects (Walsh *et al.*, 2008). Finally, correlations between the activation features in these 22 ROIs and the residual motor function after stroke were analyzed in this study.

3.1.2 Subjects

Ten right-handed patients (8 males and 2 females, mean age: 55.4 years, SD: 9.9 years) who suffered from first-ever stroke, with time elapsed after stroke more than 12 months, with left subcortical stroke, and with moderate-to-severe upper-limb impairment [Fugl-Meyer score less than 44 out of 66] (Chae *et al.*, 1998) were included in this study (Table 3.1). Ten right-handed unimpaired subjects (6 males and 4 females, mean age: 61.1 years, SD: 15.4 years) without any history of neurological and psychiatric disease were also included to serve as a reference group. In Hong Kong, people who suffer a stroke are generally at the age over 50, and a recent declining trend in age of stroke onset is observed among the patients (Hospital Authority of Hong Kong Special Administrative Region, 2014). Nevertheless, the majority of the stroke sufferers still come from the elderly group aged 65 and above. From the neuroimaging findings, age-related atrophy of the motor cortical regions and corpus callosum is revealed, which is suggested to be correlated with motor declines in the elderly, such as movement slowing, and deficits in balance, gait and coordination (Seidler *et al.*, 2010). Moreover, age-related gross and fine motor decrements, as well as higher cognitive deficits may also result from the degeneration of neurotransmitter systems (e.g. the dopaminergic system). Therefore, these age-related changes in brain structure and system may lead to the brain adaptations of recruiting more widespread brain regions for motor control in the older adults, compared to the young adults (Seidler *et al.*, 2010). In spite of this,

many life-span studies of the age-related differences in motor skill learning revealed that the motor performance declines start early in middle age and not in old age (Voelcker-Rehage and Willimczik, 2006, Voelcker-Rehage, 2008). The same might apply to the brain adaptations to normal aging (Heetkamp *et al.*, 2014). Although the stroke patients and the unimpaired subjects included in this study were relatively younger than the majority of stroke patients in overall stroke population, we speculate that the brain adaptations due to aging would already exhibit in our middle-aged stroke patients, and the findings from this study may be able to extend to the older patients. Furthermore, motor imagery has been proposed that it can be accessible through the acute phase, sub-acute phase or chronic phase of stroke rehabilitation (Johnson, 2000, Johnson *et al.*, 2002). Chronic stroke patients may even have a "hemiplegic advantage" of performing motor imagery more accurately with their paralyzed limb. This "hemiplegic advantage" may be related to a continuing focus on motor planning and/or imagining the movements involving the paralyzed limb that are currently impossible to be executed (Johnson *et al.*, 2002). Besides, chronic stroke patients often have a relatively stable condition and mental state compared with the patients in acute or sub-acute phase, so that they can withstand the long scanning time required in this study. Therefore, chronic stroke patients were recruited for this study.

All unimpaired subjects and stroke patients prior to stroke were right-handed as assessed by the Edinburgh Handedness Inventory (Oldfield, 1971). Subjects were excluded if they had history of alcohol or drug abuse or epilepsy, bilateral infarcts, uncontrolled medical problems, serious cognitive deficits, comprehensive aphasia and other MRI contraindications. A more detailed description of the inclusion and exclusion criteria of subject recruitment is documented in Appendix 1. Consent was obtained from each subject for their participation, and the procedures were approved

by the Research Ethics Committee of the Hong Kong Polytechnic University, and the IRB of Kowloon Central Cluster of Hospital Authority, Hong Kong.

Table 3.1 Demographic details of stroke patients included in fMRI study

Subject	Age/Gender	Type of stroke	Lesion location	Time between stroke and fMRI (months)	Assessment scores			
					MSS_SM (max: 29)	MSS_EF (max: 11)	FMA_SE (max: 42)	FMA_WH (max: 24)
S1	48/M	Ischemic	L PLIC	38	26.2	9.2	22	15
S2	63/M	Ischemic	L PLIC	48	21	8	23	14
S3	62/M	Ischemic	L PLIC	72	22	8	20	13
S4	66/M	Ischemic	L PLIC, putamen	50	21	8	21	17
S5	55/M	Ischemic	L PLIC	72	22.6	8	22	15
S6	47/M	Ischemic	L PLIC	38	21.4	7.6	23	17
S7	50/F	Ischemic	L. globus pallidus, putamen, insula	36	21.8	7.6	23	13
S8	65/M	Ischemic	L. globus pallidus, putamen	132	19	3.2	17	8
S9	62/M	Ischemic	L. globus pallidus, putamen	33	14.8	3.4	15	7
S10	36/F	Ischemic	L. globus pallidus, putamen, insula	12	7.4	0	4	1

L – Left; M – Male; F – Female; PLIC – posterior limb of internal capsule; MSS_SM – Motor Status Scale (Shoulder Movement); MSS_EF – Motor Status Scale (Elbow/Forearm); FMA_SE – Fugl-Meyer assessment (Shoulder/Elbow); FMA_WH – Fugl-Meyer assessment (Wrist/Hand).

3.1.3 Clinical Assessments

Assessments were made for each patient's voluntary motor function of the paretic upper limb, using the Motor Status Scale (MSS) (Ferraro *et al.*, 2002) and Fugl-Meyer Assessment (FMA) (Fugl-Meyer *et al.*, 1975) protocols, by an experienced assessor in our university clinic who was blind to the patient's group identity. The full scoring sheets of both MSS and FMA for upper extremity motor assessment are attached in Appendices 2 and 3 respectively. Since wrist motor task was used in this study, attention was particularly given to the forearm assessments, including MSS forearm movements (MSS_EF) and FMA wrist and hand movements (FMA_WH). These clinical assessments have been used in prior studies to assess for motor function in upper-extremity rehabilitation training, and have been proven to be reliable and valid for assessing motor functions following stroke (Ferraro *et al.*, 2002, Gladstone *et al.*, 2002). Excellent inter-rater reliability has been shown for FMA upper extremity motor score (intra-class correlation coefficient, ICC=0.97) (Gladstone *et al.*, 2002), and for MSS shoulder/elbow score (ICC=0.99) (Ferraro *et al.*, 2002).

3.1.4 Image Acquisition

Subjects were scanned with a 1.5-Tesla MRI scanner (Siemens Avanto, Germany) using a 12-channel head coil. The following imaging datasets were acquired: (1) standard high-resolution sagittal images, using a volumetric T1-weighted gradient echo sequence (TR = 11ms, TE = 4.94ms, flip angle = 15 degrees, FOV = 256mm×256mm, matrix = 256×224, resolution = $1\times1\times1mm^3$); (2) T2-weighted images, using a T2*-weighted turbo spin echo sequence (TR = 4330ms, TE = 98ms, flip angle = 150 degrees, FOV = 220mm×220mm, matrix = 336×384, gap = 1mm, resolution = $0.57\times0.57\times6mm^3$); (3) blood oxygenation level dependent (BOLD)

fMRI images, using a gradient-echo echo-planar-imaging sequence (TR = 2000ms, TE = 32ms, flip angle = 90 degrees, FOV = 192mm×192mm, matrix = 64×64, gap = 1mm, resolution = 3×3×6mm^3). Each fMRI session lasted for 6 minutes and was preceded by 4 dummy scans for equilibrium of MRI signals.

3.1.5 Motor Tasks

Since the stroke patients in this study experienced moderate-to-severe motor recovery for their proximal upper limb with some residual wrist motor function (FMA_WH>0), wrist movement was adopted in our study design. Moreover, in order to minimize the incidence of abnormal contraction of other muscle groups of proximal segments from the subjects, a magnetic resonance compatible wrist orthotic device, which was fully made of plastic, was designed (Figure 3.1). All subjects, including unimpaired subjects, used the wrist orthotic device to standardize their upper limb position during the MR scanning, and to fix the subjects' forearms to ensure that they focused on performing wrist movement and minimize the contraction of other irrelevant muscles.

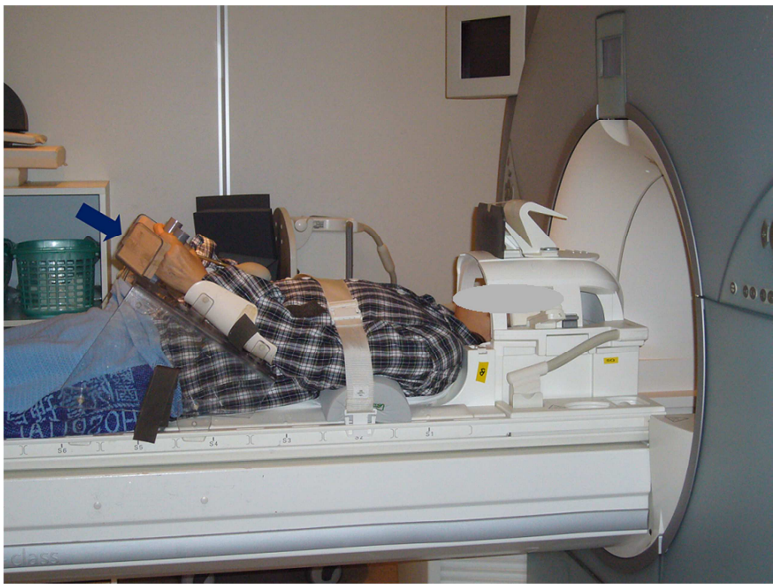


Figure 3.1 The experimental setting for fMRI experiment. The subject used the wrist orthotic device (indicated by an arrow) to standardize her upper limb position and to fix her forearms during the MR scanning. A cushion was placed under each of her upper arms, so that her elbows can rest on the cushions during the resting phase. A small projection screen was mounted on the head coil for the presentation of instructions to the subject.

Subjects were asked to perform both executed and imagined wrist extension unilaterally in the study. The sequence of motor tasks was: (1) ME_L, (2) MI_L, (3) ME_R, (4) MI_R. This sequence was chosen because all our patients in this study were right-limb impaired. We therefore would like the patients to execute or imagine with their unimpaired (left) limbs before practicing with their impaired (right) limbs. To avoid any deviations of results created from the task sequence, the unimpaired subjects were also instructed to follow the same protocol during fMRI scanning. The four motor tasks were repeated so that each subject performed 2 runs for each of the four tasks for averaging. A blocked paradigm was used, which consisted of 19 epochs: 10 resting and 9 activation epochs. Each resting and activation epoch lasted for 20 s, except for the first and the last resting epochs, which lasted for 10 s (17 epochs × 20 s + 2 epochs × 10 s = 360 s = 6 min). In the resting epochs, a white crosshair in a black background was presented. In the activation epochs, depending on the tasks

they performed, a white ‘L’ character for left wrist or a white ‘R’ character for right wrist was presented (Figure 3.2).

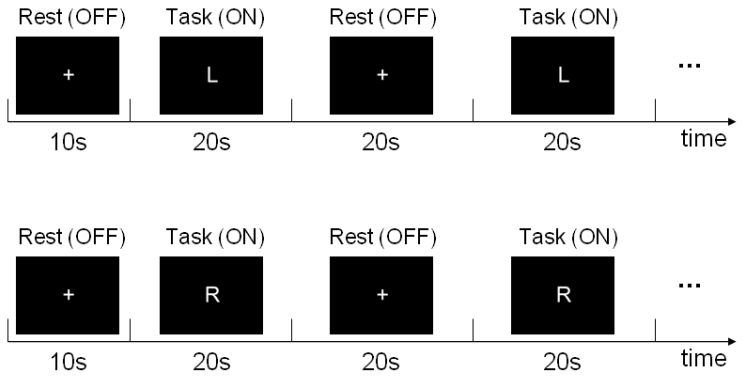


Figure 3.2 Experimental paradigm. The upper sequence was used for left motor execution or imagery, while the lower sequence was used for right motor execution or imagery.

For ME tasks, the subjects were asked to perform repetitive wrist extension during activation phase, and the pace was asked to be maintained at a period of around 2 s~4 s for one movement cycle of the wrist extension, to avoid any muscle fatigue. For MI tasks, subjects were instructed to do kinesthetic imagery during activation phase (avoiding visual imagery or counting) with the same pace as in the previous ME task. During resting phase, they were instructed to relax, not to make any movements or not to think about any movement. All subjects were instructed to keep their eyes opened during scanning.

3.1.6 fMRI Data Analysis

The data acquisition and MRI scanner operation were assisted by the radiographers at Queen Elizabeth Hospital, who were blind to the clinical data of the included subjects. Then, the MRI data processing and analyses were done by the author of this thesis. All the data were gone through the same analysis model and program to ensure that there was small chance for the author to manipulate the final

results.

All fMRI data were analyzed with a general linear model approach using Analysis of Functional NeuroImages (AFNI) software (Cox, 1996) (National Institute of Mental Health, http://afni.nimh.nih.gov). The following analysis strategy has been adopted similarly by Chan et al. (Chan *et al.*, 2009) to study language processing in healthy adults. The first 5 brain volumes in each functional dataset collected before reaching equilibrium magnetization were removed. The functional data for each subject was motion-corrected and co-registered to the 90th image of the first functional dataset using the three-dimensional volume registration. Each functional dataset was normalized to its mean intensity value across the time series. The changes of MR signal activation relative to resting were derived in multiple regression analysis. The impulse response function to each condition was then estimated with 1-sec resolution using deconvolution. A separate regressor was used to model the response in each 1-sec period in a 10-sec window following each stimulus presentation. The six motion parameters were also included as regressors for the removal of residual motion correlated activity. The hemodynamic response magnitude to each motor task was calculated by averaging the beta weights of the regressors of the response.

Individual subject analysis was performed to depict the prevalence of activation patterns across the subjects. Single subject analysis was performed on unsmoothed and normalized functional data. Individual subject brain volume with hemodynamic response magnitude to each motor task were registered onto each subject's anatomical scan and transformed to the standardized space of Talairach and Tournoux (1988). The brain volume in Talairach space, with the response magnitude to each motor task for each subject, was spatially smoothed with a Gaussian filter of full-width half-maximum (FWHM) 4mm to reduce the false

negative error due to individual variability of brain shape.

Group analysis was then performed to obtain a general indication of activated brain areas, correlated to ME and MI tasks. A two-way mixed-effect analysis of variance (ANOVA) was performed on each voxel in a standard space, considering the contrasts on the given task type (ME_L vs. MI_L; ME_R vs. MI_R) (fixed effect), and serving each individual subject as the repeated measure (random effect). The overall significance level of $\alpha<0.05$ was set to prevent type I error. Based upon Monte Carlo simulation with 1000 iteration runs via AlphaSim program in AFNI (Cox, 1996) on the brain volume, it estimated that a 162 mm^3 contiguous volume (3 voxels, each measuring $3\times3\times6mm^3$) would provide the significance level $\alpha=0.02$, which met the overall threshold of $P<0.05$. The threshold (corrected $P<0.05$) was used for group analysis to identify the statistically significant activation areas. Finally, conjunction analysis was performed to find out overlapping regions in the brain among the group of subjects during the four motor tasks, to identify the common activations between the MI and ME tasks, and between the stroke and unimpaired groups.

3.1.7 Quantitative Analysis

Quantitative measures, including laterality and overlap indices, were also derived to quantify the activation features during the tasks, and to evaluate whether these features are correlated with the motor functions. Laterality index (LI) has been used to show the lateralization of activated voxels in response to the tasks, and introduced as a quantitative indication of stroke recovery (Cramer *et al.*, 1997). It was calculated as

$$LI = \frac{ROI_C - ROI_I}{ROI_C + ROI_I}$$

where ROI_C is activation volume from anatomical areas in the hemisphere

contralateral to the limb performing the task and ROI_I is the activation volume from anatomical areas in the ipsilateral hemisphere. It has a range from -1 to +1, with a negative number indicating primarily ipsilateral activation and a positive number indicating activation primarily contralateral to the limb performing the task. A large negative or positive LI value (<−0.5 or >0.5) indicates that brain activation is lateralized heavily to hemisphere ipsilateral or contralateral to the target limb respectively. For LI value approaching zero, it reflects that similar volumes of activated voxels are recruited from both ipsilateral and contralateral hemispheres, showing weak lateralization of brain activation.

Overlap index (OI) was used to represent the percentage of overlapping of activated voxels in different tasks/groups, for assessing the functional convergence and/or segregation of task-selective brain areas. The computation of OI was based on the results from conjunction analysis as follows:

$$OI = \frac{ROI_a \cap ROI_b}{ROI_a \cup ROI_b}$$

where the numerator ($ROI_a \cap ROI_b$) is the volume of overlapping voxels between the same ROIs in two different tasks/groups, while the denominator ($ROI_a \cup ROI_b$) is the union of the two compared ROIs. In order to find out the clinical relevancy of congruence in functional neuroanatomy between the tasks using affected wrist and between stroke and unimpaired groups on the affected/right-handed tasks, the OI was derived separately for the following comparisons: (a) within-group comparison on affected-wrist tasks (stroke ME_R vs. stroke MI_R); (b) between-group comparison on MI_R task (stroke MI_R vs. unimpaired MI_R); and (c) between-group comparison on ME_R task (stroke ME_R vs. unimpaired ME_R). Besides, the OI was also derived for the comparisons: (d) within-group comparison on ME tasks (unimpaired ME_R vs. unimpaired ME_L); (e) between-group comparison on ME_L task (stroke ME_L vs.

unimpaired ME_L) to provide more information about the lateralization in the unimpaired subjects, and congruence in functional neuroanatomy between stroke and unimpaired groups on the unaffected/left-handed ME tasks.

3.1.8 Statistical Analysis

A two-group between-subjects multivariate analysis of variance (MANOVA) was conducted on the cluster volumes and LIs (dependent variables) to determine whether there were any differences between subject groups on these activation features. Furthermore, correlations between clinical assessment scores and several quantitative measures, including cluster volume, LI and OI during right/affected limb tasks, were further assessed using Spearman's correlations. All statistical analysis was conducted in IBM Statistical Package for the Social Sciences (SPSS) (version 19), and the level of statistical significance was set at 0.05.

3.2 Study II: Study on Remodeling of Structural Connectivity and Its Correlation with Motor Impairments Using DTI

3.2.1 Overview

In this study, one of the stroke patients was excluded, since the anatomical image of this patient was contaminated by motion artifacts, which led to failure in subsequent data analysis. One of the unimpaired subjects with similar age was also excluded to match the total number of stroke patients included in this study. Therefore, only nine chronic stroke patients having left subcortical ischemic lesions and right hemiparetic limbs, and nine unimpaired subjects were included to study the post-stroke structural remodeling.

Moreover, instead of using voxel-based assessment for assessing the post-stroke structural changes, network analysis was used and recommended in this study. Using voxel-based assessment for tracing the post-stroke structural changes might be quite challenging, since measurement of the changes can be highly influenced by the variations in stroke topography among stroke population (Crofts *et al.*, 2011). Moreover, since brain lesions often disrupt the neighboring white matter, it might induce erroneous judgment if fiber tracking is based on seed regions extracted from non-lesioned neuroanatomy (Schonberg *et al.*, 2006). Therefore, we applied network analysis, which is an alternative and comprehensive approach of assessing structural connectivity (Bullmore and Sporns, 2009), to characterize structural brain networks. It can avoid the assumptions on the location of seed regions by obtaining the regions of interest (ROIs) from automated parcellation of brain regions for each individual (Fischl *et al.*, 2004).

The flow of the analysis involves dividing up the brain into cortical and subcortical areas to form the nodes of the network, and measures the connectivity between nodes to characterize the properties of fiber tracts (Hagmann *et al.*, 2008).

Three main connectivity measures were included to evaluate the post-stroke structural changes: fractional anisotropy (FA), connection weight (CW) and connection strength (CS). Among these measures, FA is the commonest parameter which is used to assess white matter properties in relation to the fiber density, axonal diameter, and myelination status (Beaulieu, 2002). A decline in FA was found to be related to loss of axonal integrity, leading to Wallerian degeneration (Watanabe *et al.*, 2001). CW is used as a means to capture the connection density between two regions (Hagmann *et al.*, 2008). A higher value in CW could indicate that the connection has a shorter path length and/or greater number of fibers. CS can measure the extent to which the node is connected to the rest of the network (Hagmann *et al.*, 2008). A node with higher CS makes stronger connections (Hagmann *et al.*, 2008), since CS will increase if the connections to the region are more intensive. It can highlight the importance of structural integration at node level. These measures were used to localize which fiber tracts are affected by the lesions when compared with unimpaired subjects. Finally, correlations between the motor impairments which were assessed by clinical assessment scores (Motor Status Scale, MSS (Aisen *et al.*, 1995) and Fugl-Meyer Assessment, FMA (Fugl-Meyer *et al.*, 1975)) and the connectivity measures were evaluated in strokes to examine the remodeling of structural connectivity. The results may provide an opportunity to investigate inter-patient connectional variability and to relate it to differences in individual motor recovery levels.

3.2.2 Subjects

Nine right-handed patients (6 males and 3 females, mean age: 53.7±9.8 years) who had suffered a first-ever stroke (with time elapsed after stroke of more than 12 months; with left subcortical stroke; and with moderate-to-severe upper-limb

impairment) were included in this study (Table 3.2). Nine right-handed unimpaired subjects (5 males and 4 females, mean age: 60.2±16.0 years) without any history of neurological or psychiatric disease were also included to serve as a reference. Subjects were excluded if they had history of alcohol or drug abuse or epilepsy, bilateral infracts, uncontrolled medical problems, serious cognitive deficits, comprehensive aphasia and other MRI contraindications. Consent was obtained from each subject for their participation, and the procedures were approved by the Research Ethics Committee of the Hong Kong Polytechnic University, and the Kowloon Central Cluster of Hospital Authority, Hong Kong.

Table 3.2 Demographic details of stroke patients included in DTI study

Subject	Age/Gender	Lesion location	Lesion volume (mm^3)	Time between stroke and fMRI (months)	Assessment scores			
					MSS_SM (max: 29)	MSS_EF (max: 11)	FMA_SE (max: 42)	FMA_WH (max: 24)
S1	48/M	L PLIC	736	38	26	9.2	22	15
S2	63/M	L PLIC	280	48	21	8	23	14
S3	62/M	L PLIC	1112	72	22	8	20	13
S4	47/M	L PLIC	1968	38	21.4	7.6	23	17
S5	50/F	L pallidum, putamen, insula	17640	36	21.8	7.6	23	13
S6	50/F	L pallidum, putamen, insula	12008	28	21	8	21	12
S7	65/M	L pallidum, putamen	7560	132	19	3.2	17	8
S8	62/M	L pallidum, putamen	6192	33	14.8	3.4	15	7
S9	36/F	L pallidum, putamen, insula	28992	12	7.4	0	4	1

L – Left; M – Male; F – Female; PLIC – posterior limb of internal capsule; MSS_SM – Motor Status Scale (Shoulder Movement); MSS_EF – Motor Status Scale (Elbow/Forearm); FMA_SE – Fugl-Meyer assessment (Shoulder/Elbow); FMA_WH – Fugl-Meyer assessment (Wrist/Hand).

3.2.3 Clinical Assessments

Each patient was assessed by a blind assessor regarding their voluntary motor function of the paretic upper limb using MSS and FMA. Subdivision was done for the two assessment scores: MSS shoulder movements (MSS_SM), MSS forearm movements (MSS_EF); FMA shoulder and elbow movements (FMA_SE) and FMA wrist and hand movements (FMA_WH). These clinical assessments are used frequently for body function evaluation in upper-extremity rehabilitation training, and their reliability and validity in assessing stroke motor functions has been proven (Hsieh *et al.*, 2009).

3.2.4 Image Acquisition

Subjects were scanned with a 1.5-Tesla MRI scanner (Siemens Magnetom Avanto) using a 12-channel head coil. The following imaging datasets were acquired: (1) standard high-resolution sagittal images were acquired using a volumetric T1-weighted gradient echo sequence (TR = 11 ms, TE = 4.94 ms, flip angle = 15 degrees, FOV = 256×256 mm, matrix = 256×224, resolution = 1×1×1 mm^3); (2) T2-weighted images were acquired using a T2*-weighted turbo spin echo sequence (TR = 4330 ms, TE = 98 ms, flip angle = 150 degrees, FOV = 220×220 mm, matrix = 336×384, gap = 1 mm, resolution = 0.57×0.57×6 mm^3); (3) diffusion-weighted images were acquired using a diffusion-weighted single-shot EPI sequence (TR = 10900 ms, TE = 96 ms, FOV = 256 mm, matrix = 128×128, resolution = 2×2×2 mm^3). The diffusion-weighted images were acquired along 12 different diffusion directions with a *b*-value of 1000 s/mm^2, and an additional baseline (b=0) image. This sequence was repeated 2 times for signal averaging.

3.2.5 Diffusion Data Processing and Fiber Tractography

The data acquisition and MRI scanner operation were assisted by the radiographers at Queen Elizabeth Hospital, who were blind to the clinical data of the included subjects. Then, the MRI data processing and analyses were done by the author of this thesis. All the data were gone through the same analysis model and program to ensure that there was small chance for the author to manipulate the final results.

Raw diffusion imaging data were pre-processed with FMRIB's Diffusion Toolbox (FDT) implemented in FMRIB Software Library (FSL) (FMRIB Analysis Group, Oxford, UK). The processing included correction for eddy currents and head motion, brain extraction and fitting of diffusion tensors on corrected data (Johansen-Berg *et al.*, 2004). The pre-processed data were then reconstructed using a software package Diffusion Toolkit (Athinoula A. Martinos Center for Biomedical Imaging, USA). FA, eigenvectors, and eigenvalues of the diffusion tensor were calculated at each voxel of the diffusion image. Fiber tracking was then performed using standard fiber assignment by continuous tracking method (Mori *et al.*, 1999). Tracking stops at predefined thresholds of a diffusion-weighted image and a turning angle of 60° to limit the detection of spurious fibers. The tracks were then smoothed by a B-spline filter to remove any redundant track points and segments.

3.2.6 Structural Connectivity Mapping

Based on the T1-weighted image, white and grey matter segmentation was performed in FreeSurfer (Athinoula A. Martinos Center for Biomedical Imaging, USA) to reconstruct and parcellate brain volume to produce outputs consisting of labels corresponding to the white matter, the cortex, and the deep gray nuclei (Fischl *et al.*, 2002). The labeled mesh of each individual subject was then aligned with his/her diffusion dataset.

However, since the segmentation technique is usually applied to non-lesioned neuroanatomy, segmentation errors such as misclassifying the lesions as ventricles or nearby areas might be induced in our cases. Lesion segmentation was therefore done for each stroke patient to estimate his/her infarct extent and to correct segmentation errors induced during the automated parcellation. It was performed by feeding diffusion-weighted, T1-weighted and T2-weighted images into k-means clustering algorithm (Tou and Gonzalez, 1974) for roughly partitioning the pixel intensities into 5 clusters. The non-infracted areas, such as the areas of ventricles and cerebrospinal fluid, were removed from the lesion segmentation. After the correction of lesions from the automated parcellation, 68 cortical and 12 subcortical regions (Figure 3.3), which covered the entire cortices and subcortical structures of both left and right hemispheres, were chosen for investigation of structural connectivity changes.

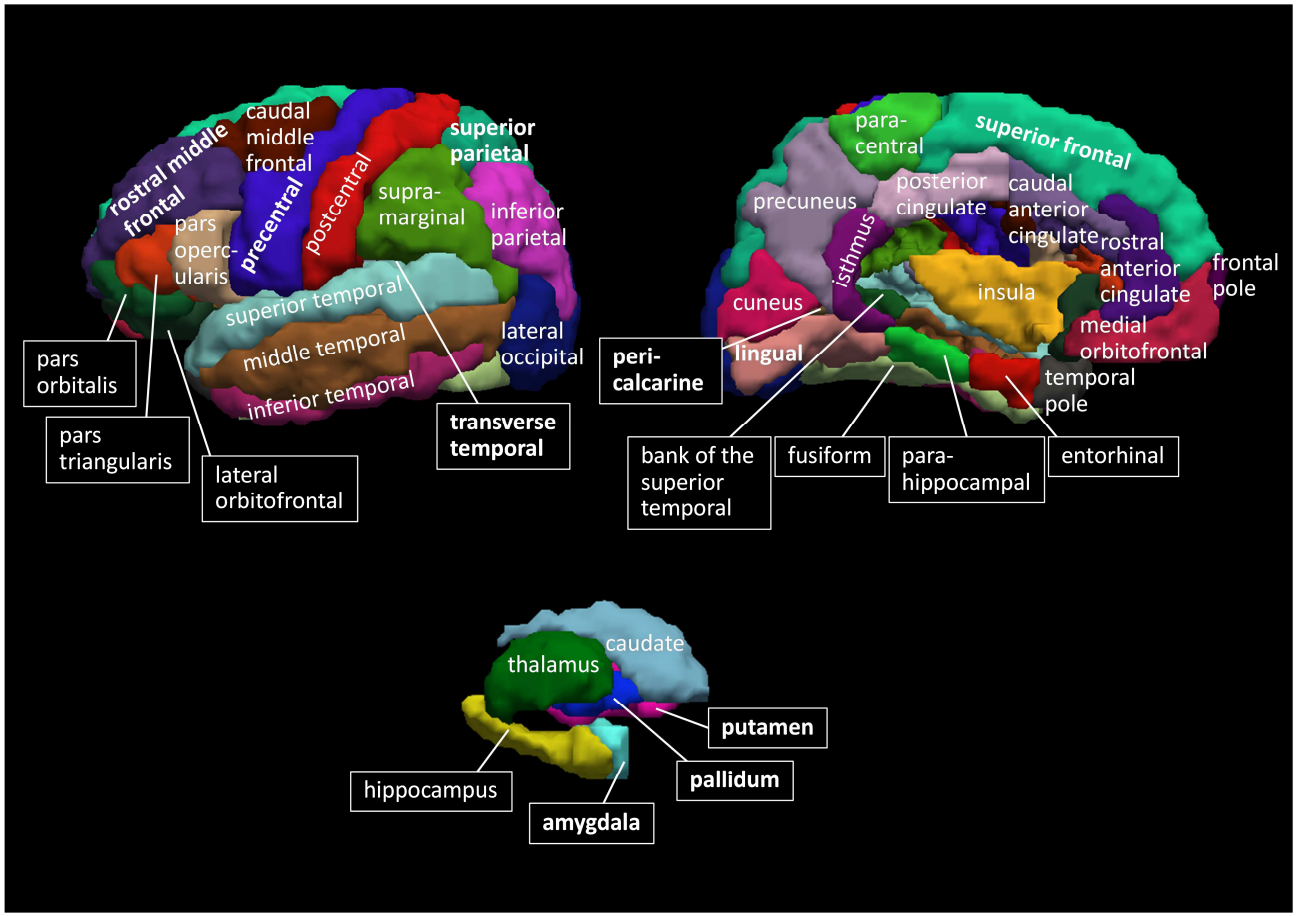


Figure 3.3 Segmentation of brain into 34 cortical and 6 subcortical regions for one hemisphere.

The outputs of fiber tractography and ROIs creation were finally combined to map connection matrices of CW and FA. Each ROI became a node in the matrix. Two nodes were connected if there was at least one fiber with end-points in them. The first connection matrix maps the CW between ROIs and is calculated as:

$$\mathrm{CW} = \frac{1}{\mathrm{V}} \sum_i \frac{1}{l(i)}$$

where V is total volume of the two ROIs, Σ_i is sum over all fibers connecting the two ROIs, and the correction term $l(i)$ is the length of a specific fiber (Hagmann *et al.*, 2008). We then re-sampled the raw CWs into a Gaussian distribution with a mean of 0.5 and a standard deviation of 0.1, to normalize the scale of the measure for each subject (Alstott *et al.,* 2009). The second matrix maps the average FA along a connection which was linking two ROIs. Finally, CS and regional FA were calculated as the sum of all the re-sampled CWs and the average FAs for each node, respectively.

3.2.7 Statistical Analysis

Pearson correlation analysis was done to identify the correlation patterns of structural connectivity properties with motor impairments. Partial correlation analysis was then done to look at the changes to the correlation patterns after controlling for the CST's structural properties. Finally, a two-group between-subjects multivariate analysis of variance (MANOVA) was done on those significantly correlated regions with their connectivity properties to assess whether there are any differences between unimpaired subjects and stroke patients. All statistical procedures were done using IBM Statistical Package for the Social Sciences (SPSS) (version 19), and the level of statistical significance was set at 0.05.

Chapter 4 Results

4.1 Study I: Study on Neural Correlates of Motor Impairment During Motor Imagery and Motor Execution Using fMRI

The ten patients (Table 3.1) were having moderate-to-severe upper-limb impairment (FMA score less than 44 out of 66). Their average scores were MSS: 26.0 (SD = 8.1) and FMA: 31.0 (SD = 10.9). The patients mainly exhibited lesions at the left posterior limb of internal capsule (PLIC), pallidum and putamen. Since all patients had left subcortical lesions, left hemisphere is regarded as ipsilesional hemisphere, whereas right hemisphere is contralesional hemisphere.

4.1.1 Activation Patterns and Clinical Correlations

The anatomical foci of positive activation clusters during ME and MI were presented in Table 4.1 and 4.2. The corresponding brain activation maps obtained by group analysis for the unimpaired and stroke groups were shown in Figure 4.1. The brain activation patterns of the unimpaired group during ME_L (non-dominant) and ME_R (dominant) were similar, in which positive activation was commonly found in M1, S1, SMA, PM, PF, IPL, insula and cerebellum in bilateral hemispheres of the unimpaired group (Table 4.1, Figure 4.1). The only difference of activation between ME_L and ME_R was demonstrated at the precuneus, which bilateral precuneus was found activated during ME_L, but only left precuneus was found activated during ME_R. During MI_L (non-dominant) and MI_R (dominant), positive activation was commonly found in SMA, PF, IPL and cerebellum in bilateral hemispheres, and in M1 contralateral to the limb performing the tasks. The differences were revealed at S1 and insula. Bilateral S1 was activated during MI_L while only left S1 was activated

during MI_R. Bilateral activation in insula was shown during MI_R but only left insula was activated during MI_L (Table 4.1, Figure 4.1).

In stroke group, positive activation was found in S1, SMA, PM, PF, IPL and cerebellum in bilateral hemispheres during ME_L (unaffected) and ME_R (affected). The differences were revealed at M1 and insula. Bilateral M1 was activated during ME_R while only right M1 was activated during ME_L. Bilateral activation in the insula was only observed during ME_L but was not shown during ME_R (Table 4.2, Figure 4.1). During MI_L (unaffected) and MI_R (affected), positive activation was found in M1, S1, SMA, PM, PF and IPL in bilateral hemispheres, and in the left precuneus. Right precuneus and right insula were found additionally activated during MI_L, while left insula was activated only during MI_R (Table 4.2, Figure 4.1).

Table 4.1 Summary of anatomical foci in Talairach coordinates showing positive BOLD responses from unimpaired group during ME_L, MI_L, ME_R and MI_R

Activated areas	ME_L				MI_L				ME_R				MI_R			
	x	*y*	*z*	volume	*x*	*y*	*z*	volume	*x*	*y*	*z*	volume	*x*	*y*	*z*	volume
Cortical region																
L. M1 (BA 4)	27	25	55	1318					32	23	55	3639	28	26	56	1051
R. M1 (BA 4)	-32	23	55	3744	-29	24	52	1139	-21	25	58	236				
L. S1 (BA 1, 2, 3, 5)	27	30	53	571	55	24	35	399	38	28	52	7625	30	29	52	195
R. S1 (BA 1, 2, 3, 5)	-36	28	52	6883	-31	29	51	1038	-56	23	32	115				
L. SMA (BA 6m, 24c)	6	8	47	4641	4	2	48	1096	5	6	47	4049	7	10	51	3139
R. SMA (BA 6m, 24c)	-7	7	49	5470	-6	4	49	2351	-4	5	51	1746	-5	3	52	2207
L. PM (BA 6l)	18	14	62	556					27	13	61	2033				
R. PM (BA 6l)	-31	12	59	1460	-27	15	58	181	-45	-1	40	591	-46	-1	39	462
L. PF (BA 46, 44)	54	-11	12	1465	52	-10	11	1676	54	-9	11	1815	53	-9	11	1796
R. PF (BA 46, 44)	-52	-22	13	3506	-57	-11	9	460	-53	-7	12	1271	-55	-9	12	1753
L. IPL (BA 40)	40	44	44	1613	57	34	26	1745	41	40	45	4020	40	46	44	326
R. IPL (BA 40)	-40	41	49	1617	-57	42	42	113	-55	27	26	827	-58	37	40	569
L. Precuneus (BA 7)	24	47	49	220					26	49	50	366				
R. Precuneus (BA 7)	-19	47	50	249												
L. Insula	36	-8	-1	295	43	-8	3	1678	43	25	17	1805	40	-7	7	1790
R. Insula	-46	28	18	1238					-43	3	5	1402	-43	-6	6	629
Subcortical region																
L. Thalamus	10	12	7	1956					13	17	7	4354	14	19	5	205
R. Thalamus	-15	16	8	3051					-10	13	5	2085				
L. Putamen	23	1	4	3234	24	1	2	1558	25	4	3	3429	26	4	5	1322
R. Putamen	-24	3	5	4007	-26	8	6	927	-22	1	6	1361	-24	4	4	720
L. Caudate nucleus	8	-5	6	352					13	9	19	130				
R. Caudate nucleus	-9	-8	6	591	-6	-5	2	111	-12	3	16	131				
Cerebellum																
L. Cerebellum	17	52	-22	20307	11	49	-15	3751	25	51	-22	3214	20	53	-24	718
R. Cerebellum	-21	57	-20	14439	-38	38	-29	165	-16	56	-20	23440	-22	51	-21	9431

Abbreviations: L – left; R – right; BA – Brodmann area. The cluster volumes are measured in mm^3. Activated clusters were significant at $p < 0.05$ corrected for multiple comparisons.

Table 4.2 Summary of anatomical foci in Talairach coordinates showing positive BOLD responses from patient group during ME_L, MI_L, ME_R and MI_R

Activated areas	ME_L – Rest				MI_L – Rest				ME_R – Rest				MI_R – Rest			
	x	*y*	*z*	volume	*x*	*y*	*z*	volume	*x*	*y*	*z*	volume	*x*	*y*	*z*	volume
Cortical region																
L. M1 (BA 4)					20	27	56	316	32	24	55	1409	30	23	54	2600
R. M1 (BA 4)	-36	22	53	3478	-28	24	65	138	-31	25	62	641	-56	15	40	126
L. S1 (BA 1, 2, 3, 5)	55	24	37	188	21	31	55	299	36	26	51	1706	40	25	48	4322
R. S1 (BA 1, 2, 3, 5)	-41	26	52	7711	-26	30	50	274	-28	32	62	620	-55	17	42	237
L. SMA (BA 6m, 24c)	5	6	55	2658	5	1	49	1782	4	11	54	2127	6	5	53	4030
R. SMA (BA 6m, 24c)	-7	5	49	4671	-5	2	50	2693	-4	4	50	2424	-7	2	50	3434
L. PM (BA 6l)	22	4	58	1537	55	0	28	1268	33	13	61	683	53	0	27	2328
R. PM (BA 6l)	-30	8	57	4068	-54	-2	33	2967	-37	3	54	936	-51	-1	42	847
L. PF (BA 46, 44)	58	-9	14	430	54	-7	14	758	57	-7	11	306	53	-6	14	1038
R. PF (BA 46, 44)	-57	-8	14	1136	-48	-44	19	1254	-61	-9	15	108	-58	-8	14	641
L. IPL (BA 40)	56	32	35	439	60	32	28	1762	37	31	40	130	54	27	27	1388
R. IPL (BA 40)	-39	40	53	1468	-39	47	54	438	-63	26	29	825	-60	33	23	155
L. Precuneus (BA 7)					18	65	47	746					27	49	50	240
R. Precuneus (BA 7)					-23	73	23	257								
L. Insula	41	-9	-5	125									45	0	11	267
R. Insula	-40	-3	9	1284	-40	-14	4	1216								
Subcortical region																
L. Thalamus	20	22	11	439					15	18	2	178				
R. Thalamus	-13	18	4	509	-14	18	3	748								
L. Putamen					27	16	6	129								
R. Putamen					-27	17	7	192								
L. Caudate nucleus													8	-14	6	793
R. Caudate nucleus													-11	-19	7	108
Cerebellum																
L. Cerebellum	21	60	-21	31921	20	61	-24	25943	18	63	-23	17424	11	60	-18	1574
R. Cerebellum	-17	60	-19	15302	-16	63	-23	15530	-13	59	-17	13331	-20	56	-20	7574

Abbreviations: L – left; R – right; BA – Brodmann area. The cluster volumes are measured in mm^3. Activated clusters were significant at $p < 0.05$ corrected for multiple comparisons.

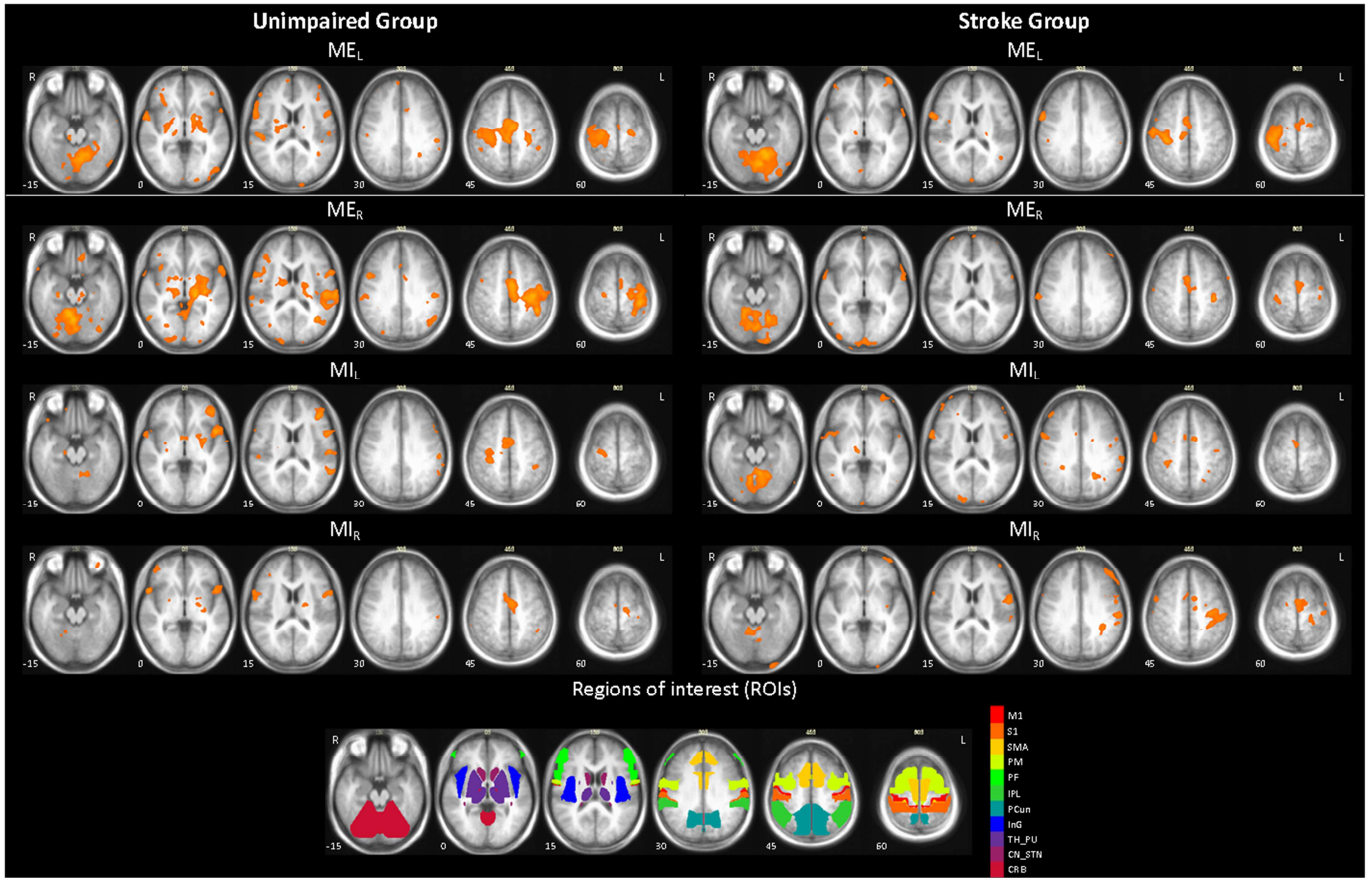


Figure 4.1 Brain activation maps obtained by group analysis for the unimpaired and stroke groups during motor execution or imagery. Activated clusters were significant at $p < 0.05$ corrected for multiple comparisons. The z-co-ordinate of each image is indicated in the axial views of the mask map (in mm).

Using Pillai's criterion, the composite dependent variate was significantly affected by stroke factor (Pillai's Trace=1.00, P=0.016). Univariate ANOVAs were conducted on each dependent measure separately to determine the locus of the statistically significant multivariate effect. Statistically significant group effects were observed for the cluster volumes of contralesional SMA (Vol_{stroke} =1963mm^3, $Vol_{unimpaired}$ =577mm^3, F[1,18]=4.53, P=0.047), and ipsilesional cerebellum (Vol_{stroke} =12050mm^3, $Vol_{unimpaired}$ =206mm^3, F[1,18]=9.27, P=0.007) during ME_R, and for the cluster volume of contralesional IPL (Vol_{stroke} =1730mm^3, $Vol_{unimpaired}$=329mm^3, F[1,18]=4.80, P=0.042) during ME_L (Figure 4.2). However, no statistically significant group effects were observed for the cluster volumes during MI tasks (Figure 4.3).

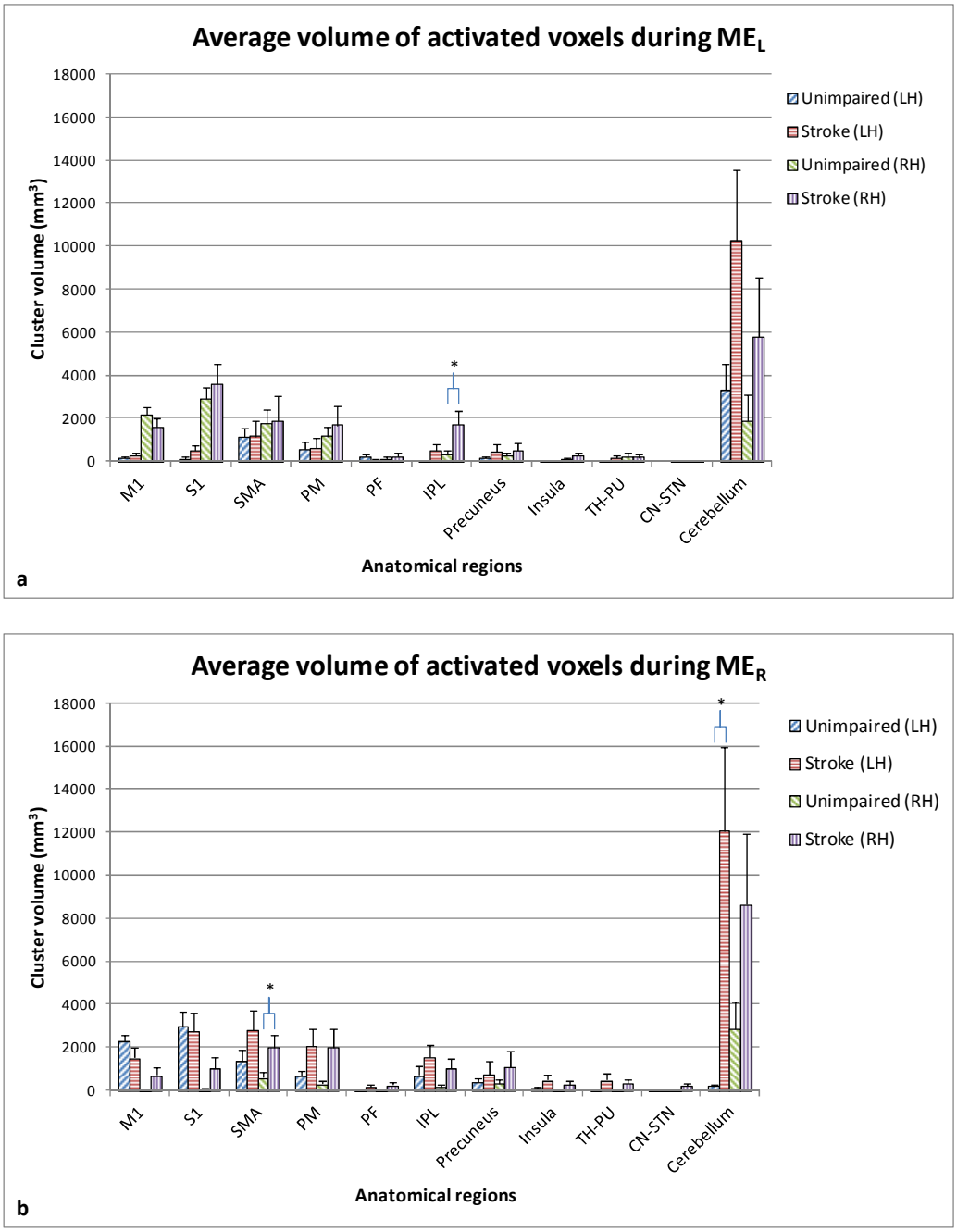
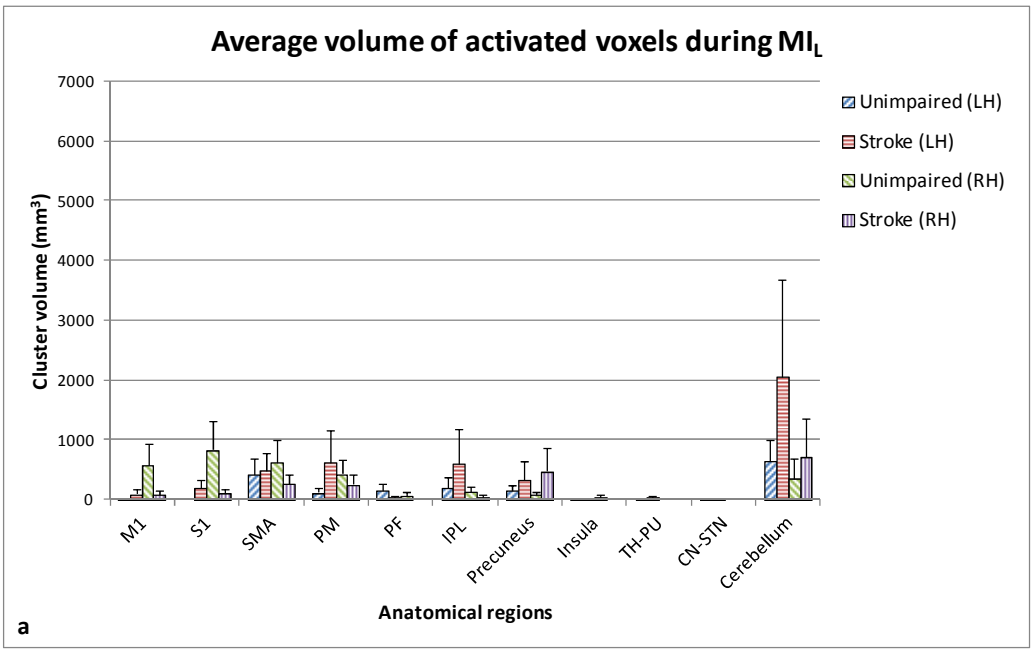


Figure 4.2 Average volume of activated voxels (SEM) during ME. LH, left hemisphere; RH, right hemisphere; LI, laterality index. The cluster volumes are measured in mm^3. Asterisks (*) indicate that significant differences between stroke and unimpaired groups were observed for the activation features at $p < 0.05$ by MANOVA.



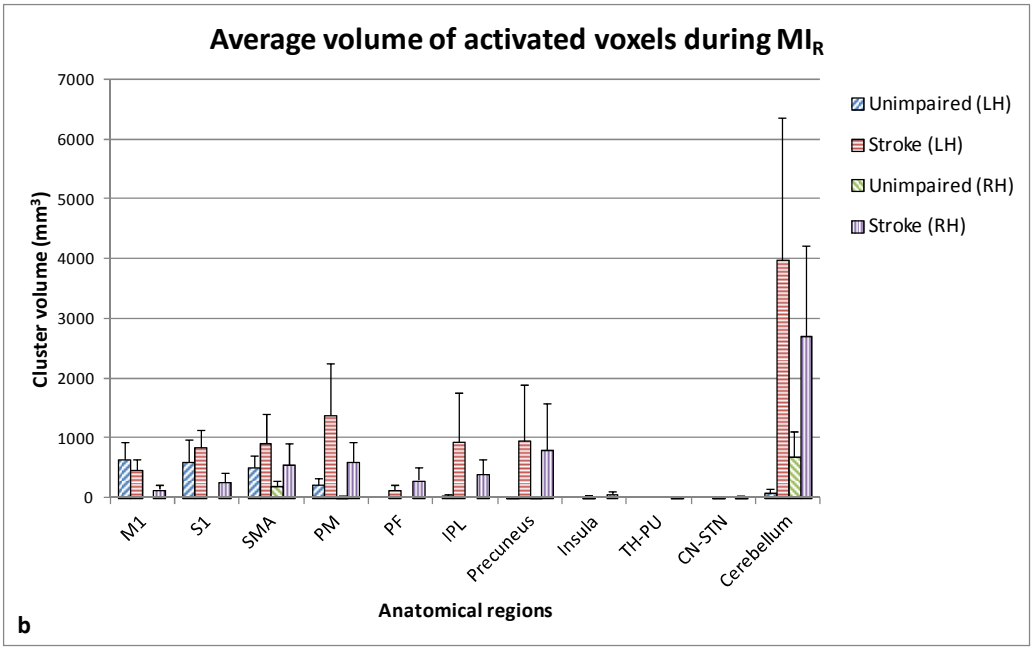


Figure 4.3 Average volume of activated voxels (SEM) during MI. LH, left hemisphere; RH, right hemisphere; LI, laterality index. The cluster volumes are measured in mm^3. There was no significant difference between stroke and unimpaired groups for the activation features at $p < 0.05$ by MANOVA.

Moreover, correlation analysis was performed on the data of ROI activation volume in order to explore the relationship between the activation patterns and the motor function outcomes. During MI_R, activation volumes in ipsilesional SMA showed significant positive correlation with the MSS_EF scores (rho=0.699, P=0.025). During ME_R, activation volumes in bilateral precuneus showed significant negative correlation with the FMA_WH scores (ipsilesional precuneus: rho=−0.652, P=0.041; contralesional precuneus: rho=−0.641, P=0.046) (Table 4.3).

Table 4.3 Correlations of quantitative measures with clinical assessment scores

Quantitative measures	Regions	Correlations	
		MSS_EF	FMA_WH
Cluster volume			
MI_R (Stroke)	L SMA	0.699*	0.539
ME_R (Stroke)	L Precuneus	-0.321	-0.652*
	R Precuneus	-0.346	-0.641*
Laterality index (LI)			
ME_R (Stroke)	PM	-0.646*	-0.434
	CN_STN	0.325	0.703*
Overlap index (OI)			
Stroke ME_R vs. Stroke MI_R	L SMA	0.669*	0.444
Stroke MI_R vs. Unimpaired MI_R	L SMA	0.702*	0.357
Stroke ME_R vs. Unimpaired ME_R	L Precuneus	-0.470	-0.713*
	R IPL	0.695*	0.135

Correlation values are Spearman's rho correlation coefficients. *P<0.05.

4.1.2 Lateralization of Brain Activation and Clinical Correlations

LI was calculated based on each subject's activation pattern obtained from individual analysis. The averages and SEMs of the LIs across the subjects were presented in Figure 4.4. From the results, large positive LI values (>0.5) were found in M1, S1, PM and IPL, and a large negative LI value (<−0.5) was found in cerebellum during both ME_L and ME_R for the unimpaired group (Figure 4.4a). Large positive LI values were additionally found in SMA and insula during ME_R. As for MI tasks, only M1, S1 and PM had a large positive LI value during MI_R, while none of

the regions showed great lateralization in brain activation during MI_L (Figure 4.4b). In stroke group, large positive LI values in M1, S1 and IPL, and a large negative LI value in cerebellum were also found during ME_L. During ME_R, only M1 and S1 had a large positive LI value. None of the regions showed great lateralization in brain activation during MI tasks (Figure 4.4b).

Statistically significant group effects were observed for the LI in M1 (LI_{stroke}=0.63, $LI_{unimpaired}$=0.99, F[1,18]=4.63, P=0.045), in SMA (LI_{stroke}=0.25, $LI_{unimpaired}$=0.70, F[1,18]=7.94, P=0.011), and in cerebellum (LI_{stroke}=0.10, $LI_{unimpaired}$=−0.72, F[1,18]=21.14, P=0.000) during ME_R, and for the LI in PF (LI_{stroke}=0.16, $LI_{unimpaired}$=−0.40, F[1,18]=5.35, P=0.033) and in CN-STN (LI_{stroke}=−0.27, $LI_{unimpaired}$=0.20, F[1,18]=5.92, P=0.026) during ME_L (Figure 4.4a). Significant effects were also observed for the LI in cerebellum (LI_{stroke}=0.25, $LI_{unimpaired}$=−0.37, F[1,18]=8.65, P=0.009) during MI_R, and for the LI in insula (LI_{stroke}=−0.20, $LI_{unimpaired}$=0.17, F[1,18]=4.44, P=0.049) during MI_L (Figure 4.4b). These results generally showed that less activation in the ipsilesional cortical regions relative to their contralesional regions and less hemispheric lateralization were observed in the stroke patients during these motor tasks.

From the correlation results (Table 4.3), there was no significant correlation found between the LI in the 11 regions and the motor scores during MI_R. During ME_R, a significant negative correlation was found between the LI in PM and the MSS_EF scores (rho=−0.646, P=0.044), and a significant positive correlation was found between the LI in CN-STN and the FMA_WH scores (rho=0.703, P=0.023).

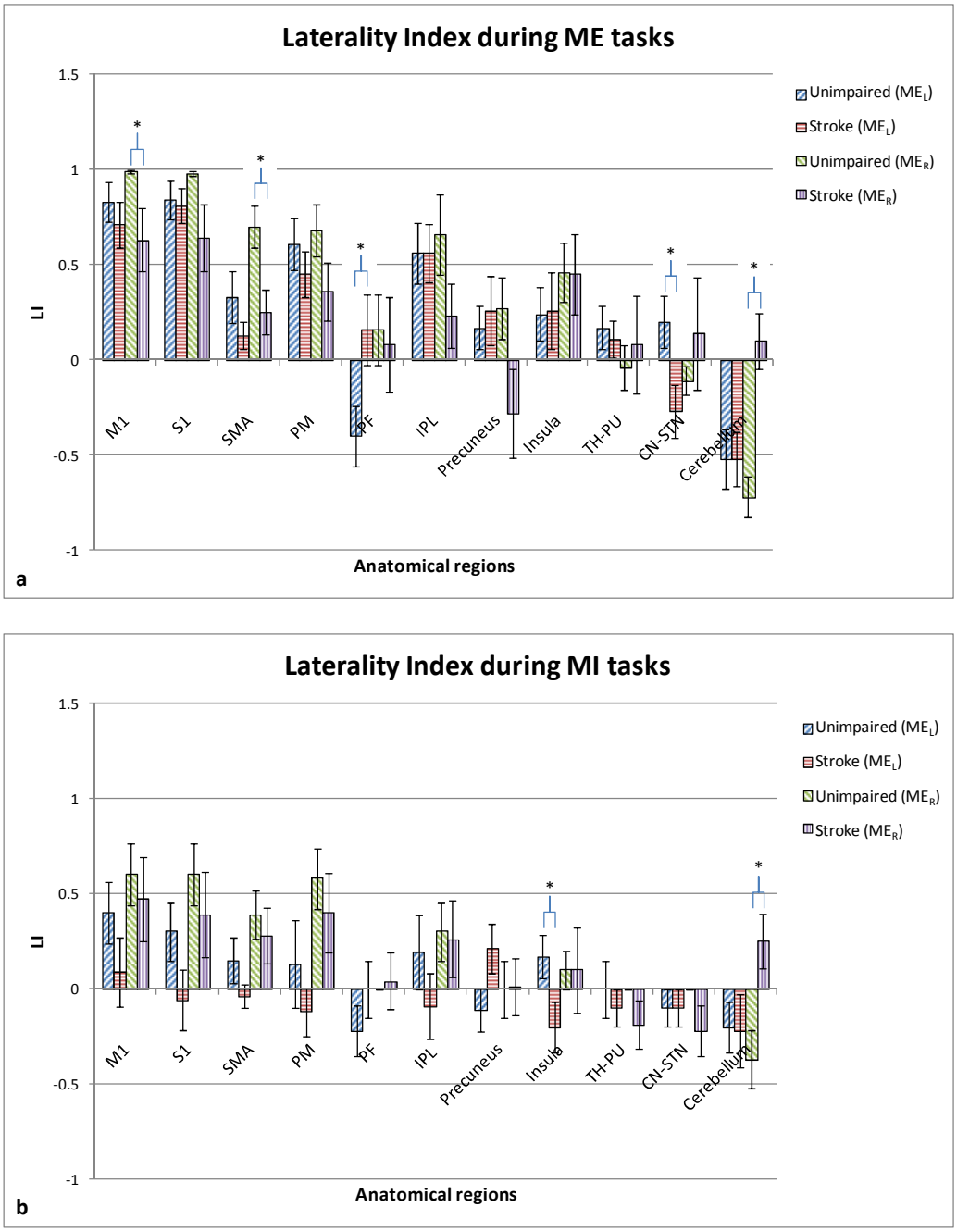


Figure 4.4 Average laterality indices (SEM) during ME and MI. LH, left hemisphere; RH, right hemisphere; LI, laterality index. Asterisks (*) indicate that significant differences between stroke and unimpaired groups were observed for the activation features at $p < 0.05$ by MANOVA.

4.1.3 Overlapping of Brain Activation and Clinical Correlations

In the comparison between ME_R and MI_R in stroke group (Figure 4.5a), the largest OI value was observed in ipsilesional primary motor and sensory areas (M1 and S1). There was a significant positive correlation between the OI in ipsilesional SMA and the MSS_EF scores (rho=0.669, *P*=0.034) (Table 4.3). In the comparison between MI_R from stroke group and MI_R from unimpaired group (Figure 4.5b), the largest OI value was observed in left/ipsilesional M1 and right/contralesional SMA. Significant positive correlations were found between the OI in left/ipsilesional SMA and the MSS_EF scores (rho=0.702, *P*=0.024). In the comparison between ME_R from stroke group and ME_R from unimpaired group (Figure 4.5c), the largest OI value was again found in left/ipsilesional M1 and S1, while in the non-primary motor related areas, bilateral SMA, left/ipsilesional PM and left/ipsilesional IPL were also found largely overlapped during ME_R between the two subject groups. From the correlation results, the IPL in right/contralesional hemisphere showed a significant positive correlation with the MSS_EF scores (rho=0.695, *P*=0.026), while the precuneus in left/ipsilesional hemisphere showed a significant negative correlation with the FMA_WH scores (rho=−0.713, *P*=0.021). In the comparison between ME_R and ME_L in unimpaired group (Figure 4.5d), the largest OI value was observed in bilateral SMA. In the comparison between ME_L from stroke group and ME_L from unimpaired group (Figure 4.5e), the largest OI values were observed in right/contralesional M1 and S1.

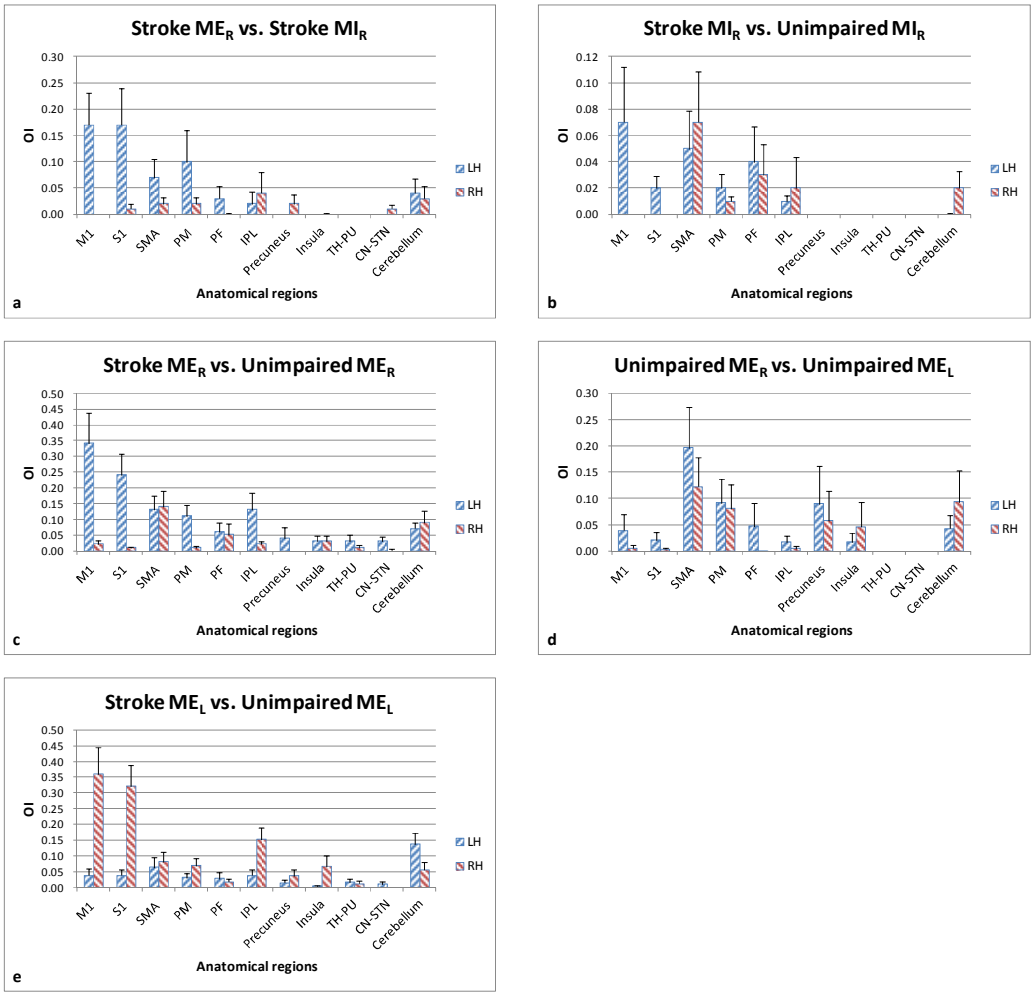


Figure 4.5 Average overlap indices (SEM) in the comparisons between different tasks/subject groups. LH, left hemisphere; RH, right hemisphere; OI, overlap index.

4.2 Study II: Study on Remodeling of Structural Connectivity and Its Correlation with Motor Impairments Using DTI

The nine patients (Table 3.2) were having moderate-to-severe upper-limb impairment (FMA score less than 44 out of 66). Their average scores were MSS: 25.5 (SD = 8.4) and FMA: 29.8 (SD = 11.0). The patients mainly exhibited lesions at the left posterior limb of internal capsule (PLIC), pallidum and putamen. Since all patients had left subcortical lesions, the left hemisphere is regarded as the ipsilesional hemisphere, whereas the right hemisphere is the contralesional hemisphere.

4.2.1 Effects of the CST's Structural Properties on Motor Outcomes

Since the structural integrity of the CST is a major determinant of motor recovery, ipsilesional and contralesional CSTs (fiber tracts passing through the sensorimotor cortex and the posterior limb of internal capsule) were illustrated in Figure 4.6 to show the integrity of the CSTs across an unimpaired subject and three stroke patients with various impairment severities. The results found that the ipsilesional CST showed a greater deterioration across the patients with increasing lesion volume (Figure 4.6). By contrast, the contralesional CST integrity of the patients was much more comparable to that of the unimpaired subject.

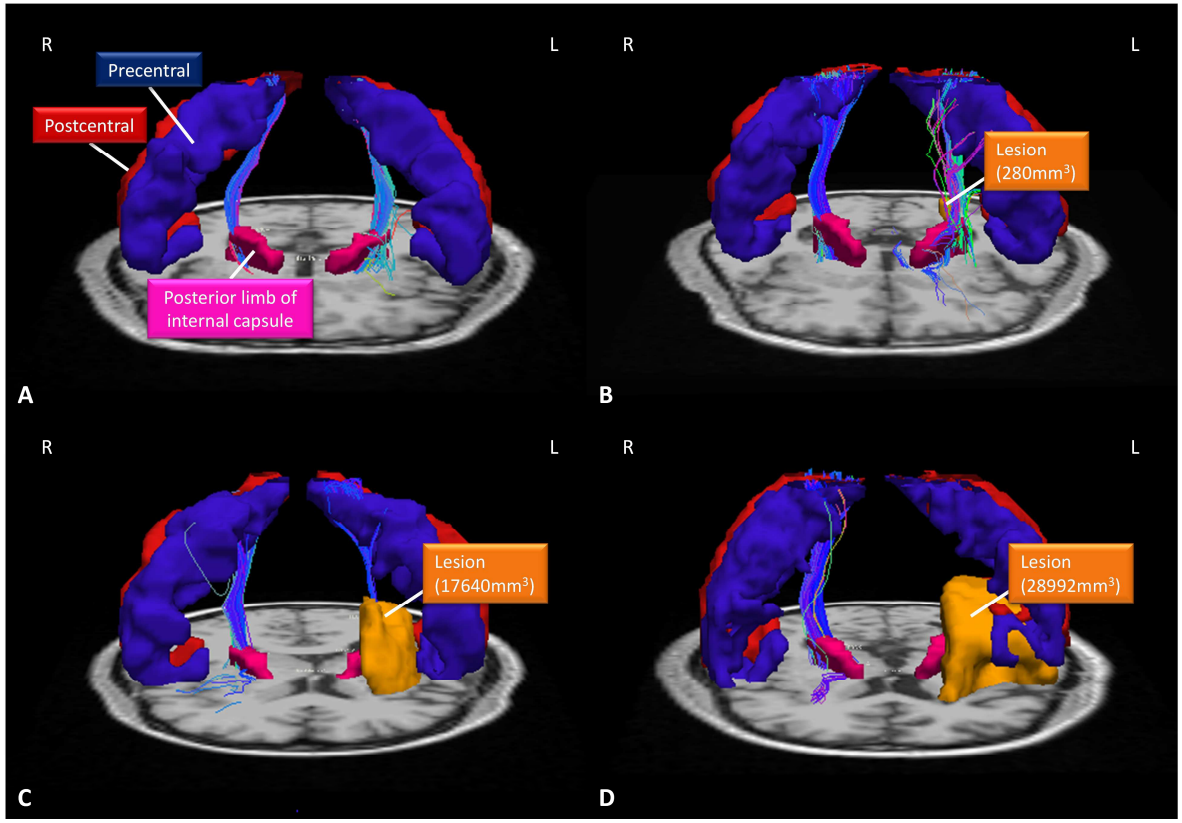


Figure 4.6 Illustration of the effect of lesion on the integrity of the corticospinal tracts (CSTs). (A) The CSTs of an unimpaired subject (U1), and (B-D) the CSTs of three stroke patients (S2, S5 and S9) with various motor impairment levels. The ipsilesional CST in stroke showed a greater deterioration across patients with increasing lesion volume, while the contralesional CST in stroke were relatively comparable to that of the unimpaired subject.

Besides, Pearson correlation analysis was done to investigate the effects of ipsilesional and contralesional CSTs' structural properties on the motor impairments. Significant positive correlation was found between FA of the ipsilesional CST and the MSS_EF score (r = 0.724, P = 0.028), indicating that smaller value in the FA of the ipsilesional CST was associated with poorer motor outcome in stroke patients. The FA of the contralesional CST showed a negative trend with the clinical scores, but not yet attained a significant level. On the other hand, there was no significant correlation between CW of the bilateral CSTs and the clinical assessment scores (Table 4.4). These results indicated that neither the FA of the contralesional CST nor the CW of the bilateral CSTs had a significant effect on the motor outcome of the stroke patients.

Table 4.4 Correlations between connectivity measures of CST and clinical assessment scores

Connectivity measures		Pearson Correlations			
		MSS_SM	MSS_EF	FMA_SE	FMA_WH
CW	L CST	0.194	0.354	0.357	0.250
	R CST	0.108	0.172	0.215	0.004
FA	L CST	0.581	0.724*	0.638	0.587
	R CST	-0.234	-0.197	-0.187	-0.056

*P<0.05.

4.2.2 Correlation Patterns Revealed by Pearson Correlation Analysis

In order to better understand the overall correlation pattern over the whole brain, all Pearson correlation coefficients of each brain region from both CS-motor and FA-motor correlations were added for the left and right cerebral hemispheres. Figure 4.7 illustrated the ranked distribution of the sum values, where the red end of the color bar indicated the regions having high ranked positive correlations, and the violet end of the color bar indicated the regions having high ranked negative correlations. The ipsilesional superior frontal cortex was found as the region with the

highest value of the summed coefficients (5.46), while the contralesional lingual gyrus was found as the region with the lowest value of the summed coefficients (-4.67). In general, the correlations between the connectivity measures (CS and FA) and the clinical scores were mainly found positive in the left (ipsilesional) regions, while the correlations between the connectivity measures and the clinical scores were mainly found negative in the right (contralesional) regions. There were ten regions which exhibited significant correlations, and they were labeled in the Figure 4.7 (8 ipsilesional regions showed significant positive correlations and 2 contralesional regions showed significant negative correlations).

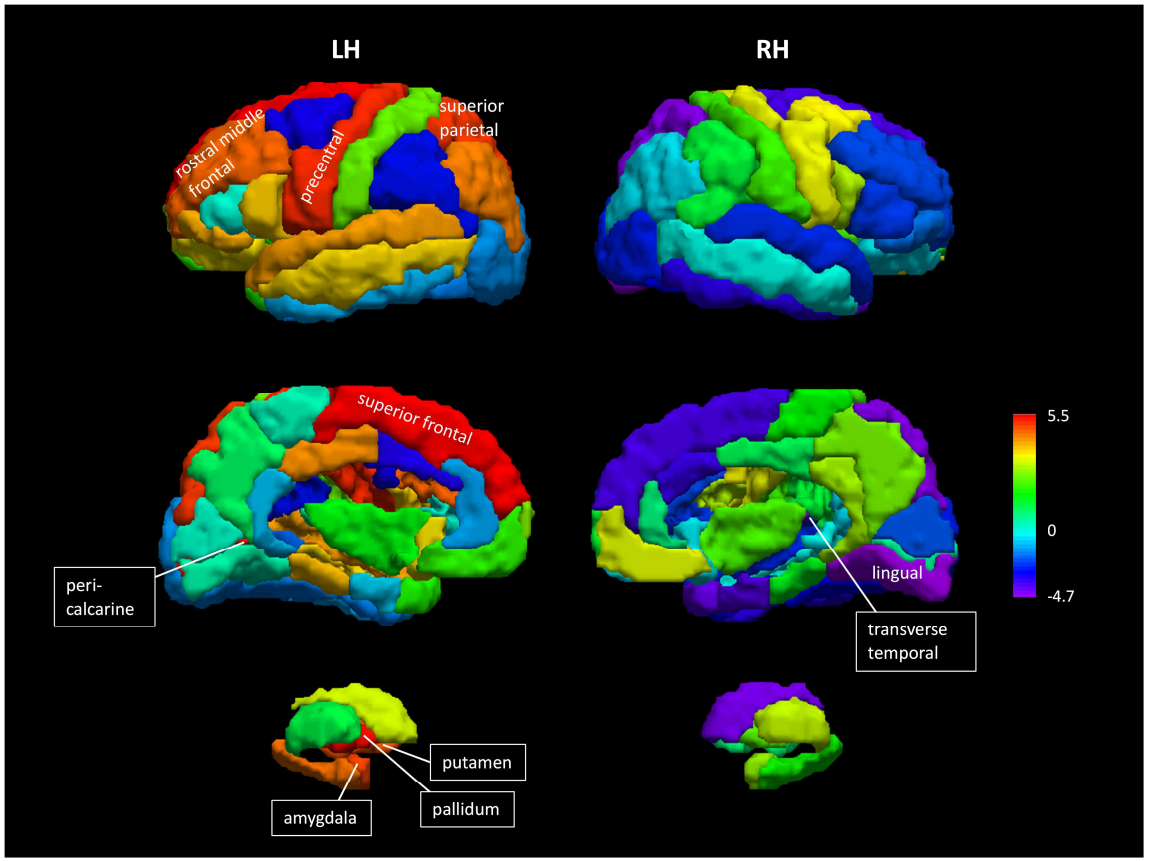


Figure 4.7 Ranking of the summed coefficients from both CS-motor and FA-motor correlations. The color bar indicates the range of sum values of all Pearson correlation coefficients of each brain region from both CS-motor and FA-motor correlations for left and right cerebral hemispheres. LH=left hemisphere, RH=right hemisphere.

From the correlation results (Table 4.5), all the significant correlations between the connectivity measures (CSs and regional FAs) of the left (ipsilesional) regions and the clinical scores were revealed as positive. Similar patterns were observed from the results of CS-motor and regional FA-motor correlation analyses. There were five regions which showed significant positive correlations, overlapped between the correlation analyses based on the two measures. They included the pericalcarine cortex, the superior parietal cortex, the putamen, the pallidum and the amygdala in the ipsilesional hemisphere. Besides these five regions, the CSs of the ipsilesional precentral gyrus and the ipsilesional rostral middle frontal cortex, and the regional FA of the ipsilesional superior frontal cortex showed additionally significant positive correlations with the clinical scores. From our results, both CSs and regional FAs demonstrated a parallel motor-correlated pattern, involving the subcortical areas (putamen, pallidum and amygdala), the parietal area (superior parietal cortex), the occipital area (pericalcarine cortex), and the frontal areas (CS: precentral gyrus and rostral middle frontal cortex; regional FA: superior frontal cortex). On the contrary, significant negative correlations were also found, rather unexpectedly, between the connectivity measures (CSs and regional FAs) of the right (contralesional) regions and the clinical scores. The involved regions were the lingual gyrus (based on CS-motor correlation) and the transverse temporal cortex (based on regional FA-motor correlation).

4.2.3 Correlation Patterns Revealed by Partial Correlation Analysis

The structural integrity of the CST is widely known as a major determinant of motor deficits, and therefore the effects of the CST's structural properties might play an important role in influencing the motor-correlated pattern over the whole brain. In order to find out whether the correlation patterns would be affected after removing

the effects of the CST's structural properties, partial correlation analysis was done to look at this issue. Based on the CS measures, after controlling for the CW of both ipsilesional and contralesional CST, there were five regions in which their CSs remained significantly correlated with the clinical scores. They were ipsilesional precentral gyrus ($r_{(PREC)(MSS_{SM})\cdot CST}$ = 0.805, P = 0.029), ipsilesional rostral middle frontal cortex ($r_{(RMF)(MSS_{SM})\cdot CST}$ = 0.759, P = 0.048), ipsilesional pallidum ($r_{(PAL)(MSS_{SM})\cdot CST}$ = 0.824, P = 0.022) and ipsilesional amygdala ($r_{(AMY)(MSS_{SM})\cdot CST}$ = 0.754, P = 0.050) which showed significant positive correlations with the MSS_SM score, and contralesional lingual gyrus ($r_{(LING)(MSS_{EF})\cdot CST}$ = -0.780, P = 0.038) which showed significant negative correlation with the MSS_EF score (Table 4.5). Based on the FA measures, after controlling for the FA of both ipsilesional and contralesional CST, there were still three regions in which their FAs remained significantly correlated with the clinical scores. They were ipsilesional pericalcarine cortex ($r_{(PCAL)(MSS_{EF})\cdot CST}$ = 0.763, P = 0.046), ipsilesional putamen ($r_{(PUT)(MSS_{SM})\cdot CST}$ = 0.787, P = 0.036) and ipsilesional pallidum ($r_{(PAL)(MSS_{SM})\cdot CST}$ = 0.848, P = 0.016) which showed significant positive correlations with the clinical scores (Table 4.5).

Table 4.5 Significant correlations between connectivity measures of brain regions and clinical assessment scores

Connectivity measures	Regions	(Pearson correlation) [Partial correlation]				MANOVAs	
		MSS_SM	MSS_EF	FMA_SE	FMA_WH	Wilks' Lambda	*P* value
CS	L pericalcarine cortex	(0.496)[0.500]	(0.711*)[0.681]	(0.592)[0.504]	(0.596)[0.617]	.221	.028
	L precentral gyrus	(0.768*)[0.805*]	(0.700*)[0.674]	(0.670*)[0.649]	(0.655)[0.622]		
	L rostral middle frontal cortex	(0.675*)[0.759*]	(0.411)[0.512]	(0.560)[0.669]	(0.412)[0.542]		
	L superior parietal cortex	(0.557)[0.551]	(0.690*)[0.692]	(0.452)[0.409]	(0.530)[0.561]		
	L putamen	(0.724*)[0.733]	(0.573)[0.595]	(0.554)[0.568]	(0.504)[0.536]		
	L pallidum	(0.638)[0.824*]	(0.643)[0.814*]	(0.631)[0.838*]	(0.711*)[0.805*]		
	L amygdala	(0.727*)[0.754*]	(0.709*)[0.684]	(0.588)[0.549]	(0.555)[0.503]		
	R lingual gyrus	(-0.594)[-0.576]	(-0.811**)[-0.780*]	(-0.675*)[-0.617]	(-0.722*)[-0.717]		
Regional FA	**L pericalcarine cortex**	(0.718*)[0.518]	(0.860**)[0.763*]	(0.662)[0.409]	(0.765*)[0.688]	.591	.490
	L superior frontal cortex	(0.785*)[0.607]	(0.767*)[0.445]	(0.754*)[0.506]	(0.704*)[0.519]		
	L superior parietal cortex	(0.775*)[0.611]	(0.803**)[0.596]	(0.632)[0.319]	(0.633)[0.399]		
	L putamen	(0.712*)[0.787*]	(0.584)[0.715]	(0.507)[0.542]	(0.519)[0.563]		
	L pallidum	(0.686*)[0.848*]	(0.691*)[0.849*]	(0.669*)[0.775*]	(0.768*)[0.825*]		
	L amygdala	(0.674*)[0.529]	(0.598)[0.286]	(0.482)[0.145]	(0.487)[0.184]		
	R transverse temporal cortex	(-0.653)[-0.403]	(-0.667*)[-0.301]	(-0.769*)[-0.574]	(-0.644)[-0.399]		

Those regions in which their CSs or FAs remained significantly correlated with the clinical scores after removing the effects of the bilateral CSTs' structural properties were highlighted in bold. *P<0.05, **P<0.01.

4.2.4 Differences Between Stroke Patients and Unimpaired Subjects

From the MANOVA results (Table 4.5), there was a statistically significant difference in the CSs of those significantly correlated regions between unimpaired subjects and stroke patients (Wilks' Lambda = 0.221, P = 0.028), but not found for the FAs (Wilks' Lambda = 0.591, P = 0.490). Followed up with univariate ANOVAs of the CSs, significant univariate group effects were found for 4 ipsilesional regions (Figure 4.8). They were precentral gyrus (CS_{stroke} = 7.10, $CS_{unimpaired}$ = 10.73, F[1,16] = 17.60, P = 0.001) and rostral middle frontal cortex (CS_{stroke} = 6.84, $CS_{unimpaired}$ = 8.19, F[1,16] = 9.33, P = 0.008) located in frontal lobe, and putamen (CS_{stroke} = 8.44, $CS_{unimpaired}$ = 11.65, F[1,16] = 4.50, P = 0.050) and amygdala (CS_{stroke} = 2.81, $CS_{unimpaired}$ = 4.11, F[1,16] = 6.44, P = 0.022) located in subcortical area. Interestingly, the CSs of these regions were found lower in stroke patients compared with the unimpaired subjects.

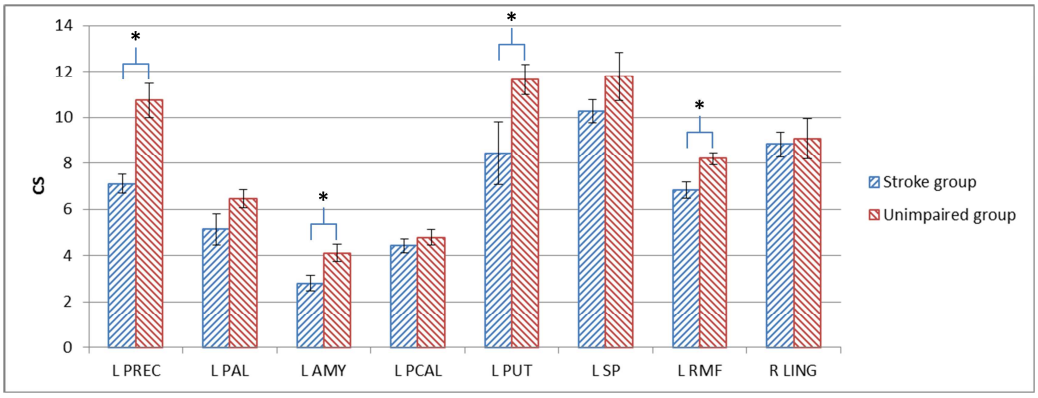


Figure 4.8 Comparison of CS between unimpaired subjects and stroke patients (*P<0.05)

Chapter 5 Discussion

5.1 Study I: Study on Neural Correlates of Motor Impairment During Motor Imagery and Motor Execution Using fMRI

This study aimed to investigate the activation patterns associated with ME and MI in stroke patients with left subcortical lesions, and to identify the clinical relevancy of activation features in between ME and MI motor tasks, and between stroke and unimpaired groups. Laterality index (LI) and overlap index (OI) were used to quantify hemispheric asymmetry and the spatial discrepancy of the cerebral activations, respectively. Our main findings showed that SMA played an important role during MI. A large OI was shown in the SMA from the comparison between ME_R and MI_R tasks using the affected wrist in the stroke patients, and from the comparison between stroke and unimpaired groups during MI_R (Figure 4.5). The OI values of the SMA showed significant positive correlations with the MSS_EF scores (Table 4.1). On the other hand, the activation features in precuneus showed significant negative correlation with the FMA_WH scores during ME_R in terms of the activation volume and the OI in the comparison between stroke and unimpaired groups. In non-primary motor areas, the activation feature in contralesional IPL showed a significant positive correlation with the MSS_EF scores indicated by the OI in the comparison between patient and unimpaired groups during ME_R, while the LI in PM showed a significant negative correlation with the MSS_EF scores.

5.1.1 Activation Features in the Supplementary Motor Area

From the brain activation patterns in stroke patients, our results showed a positive correlation that stroke patients with higher scores of forearm mobility had a larger

activation volume in the ipsilesional SMA, and exhibited more congruent functional neuroanatomy in the ipsilesional SMA associated with ME_R and MI_R. Among the activated brain areas during MI, the SMA is reported to be the most active area, which plays an important role in MI tasks and in the high-level motor control (Michelon *et al.*, 2006, Szameitat *et al.*, 2007), and is greatly involved in the preparation and readiness for action (Cunnington *et al.*, 1996). Such correlated activation features in ipsilesional SMA observed in our stroke patients reveals the importance of SMA to motor recovery after stroke. Moreover, increased overlap of activation in the ipsilesional SMA was shown in stroke patients with better motor skill, when comparing stroke and unimpaired groups during MI_R. In a previous study, contralateral control in SMA was demonstrated primarily in unimpaired subjects during MI, whereas stroke patients with severe hemiparesis displayed primarily ipsilateral control in SMA during the same condition (Kimberley *et al.*, 2006). Although these investigators did not perform any correlation analysis to study the relationship between the activation features and their patients' motor functions, their study supports a growing consensus that the best outcome is achieved when activating the brain in a pattern that most resembles the normal state (Cramer, 2004, Carey *et al.*, 2005). The correlated activity in SMA revealed in our stroke patients, which showed a relative reduction in contralesional SMA activity and increase in congruence of the functional neuroanatomy in ipsilesional SMA (more resembling the pattern observed in our unimpaired subjects), provides further converging evidence in support of the enhancement of ipsilesional activity in relation to better motor outcome.

5.1.2 Activation Features in the Precuneus

On the other hand, patients with poorer wrist and hand motor recovery showed

larger activation volume in bilateral precuneus, and exhibited more overlapping of activation in ipsilesional precuneus in the comparison between stroke and unimpaired groups during ME_R. The precuneus has recently been suggested by functional neuroimaging findings in healthy subjects that it is actively involved in a wide spectrum of highly integrated tasks, such as visuo-spatial imagery and retrieval of episodic memory and self-processing operations (Cavanna and Trimble, 2006). Moreover, precuneus and the nearby posteromedial areas are regarded as the functional cores of the default mode network (DMN), which show the highest resting metabolic rates with a characteristic of exhibiting transient reductions in the tonic activity during the involvement in non-self-referential goal-directed actions. Therefore, the precuneus is proposed to be engaged in the interconnected network of the neural substrates of self-consciousness, involving in self-related resting-state mental representations. This speculation is in line with the selective hypo-metabolism shown in the posteromedial cortex in a condition of altered consciousness, such as sleep, drug-induced anesthesia and vegetative conditions (Cavanna and Trimble, 2006). From a similar study, greater activation in bilateral precuneus was revealed in stroke patients during movements of their affected hand in comparison with healthy subjects (Lehericy *et al.,* 2004). This greater activity in the precuneus was likely attributed to the increased demand in attention required for the more complex motor tasks, such as bimanual movements (Wenderoth *et al.*, 2005), and was believed to provide compensation for the disorganized motor network (Wu and Hallett, 2005). The apparent compensatory effect in the precuneus seems to be more significant in patients with more severe motor impairments, and even during the motor execution using the distal portion of the upper limb (i.e. wrist), which may require more attention from the patients who had poor recovery of their wrist and hand functions to complete the task. This may, in turn, explain why our stroke

patients with poorer recovered wrist and hand functions exhibited more increased activity in the precuneus.

5.1.3 Activation Features in the Other Non-primary Motor-related Areas

In addition, patients with better motor recovery showed more overlapping of activation in contralesional IPL, from the comparison between stroke and unimpaired groups during motor execution using affected wrist (ME_R). In particular, the activation in PM is more lateralized to contralesional hemisphere in patients with better motor skill. Contralesional dorsal PM (PMd) is suggested to support residual motor function following stroke, since it is more active during movement of the affected hand in stroke patients, compared with healthy subjects in previous studies (Chollet *et al.*, 1991, Weiller *et al.*, 1992). The possible mechanism might be that motor-related activity in contralesional PMd may increase excitability in ipsilesional sensorimotor regions and, thereby, facilitate an increase in the gain of descending motor signals to the affected upper limb (Bestmann *et al.*, 2010). However, in contrast to the findings of previous studies, which showed those patients with more severe impairments exhibited greater contralesional activation in PMd (Johansen-Berg *et al.*, 2002, Bestmann *et al.*, 2010), our results demonstrated that patients with better motor recovery relied more on the activation in contralesional PM, relative to the ipsilesional. This discrepancy may in part be explained by differences in motor impairment levels of the stroke patients studied between our study and previous studies, since the patients included in their studies had mild motor impairment with feasible hand movements for their motor tasks (Johansen-Berg *et al.*, 2002, Bestmann *et al.*, 2010), whereas our patients had moderate-to-severe motor impairment with limited hand and wrist control. As increased cognitive demands of even simple movements is found in stroke patients (Sharma *et al.*, 2009), and the

increased attention to simple movements would, in turn, increase activity in PM and PF areas (Rowe *et al.*, 2002), our patients with better recovery might recruit more the PM activation and even in the contralesional hemisphere to support the residual motor function. In our patients with more severe hemiparesis, their motor attentional network might be disrupted more severely, leading to less recruitment of PM activity required for the motor attention and/or selection.

5.2 Study II: Study on Remodeling of Structural Connectivity and Its Correlation with Motor Impairments Using DTI

This study aimed to examine the post-stroke remodeling of structural connectivity by studying the relationships between the connectivity measures and the motor impairments in stroke patients with left subcortical lesions. Three main connectivity measures (FA, CW and CS) were used to evaluate the post-stroke structural changes. From our results, significant positive CS-motor correlations were found in five common regions (ipsilesional precentral gyrus, ipsilesional rostral middle frontal cortex, ipsilesional pallidum, ipsilesional amygdala and contralesional lingual gyrus) before and after controlling for the bilateral CSTs' connectivity property. Meanwhile, the CSs of three of these regions (precentral gyrus, rostral middle frontal cortex and amygdala) were also found significantly higher in unimpaired subjects compared with those in stroke patients, indicating that these three regions exhibited weaker connections to the rest of the network for the stroke patients.

5.2.1 Connectivity Features in the Ipsilesional Frontal Areas

Significant positive CS-motor correlations were found in the ipsilesional precentral gyrus and the ipsilesional rostral middle frontal cortex before and after controlling for the CW of the bilateral CST. Moreover, their CSs were also found

significantly lower in stroke patients compared with those in unimpaired subjects. On the other hand, from the Pearson correlation results, significant positive regional FA-motor correlations were also found in the ipsilesional superior frontal cortex. From the results, higher values in either the CS or the regional FA of these ipsilesional frontal areas were related to better motor outcomes in the stroke patients. The connections to the rest of the network were additionally found weaker in the precentral gyrus and the rostral middle frontal cortex for the stroke patients as compared with the unimpaired subjects. These ipsilesional frontal areas have been well documented to play important role in determining the motor performance. The primary motor cortex (precentral gyrus) and the frontal association areas (rostral middle frontal cortex and superior frontal cortex) are reported as the key neural substrates primarily for motor execution and planning respectively. Our observations from this study are in line with the existing knowledge established from prior studies.

5.2.2 Connectivity Features in the Ipsilesional Subcortical Areas

From the Pearson correlation results, significant positive CS-motor and regional FA-motor correlations were found in the ipsilesional lentiform nucleus (which is composed of the putamen and the pallidum). After controlling for the FA of the bilateral CST, the regional FA of the lentiform nucleus remained significantly correlated with the clinical scores. Moreover, the CS of the putamen was also found significantly lower in stroke patients compared with those in unimpaired subjects. Given the results, higher values in the CS and the regional FA of the ipsilesional lentiform nucleus were related to better motor outcomes in the stroke patients, and the connections to the rest of the network were found weaker in this region for the stroke patients as compared with the unimpaired subjects. Since all our stroke patients were having left subcortical lesions (mainly exhibiting lesions at the

posterior limb of internal capsule, pallidum and putamen), the lentiform nucleus which is composed of the putamen and pallidum would definitely bear the brunt of the brain damage induced by stroke.

Another region particularly worth mentioning is the amygdala. From the Pearson correlation results, significant positive CS-motor and regional FA-motor correlations were found in the ipsilesional amygdala. After controlling for the CW of the bilateral CST, the CS of the amygdala remained significantly correlated with the clinical scores. Moreover, its CS was also found significantly lower in stroke patients compared with those in unimpaired subjects. Consistently, higher values in the CS and the regional FA of the ipsilesional amygdala were related to better motor outcomes in the stroke patients, and the connections to the rest of the network were found weaker in this region for the stroke patients as compared with the unimpaired subjects. It is commonly known that the amygdala has a main function in the processing of memory and emotional reactions. However, a recent study suggested that the amygdala can also affect the execution of continuing movements by regulating brain circuits involved in motor control, so as to offer rapid and adaptive changes in current behavior (Sagaspe *et al.*, 2011). The amygdala has been reported acting as a primary neural substrate to orchestrate a quick coordination between emotional inputs and motor output processes (Armony *et al.*, 1997), given that it was shown to be greatly involved in promoting protective motor reactions (such as stop moving or defensive behavior) in the contexts with significant emotional involvement (LeDoux, 2000). Besides, a significant interaction between emotion and motor inhibition has been observed in the amygdala, suggesting that the emotional processing in the amygdala might facilitate motor inhibition, and such inhibition might be further strengthened when stop signals are related to threat cues (LeDoux, 2000).

5.2.3 Connectivity Features in the Contralesional Occipitotemporal Areas

On the contrary, the contralesional lingual gyrus and the contralesional transverse temporal cortex, which locate in the occipitotemporal lobe, were found showing significant negative correlations between their connectivity measures and the clinical scores. These results indicated that higher values in these connectivity measures of the contralesional occipitotemporal regions were found in relation to poorer motor outcomes in our stroke patients. Although the role of these regions in motor recovery is still uncertain, our results seem to have parallels with the previous findings from a number of functional neuroimaging studies, in which increased activity in the contralesional hemisphere during paretic limb movement was shown to correlate with poorer motor outcome. On the other hand, more findings have also been documented in some studies, demonstrating the involvement of the lingual gyrus and the temporal cortex in the processing of motion information (Servos *et al.*, 2002). Damage to the fusiform and lingual gyri that extends to lateroventral areas of the temporal lobe is believed to be the root of inducing motion perceptual incompetence (Cowey and Vaina, 2000). Furthermore, given more evidences from the functional neuroimaging (Bonda *et al.*, 1996, Howard *et al.*, 1996, Servos *et al.*, 2002), neuropsychological (Vaina *et al.*, 1990) and single-cell recording (Oram and Perrett, 1994) literature, the superior temporal cortex and the lingual gyrus are revealed playing a critical role in motion perception: The lingual gyrus may be responsible for the processing of motion information and the acquirement of global form information, while the superior temporal cortex may be responsible for the derivation of social meaning from this motion information (Servos *et al.*, 2002). Therefore, the lingual gyrus and the temporal cortex are suggested to be involved in the higher-order processing of motion information (Servos *et al.*, 2002). These findings may

complement our results, suggesting a possibility that poorer-recovered patients may require more involvement from contralesional occipitotemporal regions for motion information processing, in order to properly guide their motor behavior.

Chapter 6 Conclusion

The overall objective of this study is to find out how the functional outcomes after stroke be related to the changes in functional networks responsible for motor execution and motor imagery, and the changes in physical structure of the brain. This study is split into two parts to look into the features of brain reorganization and remodeling in patients after stroke from both functional and structural perspectives, and to correlate these features with their functional outcomes. Part I of this study explores the neural correlates of motor impairment during motor imagery and motor execution using fMRI, while part II of this study describes the remodeling of structural connectivity and its correlation with motor impairment using DTI.

In study I, LI and OI were used to quantify hemispheric asymmetry and the spatial discrepancy of the cerebral activations, respectively. Our main findings showed that SMA played an important role during MI. A large OI was shown in the SMA from the comparison between ME_R and MI_R tasks using the affected wrist in the stroke patients, and from the comparison between stroke and unimpaired groups during MI_R. The OI values of the SMA showed significant positive correlations with the MSS_EF scores. On the other hand, the activation features in precuneus showed significant negative correlation with the FMA_WH scores during ME_R in terms of the activation volume and the OI in the comparison between stroke and unimpaired groups. In non-primary motor areas, the activation feature in contralesional IPL showed a significant positive correlation with the MSS_EF scores indicated by the OI in the comparison between patient and unimpaired groups during ME_R, while the LI in PM showed a significant negative correlation with the MSS_EF scores. From the results, the non-primary motor-related areas, including the SMA during MI of affected limb,

and the precuneus, PM and IPL during ME of affected limb, seem to play important roles in regaining motor function in our stroke patients with moderate-to-severe motor impairment. In particular, more reliance on the ipsilesional SMA was seen in patients with better motor recovery when they were performing MI using their affected wrist. Further longitudinal studies are recommended to identify the neural substrates targeted by MI treatments, and to affirm their role in contributing to motor recovery.

In study II, three main connectivity measures (FA, CW and CS) were used to evaluate the post-stroke structural changes. From our results, significant positive CS-motor correlations were found in five common regions (ipsilesional precentral gyrus, ipsilesional rostral middle frontal cortex, ipsilesional pallidum, ipsilesional amygdala and contralesional lingual gyrus) before and after controlling for the bilateral CSTs' connectivity property. Meanwhile, the CSs of three of these regions (precentral gyrus, rostral middle frontal cortex and amygdala) were also found significantly higher in unimpaired subjects compared with those in stroke patients, indicating that these three regions exhibited weaker connections to the rest of the network for the stroke patients. From the results, we are able to demonstrate statistically significant alterations of neural connectivity in both the ipsilesional and contralesional hemispheres in our stroke patients compared to the unimpaired subjects, to confirm the work of previous studies, and provide more information of the alterations in connectivity profile over the whole brain. Our findings could pave the way for longitudinal studies in larger patient groups, to elucidate the role of the involved regions in the process of motor recovery.

Appendix 1 Study Inclusion and Exclusion Criteria

Inclusion Criteria – Chronic Stroke Patients:

- Between the age of 18 and 85
- First-ever stroke with time elapsed after stroke more than 12 months
- Substantial unilateral motor impairment

Exclusion Criteria – Chronic Stroke Patients:

- History of alcohol or drug abuse
- History of epilepsy
- Cerebellar lesions
- More than one stroke in the middle cerebral artery territory
- Bilateral motor impairment
- Uncontrolled medical problems (e.g. cardiovascular disease expressed as uncontrolled arrhythmias, shortness of breath, or overt signs of severe peripheral edema at the initial neurological exam, severe rheumatoid arthritis, arthritic joint deformity, active cancer or renal disease) or psychiatric problems
- Serious cognitive deficits that would prevent their ability to give informed consent and/or perform the study tasks
- Pregnancy
- MRI contraindications
- Comprehensive Aphasia

Inclusion Criteria – Healthy Volunteers:

- Between the age of 18 and 85

Exclusion Criteria – Healthy Volunteers:

- Inability to perform study tasks
- Uncontrolled medical problems
- History of epilepsy
- History of alcohol or drug abuse or psychiatric illness
- Serious cognitive deficits that would prevent their ability to give informed consent and/or perform the study tasks
- Pregnancy
- MRI contraindications

Appendix 2 Motor Status Assessment

Upper Extremity Motor
Movement scale – Shoulder/Elbow
0 = no volitional movement or no contraction
1- = contraction or patient initiating first few degrees of movement
1 = performs partly/incomplete or uncontrolled motion
1+ = lacking last few degrees of motion
2- = completes full range, decreased control or timing
2 = performs faultlessly (complete, controlled motion)
Place and hold (shoulder: 1B, 2B, 3B, 4B, 5B; elbow: 2B – 0 or 1)

Seated active range of motion (check wheelchair positioning)
Shoulder Movement

1. A. Shoulder flexion to 90°, elbow 0°, forearm neutral
   Deltoid, Rotator Cuff
   B. If placed, can position be held? (0-1)
   Deltoid, Rotator Cuff
2. A. Shoulder abduction to 90°, elbow 0°, forearm pronated
   Deltoid, Rotator Cuff
   B. If placed, can position be held? (0-1)
   Deltoid, Rotator Cuff
3. A. Shoulder flex 90°-150°, elbow 0°
   Deltoid, Rotator Cuff
   B. If placed, can position be held? (0-1)
   Deltoid, Rotator Cuff
4. A. Touch top of head
   Deltoid, Rotator Cuff, Biceps Brachii, Triceps Brachii
   B. If placed, can position be held? (0-1)
   Deltoid, Rotator Cuff, Biceps Brachii, Triceps Brachii
5. A. Touch small of back
   Subscapularis, Pectoralis Major, Latissimus Dorsi, Teres Major, Deltoid, Upper Trapezius
   B. If placed, can position be held? (0-1)
   Subscapularis, Pectoralis Major, Latissimus Dorsi, Teres Major
6. Scapular elevation
   Upper Trapezius, Levator Scapulae
7. Protraction/retraction of the scapula arm supported on table or lap
   Serratus Anterior, Rhomboids Major, Minor, Middle Trapezius
8. A. Shoulder flex 0°-30°, elbow starts at 90°
   Deltoid, Supraspinatus
   B. Shoulder to 30° extension with elbow flex, forearm supported on table
   Latissimus Dorsi, Teres Major, Posterior Deltoid
9. A. Shoulder 0°, elbow 90°, shoulder internal rotation to abdomen
   Subscapularis, Pectoralis Major, Lattisimus Dorsi, Teres Major
   B. Shoulder 0°, elbow 90°, shoulder external rotation
   Infraspinatus, Teres Minor
10. Touch opposite knee
    Pectoralis Major, Triceps Brachii, Pronator Group

Elbow/Forearm

1. A. Forearm pronation from mid-position shoulder 0°, elbow 90°
   Pronator Group
   B. Forearm supination from mid-position shoulder 0°, elbow 90°
   Biceps Brachii, Supinator
2. A. Elbow 0°, fully flex
   Biceps Brachii, Brachialis, Brachioradialis
   B. If placed, can position be held? (0-1)
   Biceps Brachii, Brachialis, Brachioradialis
3. Full elbow flexion, extend to 0° (gravity eliminated or against gravity)
   Triceps Brachii
4. Touch opposite shoulder
   Deltoid, Rotator Cuff, Pectoralis Major, Biceps

Total movement scale (MSS) (Max 40)

Appendix 3 Fugl-Meyer Assessment

Upper Extremity Motor

Position	Maximum Possible Score	Test	Scoring Criteria
Sitting	4	*I. Reflexes* • Biceps • Triceps	• No reflex activity can be elicited = 0 • Reflex activity can be elicited = 2
	12	*II. Flexor synergy* • Elevation • Shoulder retraction • Abduction (at least 90°) • External rotation • Elbow flexion • Forearm supination	• Cannot be performed at all = 0 • Performed partly = 1 • Performed faultlessly = 2
	6	*III. Extensor Synergy* • Shoulder adduction/internal rotation • Elbow extension • Forearm pronation	• Cannot be performed at all = 0 • Performed partly = 1 • Performed faultlessly = 2
	6	*IV. Movement Combining* Synergies a. Hand to lumbar spine	• No specific hand action performed = 0 • Hand must pass anterior superior iliac spine = 1 • Action is performed faultlessly = 2
		b. Shoulder flexion to 90° elbow at 0°	• Arm is immediately abducted or elbow flexes at start of motion = 0 • Abduction or elbow flexion occurs in later phase of motion = 1 • Faultless motion = 2
		c. Pronation/supination of forearm with elbow at 90° and shoulder at 0°	• Correct position of shoulder and elbow cannot be attained, and/or pronation or supination cannot be performed at all = 0 • Active pronation or supination can be performed even within a limited range of motion, and at the same time the shoulder and elbow are correctly positioned. = 1 • Complete pronation and supination with correct positions at elbow and shoulder = 2
	6	*V. Movement Out of Synergy* a. Shoulder abduction to 90° elbow at 0° and forearm pronated	• Initial elbow flexion occurs or any deviation from pronated forearm occurs = 0 • Motion can be performed partly,

			or if during motion, elbow is flexed or forearm cannot be kept in pronation = 1 • Faultless motion = 2
		b. Shoulder flexion to 90° elbow at 0°	• Initial flexion of elbow or shoulder abduction occurs = 0 • Elbow flexion or shoulder abduction occurs during shoulder flexion = 1 • Faultless motion = 2
		c. Pronation/supination of forearm with elbow at 90° and shoulder at 0°	• Supination and pronation cannot be performed at all or elbow and shoulder position cannot be attained = 0 • Elbow and shoulder properly positioned and pronation and supination performed in a limited range = 1 • Faultless motion = 2
	2	*VI. Normal Reflex Activity* • Biceps and/or finger flexors and triceps (This component is included only if the patient has a score of 6 for component V)	• At least 2 of the 3 phasic reflexes are markedly hyperactive = 0 • One reflex markedly hyperactive or at least 2 reflexes are lively = 1 • No more than one reflex is lively and none are hyperactive = 2
Wrist	10	*VII.* a. Stability, elbow at 90°, shoulder at 0°	• Patient cannot dorsiflex wrist to required 15° = 0 • Dorsiflexion is accomplished, but no resistance is taken = 1 • Position can be maintained with some (slight) resistance = 2
		b. Flexion/extension, elbow at 90°, shoulder at 0°	• Volitional movement does not occur = 0 • Patient cannot actively move the wrist joint throughout the total ROM = 1 • Faultless, smooth movement = 2
		c. Stability, elbow at 0°, shoulder at 30°	• Patient cannot dorsiflex wrist to required 15° = 0 • Dorsiflexion is accomplished, but no resistance is taken = 1 • Position can be maintained with some (slight) resistance = 2
		d. Flexion/extension, elbow at 0°, shoulder at 30°	• Volitional movement does not occur = 0 • Patient cannot actively move the wrist joint throughout the total ROM = 1 • Faultless, smooth movement = 2
		e. Circumduction	• Cannot be performed = 0 • Jerky motion or incomplete circumduction = 1

			• Complete motion with smoothness = 2
Hand	14	*VIII.* a. Finger mass flexion	• No flexion occurs = 0 • Some flexion, but not full motion = 1 • Complete active flexion (compared with unaffected hand) = 2
		b. Finger mass extension	• No extension occurs = 0 • Patient can release an active mass flexion grasp = 1 • Full active extension = 2
		c. Grasp #1 - MP joints extended and PIPS & DIPS are flexed. Grasp is tested against resistance	• Required position cannot be acquired = 0 • Grasp is weak = 1 • Grasp can be maintained against relatively great resistance = 2
		d. Grasp #2 - Patient is instructed to adduct thumb, 1st carpometacarpophalangeal and interphalangeal joint at 0°	• Function cannot be performed = 0 • Scrap of paper interposed between the thumb and index finger can be kept in place, but not against a slight tug = 1 • Paper is held firmly against a tug = 2
		e. Grasp #3 - Patient opposes the thumb pad against the pad of index finger. A pencil is interposed	
		f. Grasp #4 - The patient should grasp a cylinder- shaped object (small can), the volar surface of the 1st and 2nd finger against each other	
		g. Grasp #5 - A spherical grasp	
	6	*IX. Coordination/Speed - Finger-to-nose (five repetitions in rapid succession)* a. Tremor	• Marked tremor = 0 • Slight tremor = 1 • No tremor = 2
		b. Dysmetria	• Pronounced or unsystematic dysmetria = 0 • Slight or systematic dysmetria = 1 • No dysmetria = 2
		c. Speed	• Activity is more than 6 seconds longer than unaffected hand = 0 • 2 to 5 seconds longer than unaffected hand = 1 • Less than 2 seconds difference = 2
	66	Total Upper Extremity Motor Score	

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