Factors influencing bilateral interactions in the human motor cortex: investigating transcallosal sensorimotor networks

by

Robyn Ibey

A thesis

presented to the University of Waterloo

in fulfillment of the

thesis requirement for the degree of

Doctor of Philosophy

in

Kinesiology

Waterloo, Ontario, Canada, 2017

Examining Committee Membership

The following served on the Examining Committee for this thesis. The decision of the Examining Committee is by majority vote.

External Examiner	Dr. David F. Collins, PhD Professor University of Alberta
Supervisor	Dr. W. Richard Staines, PhD Professor University of Waterloo
Internal Member	Dr. William McIlroy, PhD Professor University of Waterloo
Internal-external Member	Dr. Britt Anderson, MD, PhD Associate Professor University of Waterloo
Committee Member	Dr. Aimee J. Nelson, PhD Associate Professor McMaster University

Author's Declaration

I hereby declare that I am the sole author of this thesis. This is a true copy of the thesis, including any required final revisions, as accepted by my examiners.

I understand that my thesis may be made electronically available to the public.

Abstract

All daily activities require the precise interaction and coordination of several brain regions to facilitate purposeful movements of the upper limbs. The mechanisms responsible for cross facilitation between the primary motor cortices are poorly understood and are important in understanding the neurophysiology of everyday upper limb movements and customizing task- and deficit- specific rehabilitation protocols following brain injury. Researchers have demonstrated activity-dependent changes in the primary motor cortex (M1) ipsilateral to the moving limb; however, the characteristics mediating this interaction between the hemispheres are not well understood. The aim of this thesis is to examine sensorimotor manipulations that modulate excitability of the resting M1 and determine the neural substrates that may be mediating these interactions. This thesis is comprised of 4 studies and we investigated corticomotor excitability changes of a resting upper limb muscle during (1) rhythmical movement at increasing force requirements, (2) rhythmical movement at increasing force requirements with the addition of sensory input (3) interhemispheric interactions and somatotopic relationships, and (4) convergence of multiple effectors. This dissertation identifies various sensorimotor manipulations that increase excitability of M1 and further informs the neurophysiological mechanisms that may be responsible for these interactions. Understanding the extent to which these mechanisms mediate activity between the upper limbs has implications in bimanual coordination and ultimately experience-dependent plasticity. The findings in this thesis have important applications for improving motor recovery with rehabilitation interventions post brain injury.

Acknowledgements

This is very nostalgic for me, a decade ago I graduated from my undergraduate degree at University of Waterloo and received "Oh, the Places You'll Go!" by Dr. Seuss. This book is about the Great Balancing Act of life itself with all the ups and downs we are presented with along the way. Dr. Seuss' book encourages us to find the success that lies within us as we embark on our journey. The journey to the end of my PhD proved to be rocky at times and I faced some of the biggest obstacles of my life thus far, but I am so fortunate to have an amazing network of people in my life – I couldn't have gotten to this point without all of you. I wish I could individually thank you all for the influence you have had, but I simply cannot do you justice in this small space. Just know I am forever grateful to have each of you in my life!

"Believe in yourself. You gain strength, courage, and confidence by every experience in which you stop to look fear in the face... You must do that which you think you cannot do... ***The future belongs to those who believe in the beauty of their dreams****."*
– Eleanor Roosevelt

One thing I have never done is quit. I am a fighter, I don't take no for an answer and I am stubborn. That being said, I was very close to walking away from this PhD. I had lost my passion, my strength, my drive, and my determination. Only a handful of people knew this and they believed in me and helped me to find myself again – I am eternally grateful to each of you:

To Sue, you helped me to finally find my way and rediscover my passion for pursuing my PhD. Everything started to become clear with your guidance and I will never forget the influence you had.

To my best friend Vincent who lives steps away, thank you for *always* being there. The countless coffee dates, dinners and trips to Home Sense, I could not have gotten to this point without you. You are one in a million and I am so fortunate to have you in my life.

To Rich, there is not enough space to thank you for everything you did for me. You are an amazing advisor, mentor and friend. You were patient through this crazy journey and you never gave up on me. Somehow you always knew just what I needed and you let me find my way. Thank you for everything – your belief and trust in me means more than you know.

To my family (Mom, Dad, Eric, Kristen, Kelly & Uncle Den), your love, support and encouragement has been the one constant my whole life. You have always been there for me and I can't imagine my life without each of you. You are my rocks. Mom & Dad I cannot remember a time in my life where you didn't encourage me to pursue my dreams. When asked "*what should I be when I grow up?*" you would say "*whatever you choose, we will be happy as long as you are happy.*" You have been there for me through it all and I can't imagine having better parents.

Dedication

In loving memory of my Baba Vickie: *your neurological injury first inspired my pursuit of this PhD, and ultimately provided me the strength and resilience to conquer and overcome my own brain injury to complete this journey*.

Table of Contents

Table of Contents *(cont'd)*

Table of Contents *(cont'd)*

Table of Contents *(cont'd)*

List of Figures

Chapter 1 – Introduction

Chapter 2 – Corticomotor excitability changes seen in the resting forearm during contralateral rhythmical movement and force manipulations: a TMS study

Chapter 3 – Convergence of transcallosal sensorimotor information: effect of rhythmical movement, force and vibration on the contralateral resting limb

Chapter 4 – Enhancing excitability of the primary motor cortex: the influence of somatotopy and convergence of multiple effectors

List of Figures *(cont'd)*

Chapter 5 – Increased corticospinal output associated with the convergence of multiple effectors: a TMS study

List of Tables

Chapter 4 – Enhancing excitability of the primary motor cortex: the influence of somatotopy and convergence of multiple effectors

List of Abbreviations

ADLs	Activities of Daily Living
ANOVA	Analysis of Variance
B	Bicep
Contra-M1	Contralateral Primary Motor Cortex
CS	Conditioning Stimulus
CST	Corticospinal Tract
cTBS	Continuous Theta Burst Stimulation
D	Deltoid
DF	Dorsiflexors
DTI	Diffusion Tensor Imaging
D-Wave	Direct Wave
ECR	Extensor Carpi Radialis
EMG	Electromyography
FCR	Flexor Carpi Radialis
FDI	First Dorsal Interossei
fMRI	Functional Magnetic Resonance Imaging
GABA	gamma-Aminobutyric acid
H-Reflex	Hoffmann Reflex
ICF	Intracortical Facilitation
IHI	Interhemispheric Inhibition
Ipsi-M1	Ipsilateral Primary Motor Cortex
ISI	Interstimulus Interval

List of Abbreviations *(cont'd)*

I-Wave	Indirect Wave
LICI	Long Latency Intracortical Inhibition
LIHI	Long Latency Interhemispheric Inhibition
LTD	Long Term Depression
LTP	Long Term Potentiation
MRI	Magnetic Resonance Image
M1	Primary Motor Cortex
MaxForce	Maximum Force
MaxTorque	Maximum Torque
MEPs	Motor Evoked Potentials
MV	Muscle Vibration
MVC	Maximum Voluntary Contraction
NMDA	N-methyl-D-aspartate
PC	Personal Computer
PF	Plantarflexors
PMd	Dorsal Premotor Cortex
PMv	Ventral Premotor Cortex
Pre-SMA	Pre-Supplementary Motor Area
RMT	Resting Motor Threshold
S	Soleus
SI	Primary Sensory Cortex
SIHI	Short Latency Interhemispheric Inhibition

List of Abbreviations *(cont'd)*

SII	Secondary Somatosensory Cortex
SICI	Short Latency Intracortical Inhibition
SMA	Supplementary Motor Area
TA	Tibialis Anterior
TES	Transcranial Electrical Stimulation
TMS	Transcranial Magnetic Stimulation
TS	Test Stimulus

Chapter 1: Introduction

1.1 General objective of thesis

The general objective of this thesis is to examine the role of sensorimotor manipulations on bilateral activation of the primary motor cortex (M1). Many daily activities require the precise interaction and coordination of several brain regions to facilitate purposeful movements of the upper limbs. During bimanual coordination, the movement is more stable when the homologous muscles are active simultaneously rather than in an alternating manner (Cohen, 1971). It is well-established that sustained contractions on one side of the body lead to increases in excitability of the contralateral homologous motor pathways (Cernacek, 1961). Further, corticospinal projections from the M1, which play a very important role in voluntary movement, are facilitated when the contralateral homologous muscles of the opposite limb are contracting (i.e. Perez & Cohen, 2008). The neural mechanisms that mediate these bilateral interactions between muscles of the upper limbs are not currently well understood (Carson, 2005; Swinnen, 2002).

For years researchers have demonstrated activity-dependent changes in the primary motor cortex ipsilateral (ipsi-M1) to the moving limb (Carson, 2005). More recent literature has shown that unilateral motor practice increases motor output to both the trained and the untrained limb, a phenomenon known as cross education (Carroll, Barton, Hsu, & Lee, 2009; Farthing, 2009). Cross education occurs without muscle hypertrophy and is believed to be centrally driven; however, the neural mechanisms mediating this interaction have not been well established. One hypothesis suggests that the transfer of performance gains may be mediated by interactions between M1s (Hinder, Schmidt, Garry, Carroll, & Summers, 2011).

Excitability of the ipsi-M1 increases during unilateral voluntary isometric contractions and increases with increasing contraction intensity (Hortobagyi, Taylor, Petersen, Russell, & Gandevia, 2003; Muellbacher, Facchini, Boroojerdi, & Hallett, 2000; Perez & Cohen, 2008, 2009). There is an increase in corticomotor drive that persists to the untrained limb after unilateral training (Lee, Gandevia, & Carroll, 2009). While M1 may be a prime mediator of this cross education effect, we must first better understand the mechanisms of cross facilitation that are driving these plasticity changes.

Much of the literature has focused on cross excitability changes in the homologous muscle that occur in M1 during isometric contractions (i.e. Perez & Cohen, 2008) and rhythmical movement (i.e. Carson et al., 2004). Not as extensively studied are corticospinal excitability changes in M1 with additional afferent feedback to the homologous muscle (Kossev, Siggelkow, Kapels, Dengler, & Rollnik, 2001) and with remote segment activation (Hortobagyi et al., 2003). Everyday movements are influenced by converging information from multiple areas, including the somatosensory cortex and non-primary motor regions. There is evidence that cross education may have therapeutic utility and if it is mediated by cross excitability changes in M1 we need to gain a better understanding of these mechanisms. This thesis will examine various sensorimotor manipulations and determine whether they enhance excitability of M1 and explore the neurophysiological mechanisms that may be mediating these interactions.

1.2 Background Research

1.2.1 Functional organization of sensorimotor cortex

1.2.1.1 The Motor Cortex

The motor cortex is the main area of the brain involved in motor function and is comprised of Brodmann areas 4 and 6 located in the frontal lobe (Fulton, 1935). Brodmann area 4 is entitled the primary motor cortex (M1) as it contains the largest concentration of corticospinal tract neurons and is involved in the execution of voluntary movement (Dum & Strick, 1991). It lies anterior to the central sulcus on the precentral gyrus and controls voluntary movement on the contralateral side of the body. Brodmann area 6 lies anterior to area 4 and is subdivided into superiorly placed supplementary motor area (SMA) and inferiorly positioned premotor area (Picard & Strick, 1996). Premotor area is comprised of dorsal premotor cortex (PMd) and ventral premotor cortex (PMv) (Picard & Strick, 2001). The non-primary motor areas encompass all areas in the frontal lobe that can influence motor output at both M1 and the spinal cord (Dum & Strick, 2002). The motor cortex also receives information from three other primary sources—somatosensory cortex, cerebellum and basal ganglia. Somatosensory input is relayed directly to M1 from the thalamus. The thalamus also relays information to the motor areas from the cerebellum and basal ganglia.

In the mid-twentieth century, Penfield and Rasmussen (1950) used low intensity electrical stimulation to map the output of M1 and found a somatotopically ordered representational map for movements (or muscles). The somatotopic organization for joints or movements of the motor cortex is very similar to the

somatosensory cortex (Rasmussen & Penfield, 1947). The motor homunculus represents the amount of cortical area dedicated to motor control of a particular body part or region. The most lateral aspect of the homunculus contains the mouth and face, as you move upwards there are the hands, trunk, lower extremities and feet most medially. Both the SMA and premotor areas are somatotopically organized as well. More recent research has upheld the somatotopy of functional subregions for the legs, arms, and head; however, precise topography of specific body parts has been challenged (Sanes & Donoghue, 1997).

The primary motor cortex can be stratified into six cortical layers (1-6, superficial to deep), but lacks granular layer 4. The strong presence of corticospinal neurons is a unique feature of M1; these neurons represent 79% of all corticospinal neurons (Dum & Strick, 1991). Most neocortical neurons (70-80%) are excitatory pyramidal neurons (DeFelipe & Fariñas, 1992). The remaining 20-30% are interneurons, which are mainly inhibitory (White & Keller, 1989). M1 contains both pyramidal neurons and interneurons. Horizontal cortico-cortical connections are contained in layers 1-3; these connections influence the activity of the layer 5 output neurons from M1. Interneurons can release either excitatory or inhibitory neurotransmitters onto pyramidal neurons. The majority of interneurons are inhibitory and utilize gamma-aminobutyric acid (GABA) as a neurotransmitter. The pyramidal neurons in layer 5 utilize the excitatory neurotransmitter glutamate and send motor commands to the spinal cord. The CST forms the main output pathway for the execution of voluntary movement. Approximately 75% of the corticospinal

neurons decussate in the pyramids, while approximately 15% decussate in the spinal cord and the remaining 10% do not cross (Ralston & Ralston, 1985).

1.2.1.2 The Somatosensory Cortex

The somatosensory cortex can be divided into the primary somatosensory cortex (SI), secondary somatosensory cortex (SII) and posterior parietal cortex. SI is located posterior to the central sulcus on the postcentral gyrus and is comprised of Brodmann areas 3a, 3b, 1, and 2. Areas 3b and 1 receive independent thalamocortical projections from various cutaneous afferents, while 3a and 2 receive proprioceptive inputs from peripheral muscle and joint afferents. SII is superior to the lateral sulcus and receives input from SI. The posterior parietal lobe is behind SI and consists of areas 5 and 7. Area 5 integrates tactile input from mechanoreceptors of the skin with proprioceptive input from muscles and joints. The sensory homunculus is much like the motor homunculus. Certain areas of the body are exaggerated owing to greater sensory innervation density and the importance of sensory input from that area as it relates to function.

1.2.1.3 Functional connectivity within & between sensorimotor cortices

We must consider whether there are sources of bilateral functional connectivity within the motor cortical network that have the potential to mediate cross facilitation and thereby provide a mechanism for cross education to function (Farthing, Borowsky, Chilibeck, Binsted, & Sarty, 2007). During unimanual movement, ipsilateral activation has been found in M1, premotor cortex, SMA, primary somatosensory cortex, cerebellum, parietal lobe and cingulate cortex (Dai, Liu, Sahgal, Brown, & Yue, 2001).

In nonhuman primates, the density of connections within the corpus callosum follows a rostrocaudal gradient for M1, SMA-proper and pre-SMA, where the hand representation has the weakest connection (Liu, Morel, Wannier, & Rouiller, 2002). The pre-SMA has denser connections than both the SMA proper and M1. Therefore regions more heavily involved in producing motor output exhibit less transcallosal connections. Fling and colleagues (2013) provided evidence through diffusion tensor imaging (DTI) that interhemispheric transcallosal projections are largely homotopic and there are differences in the quantity and strength of these projections within the motor cortex during voluntary movement. There are more fibers connecting homologous SMA regions than M1, S1, pre-SMA or dorsal premotor cortices (PMd) (Fling, Benson, & Seidler, 2013). Within humans there are more fibers connecting SMA than pre-SMA. Further, there are more fibers connecting SMA than for M1, S1 or PMd. On the other hand M1 does exhibit strong interhemispheric connections between homologous regions. Interhemispheric connections within the SMA have been found to be quite dense in some species (Gould, Cusick, Pons, & Kaas, 1986). Even though the density of these connections has been established and many have considered the SMA to have a critical role in bimanual movements, the SMA has not been given much consideration with respect to cross facilitation. It is clear that even though there is now a vast amount of structural knowledge documenting the presence of anatomical connections between the motor cortices (i.e. Gould et al., 1986), we have not established all the functional roles of these pathways.

As stated above, there is controversy with respect to within-limb somatotopy in the primary motor cortex. Some researchers believe that the organization of representations of within-limb segments (i.e. elbow, fingers, shoulder) overlap, while others believe there is somatotopic distinction despite the overlap studied in primates (Gould et al., 1986; Kwan, Mackay, Murphy, & Wong, 1978; Nudo, Jenkins, Merzenich, Prejean, & Grenda, 1992) and humans (i.e. Kleinschmidt, Nitschke, & Frahm, 1997). Overlap has been shown with various neuroimaging studies in humans (i.e. Grafton, Woods, & Mazziotta, 1993). This overlap is attributed to bidirectional horizontal connections (Huntley & Jones, 1991), convergence of corticospinal output from multiple locations (i.e. Donoghue, Leibovic, & Sanes, 1992) and divergence of corticospinal output to multiple segments (i.e. McKiernan, Marcario, Karrer, & Cheney, 1998). Plow and colleagues (Plow, Arora, Pline, Binenstock, & Carey, 2010) found that somatotopic gradients exist in M1 despite the overlap, which supports functional somatotopy. Functional somatotopy states that within-limb representations have overlap for multi-joint coordination, though there still exists distinct centers for control of individual muscles.

1.2.2 Basics of Transcranial Magnetic Stimulation

1.2.2.1 Background of Transcranial Magnetic Stimulation

Transcranial Magnetic Stimulation (TMS) was originally developed by Barker and colleagues (1985) and provides a means with which we can evaluate corticospinal excitability in humans noninvasively. TMS uses the principal of electromagnetic induction by placing a coil over the scalp. A brief, high current pulse

is produced which produces a magnetic field with the lines of flux passing perpendicular to the plane of the coil (tangential to scalp). The magnetic field can reach 2 Tesla and typically lasts for 100 μs. An electrical field is induced perpendicular to the magnetic field (Hallett, 2007). TMS can activate neurons that lie 1.5 to 2.0 cm below the scalp surface (Rudiak & Marg, 1994). The targets of TMS are typically cortical association fibers or horizontal fibers found in layers 1-3 that synapse with motor neurons (Terao & Ugawa, 2002). Detailed study of the corticofugal discharge in response to the motor stimulus has been studied by Amassian and colleages (Amassian, Cracco, & Maccabee, 1989). There is a short latency direct wave (D-wave) followed by several longer latency indirect waves (I-waves). The D-wave is thought to result from direct depolarization of the initial axon segment of the corticospinal neuron and is most effectively activated by transcranial electrical stimulation (TES) (Merton & Morton, 1980). The I-waves follow the D-wave, approximately 1.5 ms later reflecting the delay required for synaptic discharge. Therefore, the first I-wave (I_1) is thought to be generated through the depolarization of an axon synapsing directly onto a corticospinal neuron, while the following I-waves (I_2,…) may require local polysynaptic circuits. I-waves are elicited using relatively low TMS intensities. TMS can evoke both D-waves and I-waves that arise from trans-synaptic activation of corticospinal neurons.

1.2.2.2 Transcranial Magnetic Stimulation techniques

The output of M1 can be objectively measured using electromyography (EMG) over the muscle of interest. The EMG will record a resultant motor evoked potential (MEP) in the muscle when a single suprathreshold TMS pulse is delivered

to M1 over the muscle representation of interest. This technique is used to measure excitability of specific muscle representations in M1. The resting motor threshold (RMT) is determined as the lowest stimulation intensity that produces an MEP of at least 50 μV on 5 out of 10 consecutive trials. The suprathreshold TMS pulse is usually set at 120% RMT during single pulse TMS conditions to measure cortical excitability.

Paired-pulse TMS techniques can be used to investigate intracortical inhibitory or excitatory circuits. Applying a conditioning stimulus over the same cortical area at a specific time preceding the test stimulus allows us to modulate the amplitude of the MEP. Short –interval intracortical inhibition (SICI) and intracortical facilitation (ICF) are paired-pulse TMS conditions which both use a subthreshold conditioning stimulus (CS) prior to a suprathreshold test stimulus (TS). The subthreshold CS (80% RMT) is high enough to activate cortical neurons; however, it will not have any descending influence on the spinal cord and will not result in an MEP. When the interstimulus interval (ISI) is between 1 and 4 ms it will result in inhibition, while intervals between 8 and 20 ms will result in facilitation (Ziemann, Rothwell, & Ridding, 1996). SICI is likely mediated by GABA-A receptors (Di Lazzaro et al., 2000). Long-interval intracortical inhibition (LICI) uses a paired-pulse TMS technique with two suprathreshold TMS pulses with an ISI of 100-150 ms. LICI is likely mediated by GABA-B receptors.

Interhemispheric interactions can be investigated with dual coil paradigms. Interhemispheric inhibition (IHI) is another paired-pulse TMS technique measured by applying a CS to one primary motor region shortly before a second TS is applied

over the motor region of interest in the opposite hemisphere. IHI has an ISI of 6-50 ms and the magnitude of the MEP is smaller (Chen, Yung, & Li, 2003; Ferbert et al., 1992; Gerloff, Corwell, Chen, Hallett, & Cohen, 1998). The evidence suggests that the CS activates excitatory transcallosal projections that synapse onto networks of inhibitory interneurons in the opposite hemisphere that alter excitability of the pyramidal output neurons (Di Lazzaro et al., 1999; Ferbert et al., 1992; Hanajima et al., 2001). Short latency IHI (SIHI) is obtained with an ISI close to 10 ms and long latency IHI (LIHI) is obtained with an ISI close to 50 ms. Both of these variants appear to be mediated by different neurotransmitters and receptors. LIHI appears to be mediated by GABA-B receptors, while the substrates mediating SIHI are unknown (Daskalakis, Christensen, Fitzgerald, Roshan, & Chen, 2002; Irlbacher, Brocke, Mechow, & Brandt, 2007).

1.2.3 Plasticity in M1

There exists a large body of literature, using both human and animal models, showing that the cerebral cortex is modifiable. The ability of neurons to change their structure and function in response to new experiences is termed neuroplasticity. Intracortical electrical stimulation mapping has demonstrated that M1 maps are able to exhibit long-lasting reorganization (Sanes, Wang, & Donoghue, 1992). The main mechanisms that have been suggested to mediate reorganization in the cerebral cortex involve unmasking of existing but latent horizontal connections (Sanes & Donoghue, 2000) and modulation of synaptic efficiency such as long-term potentiation (LTP) (Hess & Donoghue, 1994) or long-term depression (LTD) (Hess

& Donoghue, 1996). The neurotransmitter systems involved in mediating these effects include the inhibitory Ɣ-aminobutyric acid (GABA)ergic system (Hess, Aizenman, & Donoghue, 1996; Jacobs & Donoghue, 1991) as well as the excitatory glutamatergic system with activation of N-methyl-D-aspartate (NMDA) receptors (Hess et al., 1996; Hess & Donoghue, 1994). M1 reorganization likely depends on the balance of excitatory and inhibitory influences.

Transcranial magnetic stimulation (TMS) has provided a means of exploring M1 mapping in humans. M1 movement representations and modifications have been studied in many populations, including amputations (Cohen et al., 1991; Hall, Flament, Fraser, & Lemon, 1990), spinal cord injuries (Topka, Cohen, Cole, & Hallett, 1991), temporary modifications of sensory input (Brasil-Neto et al., 1992) and immobilization (Liepert, Tegenthoff, & Malin, 1995). TMS is assumed to activate neurons superficial to layer V; however, many cortical and subcortical neurons are activated, so we cannot be sure that M1 is the source of modifications seen in these populations. Animal studies show that motor representations can be altered in regions of the cortex adjacent to a lesion, however undamaged motor representations may reduce further in topographical size in the absence of proper motor training (Castro-Alamancos & Borrel, 1995). Daily repetitive training after an injury can preserve motor representations that were unaffected by the infarct, suggesting that these areas may have a role in recovery due to cortical plasticity (Nudo & Milliken, 1996; Nudo, Milliken, Jenkins, & Merzenich, 1996). It is important that we increase our understanding of the factors that influence excitability changes within M1 in order to better understand what is mediating neuroplasticity.

1.2.4 Bilateral interactions between limbs

1.2.4.1 Motor irradiation

Involuntary contractions during intended unilateral activation of the homologous muscles of the opposite limb is observed in children under the age of 10 and is referred to as mirror movements. Mirror movements are a characteristic developmental pattern and they decrease in occurrence with age (i.e. Mayston, Harrison, & Stephens, 1999). Neurophysiological studies suggest that mirror movements in a healthy child are caused by corresponding activation of the crossed CSTs from both M1s (Mayston et al., 1999; Reitz & Müller, 1998). These movements are abnormal in the mature motor system; however, contraction of muscles on one side of the body can lead to increased excitability of the opposite resting homologous motor pathway. In the mature motor system this is called motor irradiation, which is demonstrated using surface EMG (Cernacek, 1961). EMG activity in the resting homologous muscles can be observed occasionally during repetitive movements of a distal upper limb muscle. These effects are more pronounced when they are performed against an external resistance (Cernacek, 1961) or with increased effort (Hopf, Schlegel, & Lowitzsch, 1974). The amount of neural drive to the active muscle does appear to affect the amount of motor irradiation seen in the quiescent muscle.

1.2.4.2 Cross facilitation between homologous muscles

TMS has provided a method to assess excitability changes of the corticomotor pathway to the muscle beyond EMG. Excitability changes that arise in a quiescent muscle during voluntary activation of its contralateral homologous

muscle have been extensively studied. Voluntary contractions of one limb give rise to increases in the excitability of descending projections to the homologous muscles of the opposite limb (Hess, Mills, & Murray, 1986). Many factors that modulate cross facilitation are also those that when manipulated alter the level of cross education that is brought on by unilateral training.

MEPs induced in musculature of the hand by TMS of the motor cortex are facilitated by isometric contraction of homologous muscles of opposite limb (Hess, Mills, & Murray, 1987, 1986; Liepert, Dettmers, Terborg, & Weiller, 2001). This phenomenon has also been reproduced in more proximal muscle groups (Hortobagyi et al., 2003; Perez & Cohen, 2008). Furthermore, the amplitude of MEPs in resting muscles increases with increased force of contraction of the active muscle (Dettmers et al., 1995; Hess et al., 1986; Hortobagyi et al., 2003; Meyer, Roricht, Grafin von Einsiedel, Kruggel, & Weindl, 1995; Muellbacher et al., 2000; Perez & Cohen, 2008, 2009). Modulation of excitability of corticospinal pathways of the opposite limb also occurs during rhythmic movements and appears to require voluntary drive of the muscle (Carson et al., 2004; Carson, Welsh, & Pamblanco-Valero, 2005; Carson, Riek, & Bawa, 1999). Continuous cyclical contractions have demonstrated cross excitability changes during active rhythmical movements of the wrist (Carson et al., 2004) and finger (Uehara, Morishita, & Funase, 2011; Uehara, Morishita, Kubota, & Funase, 2013). The modulation of excitability is phase dependent – excitability is increased when the homologous muscle is most strongly engaged.

The type of contraction and strength of the contraction affect cross facilitation. Tonic contractions appear to facilitate excitability more than phasic contraction (Liepert et al., 2001). However, in a more proximal muscle group, Uematsu and colleagues (2010) and Howatson and colleagues (2011) both found that lengthening contractions produced greater increases in excitability when compared to shortening and isometric contractions. Cross facilitation is also augmented by the strength of the contraction (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2008; Stinear, Walker, & Byblow, 2001). At low levels of force (under 30% maximum voluntary contraction, MVC) excitability is not consistently amplified (Liepert et al., 2001; Muellbacher et al., 2000; Stinear et al., 2001). It appears with some more complex movements, i.e. dextrous finger movements, that do not require increased levels of force production, excitability is augmented (Tinazzi & Zanette, 1998). There may be an influence of increased recruitment of other muscles that may help to influence corticomotor drive to the resting muscle. Past studies have seen an increase in excitability of both the homologous and non-homologous muscles during isometric contractions (i.e. Hortobagyi et al., 2003); however, facilitation is greatest in the homologous muscle. The spread of excitability to the non-homologous muscle has not been well studied to date, so the spread of cross facilitation is not yet known. Voluntary drive to the muscle also appears to be required to see a change in cortical excitability. During passive movement, excitability changes are not seen in the ipsilateral motor cortex (Carson & Riek, 2000).

Since corresponding changes in response amplitude are not obtained when potentials are evoked by stimulating the corticospinal pathway at the level of the cervicomedullary junction (Carson et al., 2004; Hortobagyi et al., 2003), it has been concluded that the phenomenon of cross facilitation is modulated by interhemispheric interactions between cortical motor areas. Further, there is often a reduction in SICI that parallels increased excitability in the resting motor cortex during movement of the homologous muscle (Perez & Cohen, 2008). Modulation has also been seen in IHI from the active M1 to the resting M1 (Hinder, Schmidt, Garry, & Summers, 2010; Nelson, Hoque, Gunraj, Ni, & Chen, 2009; Perez & Cohen, 2008). Many studies have found there is an increase in IHI during voluntary contraction (Ferbert et al., 1992; Hinder et al., 2010), but the relationship these pathways have with intracortical circuits may be the reason there is still a resultant increase in cross excitability.

It cannot be assumed that direct interactions between primary motor cortices represent the source of cross facilitation. In monkeys, mirror movements are abolished by the temporary inactivation (through injection of muscimol) of M1 ipsilateral to the actively moving limb, whereas they are largely preserved, or enhanced, in circumstances in which the opposite M1 (i.e. contralateral to the moving limb) is injected (Tsuboi, Nishimura, Yoshino-Saito, & Isa, 2010). This suggests cross facilitation arises from common drive to both primary motor cortices from other centers in the motor network. Cross facilitation is present in patients with agenesis of the corpus callosum (Meyer et al., 1995), supporting the idea that this is not only an M1-M1 interaction.

1.2.4.3 Surround inhibition

Surround inhibition is a neural mechanism to focus neuronal activity in the central nervous system (Angelucci et al., 2002). This mechanism is more heavily studied in sensory systems, but there is recent research exploring this occurrence in the motor cortex during different skilled behaviours. Within the motor cortex it is thought to be mediated by GABA receptors to support the selective execution of desired movements (Hallett, 2003; Ziemann, Rothwell, & Ridding, 1996). During active contractions the muscle of interest will display increases in excitability, while neighbouring muscles will be inhibited (Sohn & Hallett, 2004). This mechanism (surround inhibition) is typically present at the initiation of movement, but is absent during tonic contractions (Beck et al., 2008). It is also found to be stronger at lower force levels (i.e. 10% max force) and disappears at higher force levels (i.e. 40% max force) (Beck, Schubert, Richardson, & Hallett, 2009). With callosal neurons being glutamatergic and facilitatory to their immediate targets, this may be one mechanism aiding in the selective excitability of the contralateral homologous muscle. Surround inhibition may be mediated locally (i.e. within M1) and/or it may be mediated by inputs from other motor regions (Beck & Hallett, 2011).

1.2.4.4 Cross education

It is well known that the contralateral untrained limb can benefit from unilateral strength training, known as cross education. This effect is task specific and occurs in the opposite homologous muscles (Carroll, Herbert, Munn, Lee, & Gandevia, 2006; Lee & Carroll, 2007; Munn, Herbert, & Gandevia, 2004; Zhou, 2000). This phenomenon has been documented as early as 1894 (Scripture, Smith, &

Brown, 1894) and has relevance with respect to clinical rehabilitation. There is a large body of literature studying cross education, demonstrating that there is not only a transfer of strength, but also motor skill (Imamizu & Shimojo, 1995; Laszlo, Baguley, & Bairstow, 1970; Parlow & Kinsbourne, 1989). Despite the longstanding interest in this concept, there is little consensus concerning the mediating neural mechanisms.

Execution of many unilateral tasks is associated with increased excitability of both contralateral and ipsilateral cortical motor areas. One model thought to be responsible for cross education is 'cross activation.' This model suggests that bilateral cortical activity generated during unilateral training drives simultaneous neural adaptations in both cerebral hemispheres. Unilateral training induces task specific changes in the configuration of cortical motor networks that normally control the muscles of the opposite (quiescent) limb (Hellebrandt, 1951). Since the magnitude of the cross-activation is contingent on the intensity of the unilateral contraction (Perez & Cohen, 2008), the degree of transfer is predicted to scale with the amount of neural drive required to perform the training task. Bilateral variations in the excitability of the corticospinal projections during movements that are by intention unilateral have been demonstrated using TMS. MEPs induced by TMS are increased in amplitude by isometric contractions of the homologous muscles in the opposite forearm (Hortobagyi et al., 2003). The amount of potentiation or cross facilitation is positively correlated with the amount of force that is generated by the contractions of the opposite limb (Perez & Cohen, 2008).

With cross education research, no relationship has been found across participants between the degree of cross education and increases in the excitability of corticospinal projections to the homologous muscles engaged in training, when these are assessed at rest in the context of either acute (Carroll, Lee, Hsu, & Sayde, 2008; Hinder et al., 2011) or chronic (Hortobágyi et al., 2011) training protocols. Based on this, it would appear reasonable to consider whether the functional adaptations that support interlimb transfer gains in performance either occur in areas upstream of the primary motor cortex or via changes in the effectiveness of synaptic transmission through projections from these areas onto M1 targets.

1.2.4.5 Influence of lower limb on upper limb motor excitability

In the past, voluntary activation of the upper limb muscles has been shown to facilitate the monosynaptic reflex elicited in the lower limb muscles, known as the Jendrassik maneuver. Corticospinal excitability changes have since been investigated across different interlimb interactions of the upper and lower limb. Within M1 there is no overlap or neurophysiological connections between arm and leg muscle representations (Brown, Day, Rothwell, Thompson, & Marsden, 1991; Huntley & Jones, 1991). Excitability changes arising in upper limb M1 representations during lower limb contraction are likely due to secondary motor areas where arm and leg regions overlap significantly, rather than horizontal connections in M1. There is a large body of literature that has investigated the stability and preferred isodirectional movement of the hand and foot (Jeka & Kelso, 1995). More recently TMS has been used to probe the neural correlates associated with the changes observed in both isodirectional and nonisodirectional

coordination of the hand and foot. Byblow and colleagues (2007) explored the functional connectivity between secondary and primary motor areas during foot movement. They found that upper limb corticomotor excitability and SICI were altered by movement conditions involving leg muscle activation and connections between SMA-M1 appeared to facilitate forearm corticospinal excitability in a non-specific manner (Byblow et al., 2007). Past research has demonstrated modulation of GABA-mediated inhibitory networks in upper limb M1 representations during foot movements (Baldissera & Borroni, 2002; Borroni, Cerri, & Baldissera, 2004). Previous studies have also found a reduction in upper limb corticomotor excitability when conditioning SMA at rest (Civardi, Cantello, Asselman, & Rothwell, 2001); however, lower limb dorsiflexion and plantarflexion appear to cause facilitation relative to rest. Reduced cortical inhibition in the hand muscles as a result of discrete (Sohn, Kang, & Hallett, 2005) and phasic (Tazoe, Endoh, Nakajima, Sakamoto, & Komiyama, 2007) dorsiflexion movement of the ipsilateral foot has also been observed during measurements of silent period duration. Fujiyama and colleagues (2012) found that coordination of contralateral limbs (right hand, left foot) resulted in decreased corticospinal inhibition compared to coordination of ipsilateral limbs (right hand, right foot). Borroni and colleagues (2004) found that H-reflex excitability modulations in the upper limb remained phase linked to muscle contractions, not movement, of the lower limb. Force signals generated by Golgi tendon organs during movement of the foot muscles may in fact reach the hand motor area, modulating its excitability (McIntyre, Proske, & Rawson, 1984).

1.2.4.6 Effect of sensory input on motor excitability

Proprioceptive input has a major impact on motor control at both the spinal and the cortical level (Gandevia, 2001; Wiesendanger & Miles, 1982). Motor neuron firing rate may be reduced by 30% in the absence of afferent feedback (Macefield, Gandevia, Bigland-Ritchie, Gorman, & Burke, 1993). Peripheral electrical stimulation can modulate afferent input and induce neuroplastic changes in motor cortical areas (Kaelin-Lang et al., 2002; Ridding, Brouwer, Miles, Pitcher, & Thompson, 2000); this suggests projections from the somatosensory cortex can modulate motor cortical excitability. In the motor cortex peripheral nerve lesions (Kolarik, Rasey, & Wall, 1994) and limb amputations (Chen, Corwell, Yaseen, Hallett, & Cohen, 1998; Qi, Stepniewska, & Kaas, 2000) result in reorganization of the motor representation in the deafferented hemisphere (Cohen et al., 1991). Most reports concentrate on the effects of deafferentation on contralateral cortical representations; however, decreased somatosensory input also elicits changes in the contralateral hemisphere. Acute deafferentation of a limb results in bilateral cortical reorganization. This may be driven by transcallosal connections between homologous muscle groups.

Many studies have used TMS to evaluate the impact of afferent sensory input on excitability of the human motor cortex (i.e. Rosenkranz & Rothwell, 2003). Muscle vibration has been a strong tool for modifying MEPs (Claus, Mills, & Murray, 1988; Kossev, Siggelkow, Schubert, Wohlfarth, & Dengler, 1999; Rosenkranz, Altenmüller, Siggelkow, & Dengler, 2000; Siggelkow et al., 1999). Modulation of afferent input through tendon and muscle vibration mainly excites 1a fibers of muscle spindle afferents (Burke, Hagbarth, Löfstedt, & Wallin, 1976; Collins & Prochazka, 1996; Gandevia, 1985; Martin & Park, 1997; Roll, Vedel, & Ribot, 1989).

Muscle vibration to forearm muscles augments MEPs in the resting contralateral muscle (Claus et al., 1988; Kossev et al., 1999), while MEPs in the non-vibrated functional antagonist show depression (Siggelkow et al., 1999).

1.2.4.7 Sources of functional connectivity

It is believed that interhemispheric interactions between cortical motor centres are mediated in large measure by the fibers of the corpus callosum (Kobayashi, Hutchinson, Schlaug, & Pascual-Leone, 2003). During everyday tasks involving the upper extremities, some movements require the two upper limbs to execute quite different actions, while others require symmetry. Therefore the corpus callosum provides a basis for highly organized patterns of facilitation and inhibition (Carson, 2005). Researchers have been developing various experimental methods capable of elucidating these patterns during voluntary movement tasks.

So far, the expression of IHI has been examined almost exclusively via changes in the excitability of corticospinal projections to intrinsic hand muscles (i.e. Ferbert et al., 1992). This is an important consideration, as the cortical representations of proximal and distal muscles are distinguished in terms of their structural and functional interhemispheric relations. With respect to their structural connectivity in nonhuman primates, there is a greater density of callosal projections between cortical areas that represent proximal muscles, than between those corresponding to distal muscles. Recently Ibey and colleagues (Ibey, Bolton, Buick, Staines, & Carson, 2015) investigated whether the IHI parameters established for the hand can be applied more generally to muscles of the forearm. They found that

like the hand, IHI increased as a function of the CS and was exhibited between 7 and 15 ms with 10 ms being the most effective ISI to induce SIHI.

Unimanual movement and practice may exert its effects on the functional capacity of the opposite limb through modification of the focal excitatory relationship between the primary motor cortices or it may have an effect on intracortical circuits. In a condition in which sizes of the conditioned and test MEP were matched across torque levels, Perez and Cohen (Perez & Cohen, 2008) reported that IHI measured in the resting FCR muscle during isometric flexion of the opposite wrist was lower when torque was generated at 30% and 70% MVC, but not at 10% MVC. In contrast, when the MEPs generated by the CS were not matched across conditions, an increase in IHI was obtained. IHI is abolished when forces greater than 50% MVC are generated by the muscle in which the test MEP is recorded (Chen et al., 2003). Interhemispheric inhibition (and facilitation) provides only a partial indication of the relationship between the physiological processes that are operating during the execution of movements.

1.2.5 Applications

With structure and function of the cerebral cortex being modifiable post injury, it is no wonder why treatments that maximize neuroplasticity are becoming increasingly popular. Bimanual movement training is proving to be an effective training method to enhance sensorimotor control of upper extremity musculature following brain injury due to stroke (Cauraugh, Lodha, Naik, & Summers, 2010; McCombe Waller & Whitall, 2008; Stinear, Barber, Coxon, Fleming, & Byblow, 2008;

Summers et al., 2007); however, there is little understanding of the neurophysiological changes that drive behavioural improvements. Bilateral movement has been shown to enhance activity in the primary motor cortex of the affected hemisphere sub-acutely in individual patients with hemiparesis (Staines, McIlroy, Graham, & Black, 2001). The mechanisms responsible for cross facilitation between the primary motor cortices are poorly understood in healthy individuals and are important in understanding the neurophysiology of everyday upper limb movements, which can then be utilized in customizing rehabilitation protocols following brain injury.

In addition, there is significant risk associated with loss of muscle strength and general capacity resulting from limb immobilization in the elderly. They are at higher risk of falls and also unable to perform activities of daily living and may be unable to live independently. Even in younger people with functional reserves, 3 weeks of immobilization leads to declines in strength in the order of 50% of initial capacity (Hortobágyi et al., 2000). Some researchers have found that if the opposite limb is trained during period of immobilization, the loss of functional capacity is lessened (Farthing, 2009; Magnus, Barss, Lanovaz, & Farthing, 2010; Pearce, Hendy, Bowen, & Kidgell, 2013). Given the therapeutic potential, there is a need to provide a neurophysiological basis upon which to design interventions and protocols to individual requirements.

1.3 Setup and Terminology

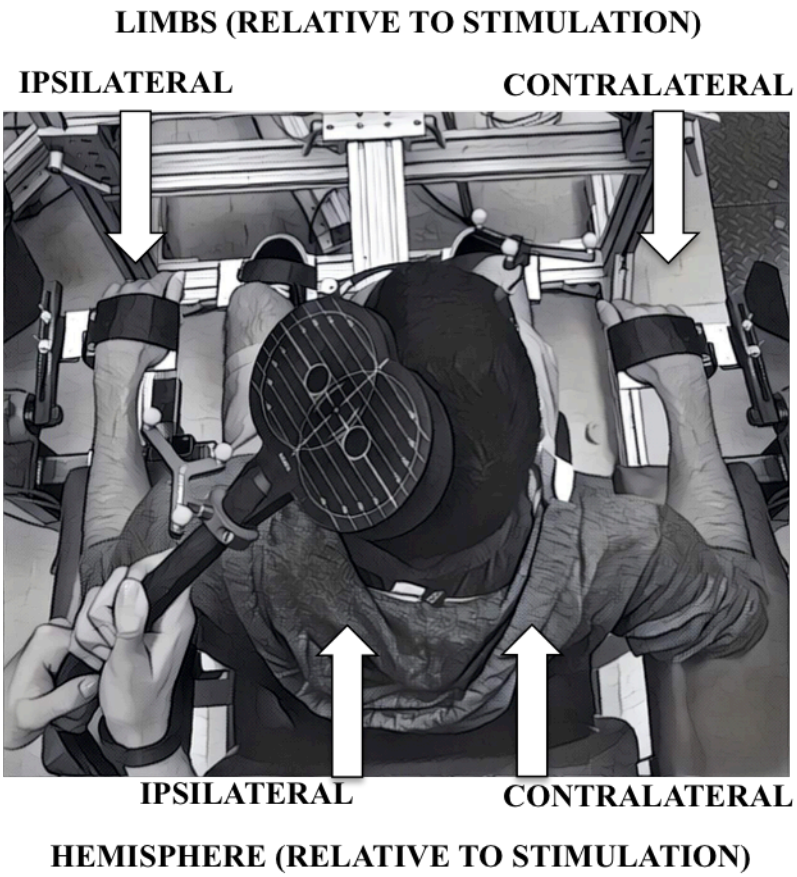

Figure 1.1. Thesis setup terminology. This diagram signifies that the terms ipsilateral and contralateral are relative to the hemisphere of stimulation.

1.4 Specific research objectives

1.4.1 Research objective 1

To investigate ways in which the primary motor cortices interact during rhythmical movement at increasing load requirements.

Research has demonstrated that corticospinal excitability to the resting muscle increases during both isometric contractions (Perez & Cohen, 2008) and rhythmical movement (Carson et al., 2004) involving the contralateral homologous muscle. Excitability also depends on the force of contraction (Perez & Cohen, 2008). There is also evidence that MEPs may be influenced by the amount of motor drive to the contracting muscle; they are potentiated when the muscle is most strongly engaged (Carson et al., 2004). The majority of the literature has focused on static isometric contractions against increasing forces and not much focus has been on dynamic movements. The aim of this study was to examine changes in corticomotor excitability to a resting wrist extensor muscle during contralateral rhythmical isotonic and static isometric wrist contractions (flexion/extension) at different loads and positions using TMS. We hypothesized that voluntary activation of the ipsilateral homologous muscle would increase primary motor cortex excitability and this would be further potentiated with increased movement loads.

1.4.2 Research objective 2

To examine the effect of added sensory input on cortical projections to the contralateral resting limb during rest and during movement and force manipulations.

Past research has looked at the impact of afferent sensory input on excitability of M1. Muscle vibration has been used as a means to explore the impact of muscle spindle afferents on M1 excitability. Kossev and colleagues (2001) showed cross excitability changes during muscle vibration to the resting homologous wrist muscles. We wanted to explore the influence of additional afferent information on cross excitability during contralateral isotonic rhythmical contractions. The goal of this study was to examine the effect of added sensory input on corticomotor projections to the contralateral resting limb during rest and rhythmical movement with increased load using TMS. We hypothesized that low amplitude vibration of forearm muscles ipsilateral to stimulation would increase excitability to the contralateral resting muscle through transcallosal projections and decreased GABAergic activity. The force of contraction of the vibrated muscle would further augment MEP excitability.

1.4.3 Research objective 3

To investigate interhemispheric interactions and somatotopic relationships influencing excitability of M1 and to determine how activity of multiple effectors influences excitability of M1 (convergence).

Excitability of the corticomotor projections to the resting limb increase with contraction of the homologous muscle. This has been studied extensively during both isometric and isotonic contractions (Carson et al., 2004; Ibey & Staines, 2013; Perez & Cohen, 2008). Both Ibey and Staines (2013) and Hortobagyi and colleagues (2003) found non-homologous muscle activation also contributed to cross

excitability changes. The question arose of whether somatotopy and spatial location of the muscle representation would have an influence on cross excitability changes. Motor excitability changes are seen in the upper extremity when the remote segment (leg) is activated (Hortobagyi et al., 2003). There is a homuncular sequence to the body representation in M1, however there is debate regarding the degree of overlap (Plow et al., 2010). If cortical representations overlap, contracting nearby muscles (to the muscle of interest) should have an effect on excitability changes in M1 seen with TMS. The goal of this study was to investigate interhemispheric interactions and somatotopic relationships influencing excitability of M1 and to determine how the coordination of multiple effectors influences excitability of M1. We hypothesized that motor cortical excitability will be enhanced during ipsilateral activation of the homologous muscle and will decrease as a function of spatial position during contralateral activation. Recruitment of dual effectors (hand-foot) will increase M1 excitability greater than one effector (hand).

1.4.4 Research objective 4

To probe the neural mechanisms contributing to changes in M1 during isometric contraction of multiple effectors – studying the effect of primary and non-primary motor areas to excitability changes in the resting motor cortex.

Past research has demonstrated excitability changes in the resting upper limb during activity of the lower limb. Motor excitability is increased in the upper limb during contraction of the lower limb (Hortobagyi et al., 2003) and also increases with contraction of the contralateral homologous muscle (Perez & Cohen,

2008). The previous study in this thesis demonstrated excitability changes with the contraction of the remote effector and with contraction of the remote effector with the homologous muscle. The aim of this study was to investigate the neural mechanisms contributing to changes in M1 during the convergence of M1 and non-primary motor cortices. We hypothesized that decreased GABAergic activity within M1 will influence excitability changes.

Chapter 2: Corticomotor excitability changes seen in the resting forearm during contralateral rhythmical movement and force manipulations: a TMS study

Ibey RJ, Staines WR. (2013). Corticomotor excitability changes seen in the resting forearm during contralateral rhythmical movement and force manipulations: a TMS study. *Behav Brain Res,* 257: 265-74.

2.1 Overview

The aim of this study was to examine changes in corticomotor excitability to a resting wrist extensor muscle during contralateral rhythmical isotonic and static isometric wrist contractions (flexion/extension) at different loads and positions, using transcranial magnetic stimulation (TMS). TMS-induced motor-evoked potentials (MEPs) were recorded from the relaxed right extensor carpi radialis (ECR) and flexor carpi radialis (FCR) respectively, while the left arm underwent unimanual manipulations. Rhythmical isotonic (0.5 Hz) flexion and extension movements of the left wrist under 3 load conditions (no, low and high force) and a frequency matched passive movement condition were collected, along with isometric flexion/extension contractions in each position (low and high force). TMS was delivered at eight positions (4 in the flexion phase and 4 in the extension phase) during the continuous movement conditions and each of these positions was sampled with isometric contraction. The potentials evoked by TMS in right ECR were potentiated when the left ECR was engaged, independent of position within that phase of contraction or contraction type (isotonic and isometric). Motor cortical excitability of the resting right ECR increased as load demands increased to the left wrist. Passive rhythmical movement did not influence excitability to the resting ECR implying that voluntary motor drive is required. Our findings indicated that the increase in corticomotor drive during both rhythmic isotonic and static isometric contractions of the opposite limb is likely mediated by interhemispheric interactions between cortical motor areas. Improving our understanding of these cortical networks can be useful in future methods to enhance neuroplasticity

through neurorehabilitation methods.

2.2 Introduction

For years researchers have demonstrated activity-dependent changes in the primary motor cortex ipsilateral to the moving limb (ipsi-M1) (Carson, 2005). More recent literature has shown that unilateral motor practice increases motor output to both the trained and the untrained limb, a phenomenon known as cross education (i.e. Carroll et al., 2008). One hypothesis suggests that M1 mediates this interaction. Excitability of the ipsi-M1 goes up during unilateral voluntary isometric contractions and increases with increasing contraction intensity (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008). There is an increase in corticomotor drive that persists to the untrained limb after unilateral training (Lee et al., 2009). While M1 may be a prime mediator of this cross education effect, we must better understand the mechanisms of cross facilitation that are driving these plasticity changes. The aim of this study was to examine changes in corticomotor excitability to a resting wrist extensor muscle during contralateral rhythmical isotonic and static isometric wrist contractions (flexion/extension) at different loads and positions, using transcranial magnetic stimulation (TMS).

In primates, control of the distal muscles of the upper limb was thought to be regulated by the contralateral motor system and devoid of transcallosal connections between the individual primary motor cortical (M1) representations (Brinkman & Kuypers, 1973). More recent research with primates (Aizawa, Mushiake, Inase, &

Tanji, 1990; Cisek, Crammond, & Kalaska, 2003; Donchin et al., 2002; Donchin, Gribova, Steinberg, Bergman, & Vaadia, 1998; Matsunami & Hamada, 1981; Tanji, Okano, & Sato, 1988) and humans (Chiappa et al., 1991; Wassermann, Pascual-Leone, & Hallett, 1994) has demonstrated ipsilateral motor cortical activation during unimanual movements. Functional magnetic resonance imaging (fMRI) studies show blood oxygen level-dependent signal activity in ipsi-M1 to the active arm (Dai et al., 2001; Dettmers et al., 1995; Thickbroom, Phillips, Morris, Byrnes, & Mastaglia, 1998; van Duinen, Renken, Maurits, & Zijdewind, 2008). Transcranial magnetic stimulation (TMS) studies reveal increased corticospinal tract (CST) activation to the resting contralateral homologous muscle with unimanual contraction (Hess et al., 1986; Hortobagyi et al., 2003; Meyer, Röricht, von Einsiedel, Kruggel, & Weindl, 1995; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman, Davey, & Ellaway, 1998; Stinear et al., 2001; Tinazzi & Zanette, 1998; Zwarts, 1992) and rhythmical movement (Carson et al., 2004, 2005; Carson et al., 1999; Stinear & Byblow, 2002; Uehara et al., 2013). While there is increasing evidence that excitability in ipsi-M1 is modulated during unimanual motor activity, the characteristics mediating this interaction between the hemispheres are not well understood.

Electrophysiological studies in primates have shown that activity in the CST contralateral to an active upper limb may be indirectly modulated by both joint torque and position in static conditions, though dynamic movements have not been studied extensively (Ashe, 1997; Evarts, 1968; Evarts, Fromm, Kroller, & Jennings, 1983; Werner, Bauswein, & Fromm, 1991). Similarly in humans, both fMRI (Dai et

al., 2001; Dettmers et al., 1995) and TMS (Abbruzzese, Morena, Spadavecchia, & Schieppati, 1994; Sekiguchi, Kimura, Yamanaka, & Nakazawa, 2001; Sekiguchi, Nakazawa, & Suzuki, 2003) studies support these claims demonstrating that activity in contralateral M1 (contra-M1) to a contracting limb is related to both the magnitude of the contraction and the position of the limb. TMS studies have explored whether excitability changes in ipsi-M1 are correlated to changes in contra-M1. These studies have found excitability of the CST projections to the resting homologous muscle increases with greater force of contraction (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). Both Stinear and colleagues (2001) and Perez and Cohen (2008,2009) found positive correlations between the amplitudes of motor evoked potentials (MEPs) in both contra-M1 and ipsi-M1 with increasing levels of contraction. There is also evidence that MEPs may be influenced by the amount of motor drive to the muscle; they are potentiated when the muscle is most strongly engaged (Carson et al., 2004; Yahagi et al., 2003).

The regions and pathways involved in this bi-hemispheric interaction are not well understood. The majority of literature has focused on static movements against increasing forces and the mechanisms involved; thus, how these mechanisms are modulated during dynamic movement with increasing load requirements is not yet known. Many ADLs require movements to be rhythmical or dynamic in nature in order to accomplish a task. The aim of the present study was to investigate ways in which the primary motor cortices interact during rhythmical movement at increasing load requirements. The mechanisms responsible for changes in ipsi-M1

relative to sensory and motor manipulations in contra-M1 (cross facilitation) remain poorly understood. We used single pulse TMS to explore changes in corticomotor excitability during both rhythmical isotonic and isometric contractions at different loads. It was hypothesized that voluntary activation of the left wrist extensors (homologous muscle) would increase ipsi-M1 excitability of the homologous extensors and this would be further potentiated at increased movement loads. The pattern of this excitability modulation was hypothesized to be specifically related to the movement phase of the contralateral limb (i.e. highest when the homologous muscle to that tested was functionally active). Further, excitability would be higher when the muscle is most strongly engaged within that phase of movement. We additionally hypothesized that non-homologous muscle activation would not increase ipsi-M1 activation.

2.3 **Materials and Methods**

2.3.1 Participants

Eight right-handed healthy volunteers (4 female, 4 male) with an average age of 25.8 ± 4.1 years participated in each of three experiments collected in a single session. Handedness was confirmed using the Edinburgh Inventory (Oldfield, 1971). All participants had no contraindications to TMS or any known neurological impairments. Participants gave their informed written consent to participate in the studies and completed a screening form for TMS. All of the experimental procedures were approved by the Office of Research Ethics at the University of Waterloo.

2.3.2 Experimental Setup

Participants were seated in an armchair with their forearms fully supported on an adjustable table. Arms were positioned 20–30° from their torso with both forearms in 90–100° of flexion. The right forearm rested in a neutral (semiprone) position during the entire experiment. The left forearm was secured semiprone in a custom-made manipulandum with the hand fastened midpalm thereby isolating wrist movement and eliminating grip (Fig. 2.1A). A rotating shaft and potentiometer were located above the wrist joint axis allowing for full extension and flexion range of motion. The rotating shaft was equipped with a wheel where resistance could be added using a fastener, providing tension in both directions of movement. The potentiometer monitored displacement of the wrist joint through a custom LabVIEW program (National Instruments, Austin, TX, USA) used to trigger TMS stimulation during active or passive wrist movements. A metronome provided auditory cuing for movements of the left wrist. The tones were presented to the right of the participant (opposite to the side of the TMS trigger). At the beginning of each experiment subjects performed 3 trials of maximum voluntary isometric contractions for both wrist flexion and wrist extension. Maximum torque (MaxTorque) was used as an estimation of maximum force output (MaxForce). MaxForce was measured using a digital force/torque gauge (Chatillon Force Measurement, Greensboro, NC, USA) recording their MaxForce in Newtons. Subjects alternated between wrist flexion and extension MaxForce acquisition with a rest period between each contraction (~20 s). The MaxForce for both wrist flexion and wrist extension were averaged and the lesser of the two was used to calculate the

10% and 30% MaxForce for the experimental conditions (resistance on the manipulandum had to be the same in both directions). MaxForce was used to determine their contraction level, which was continuously monitored and collected using electromyography over extensor carpi radialis longus (ECR) and flexor carpi radialis (FCR) in Neuroscan 4.3 (Compumedics, Charlotte, NC, USA). Since resistance was the same for both wrist flexors and wrist extensors, we have termed no resistance as no force, 10% as low force and 30% as high force.

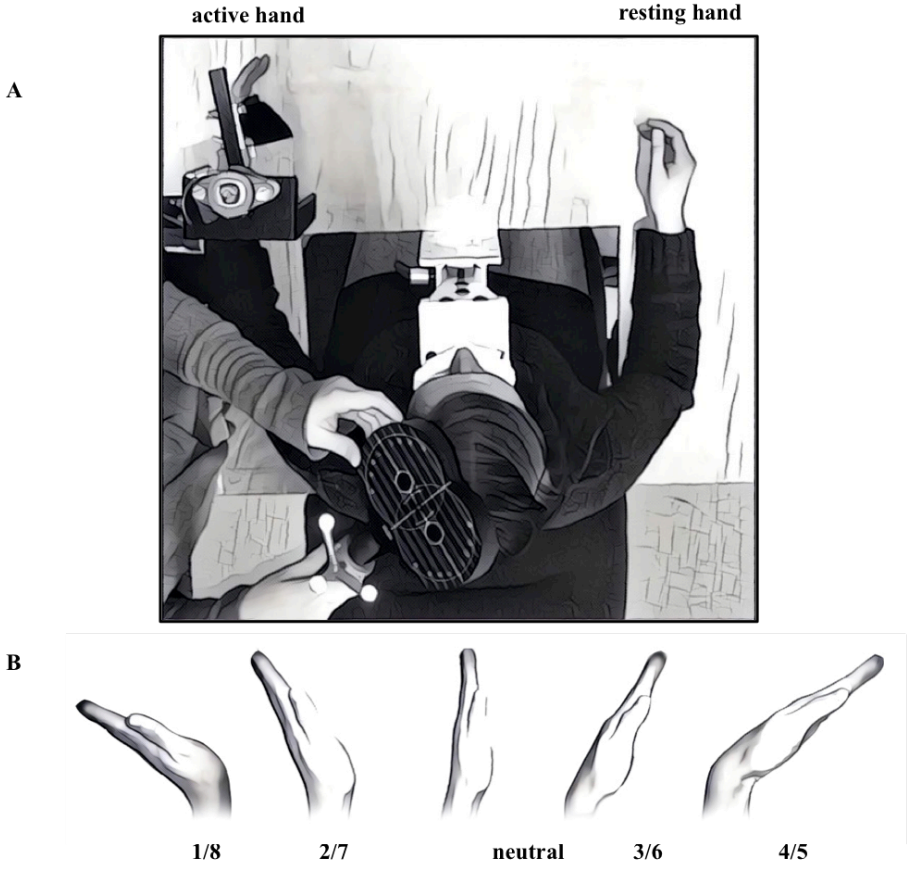

Figure 2.1. Experimental set-up. (A) Overhead view of the experimental set-up where subjects were instructed to perform either rhythmical flexion and extension at the left wrist or stationary (isometric) contractions into either flexion or extension at various wrist positions, while they were fixed in a custom made manipulandum. The right arm was at rest in all experimental conditions. (B) Wrist orientations representing the 4 TMS and median nerve stimulation positions during isotonic and passive movement. Positions 1 to 4 represent the flexion phases

(moving from full wrist extension to full wrist flexion) while positions 5 to 8 represent the extension phases (moving from full wrist flexion to full wrist extension). The 5 hand positions represent the wrist orientations during both isometric flexion and extension.

2.3.3 Electromyography

Electromyographic (EMG) activity of ECR, FCR and first dorsal interosseous (FDI) was recorded from both upper limbs using bipolar surface electrodes placed longitudinally over the muscle bellies. EMG signals were amplified (1000×), filtered (bandpass 1–200 Hz) and recorded using a SynAmps^2 amplifier and Neuroscan 4.3 (Compumedics, Charlotte, NC, USA) with a sampling rate of 1000 Hz. EMG data from the left FCR was collected through a standard EMG amplifier (amplified 5000×, Grass Technologies, Model QP511, West Warwick, RI) at the same sampling rate as a high-level input to Neuroscan. All EMG data were stored on a PC for off-line analysis.

2.3.4 Transcranial Magnetic Stimulation (TMS)

Motor evoked potentials (MEPs) were collected from the resting right arm by stimulating the left motor cortex with a MagPro x100 stimulator (Medtronic, Minneapolis, MN, USA) through a figure-eight coil (loop diameter 10 cm; model MC-B70) using a biphasic waveform. Coil placement and orientation was continuously monitored using Brainsight (Brainsight 2; Rogue Research, Montréal, QC, Canada), a TMS neuronavigation system that displays real-time coil placement and target location on an anatomical magnetic resonance image (MRI). The coil was oriented tangential to the scalp with the handle pointing posterior and 45° away from the midline to activate the corticospinal system. This particular orientation induces a posterior-anterior directed current in the motor cortex (Kammer, Beck, Erb, &

Grodd, 2001).

TMS was delivered over the optimal location to elicit a response in the ECR muscle of the right arm and was delivered using single pulses. Resting motor threshold (RMT) was defined as the lowest stimulation intensity where potentials with peak to peak amplitude of a minimum of 50 μV were evoked in at least five out of ten trials (Rossini & Rossi, 2007). TMS was triggered externally using a customized LabVIEW program (National Instruments, Austin, TX, USA) and the magnitude of individual TMS pulses was 120% of RMT.

2.3.5 Experimental Procedures

2.3.5.1 Experiment 1: Effect of Isotonic Contraction

Participants were instructed to perform rhythmic flexion and extension of their left wrist, using their full range of motion, to the beat of an auditory metronome at a frequency of 0.5 Hz. The metronome ran at a frequency of 1 Hz so subjects could coordinate maximum flexion and extension with each beat. Each trial (~3 min) began with a movement calibration where a custom program measured full wrist displacement. The total displacement was divided into four equal intervals corresponding to 8 position targets (4 moving from maximum extension to maximum flexion and 4 moving from maximum flexion to maximum extension) (Fig. 2.1B). TMS was triggered ten times in each phase position resulting in a total of 80 MEPs collected during each condition. All eight-phase positions were triggered in a random order before repeating. For the low force and high force isotonic conditions the resistance was applied prior to the trial and measured by the digital

force/torque gauge. Resistance was re-measured at the end of each trial to ensure the resistance level remained consistent throughout the condition.

2.3.5.2 Experiment 2: Effect of Isometric Contraction

Participants were positioned in each of the five positions represented in Fig. 2.1B and asked to isometrically contract into either extension or flexion while an experimenter secured the manipulandum still with the digital force/torque gauge. The positions that were sampled during the rhythmical isotonic contractions were duplicated with the addition of neutral. The digital force/torque gauge measured the force of contraction. A researcher held the instrument and provided consistent feedback to the participant so they could gauge their level of contraction during a practice trial, as well as during the actual trial. Each position was sampled 10 times resulting in 100 MEPs (5 positions, each isometrically contracting into flexion and extension). This was completed for both low and high force.

2.3.5.3 Experiment 3: Effect of Passive Movement

Participants were secured semiprone in the same custom-made manipulandum used for the first two experiments with the hand fastened midpalm, thereby isolating wrist movement and eliminating grip. A researcher rhythmically moved the participant's left wrist through flexion and extension to the beat of an auditory metronome at a frequency of 0.5 Hz. The metronome ran at a frequency of 1 Hz so the researcher could coordinate maximum flexion and extension with each beat. MEPs were elicited in the right ECR by triggering the TMS at the 8 phase positions using the same procedure as in Experiment 1.

Experiments 1 through 3 were collected together within one session. All conditions were pseudo-randomized with each participant. Vision was eliminated for all conditions – resting and movement tasks. Resting MEP data (20 MEPs) were obtained with both hands stationary in semiprone position. Resting values were acquired at each of the 4 positions sampled for both isotonic and isometric contractions. During all conditions experimenters monitored motor activity in the right resting limb and provided auditory feedback to the participant where necessary to ensure all muscles were silent.

2.3.6 Data Analysis

The mean EMG amplitude of ECR (rectified) was calculated at each position for both isotonic and isometric conditions. A 200 ms time window was epoched around the stimulus (0 ms represented stimulus onset). Fig. 2.2 shows the raw electromyographic profile from one participant for both the isotonic (A) and isometric (B) conditions. There were ten EMG profiles for each position, which were then rectified, averaged and the mean amplitude was calculated. The mean amplitudes for each position for all 8 participants were averaged together (Fig. 2.3). A one-way ANOVA was run on the raw EMG at each position for each of the isotonic movement conditions to test the effect of phase position. Pre-planned contrasts were run to test if there was a difference in EMG activity from position 5 and 8 based on the hypothesis that excitability would be greater when the muscle is most strongly engaged in that phase of movement. A frequency analysis was also performed on the potentiometer data for the no force rhythmical movement and passive movement condition to ensure the frequency was equivalent.

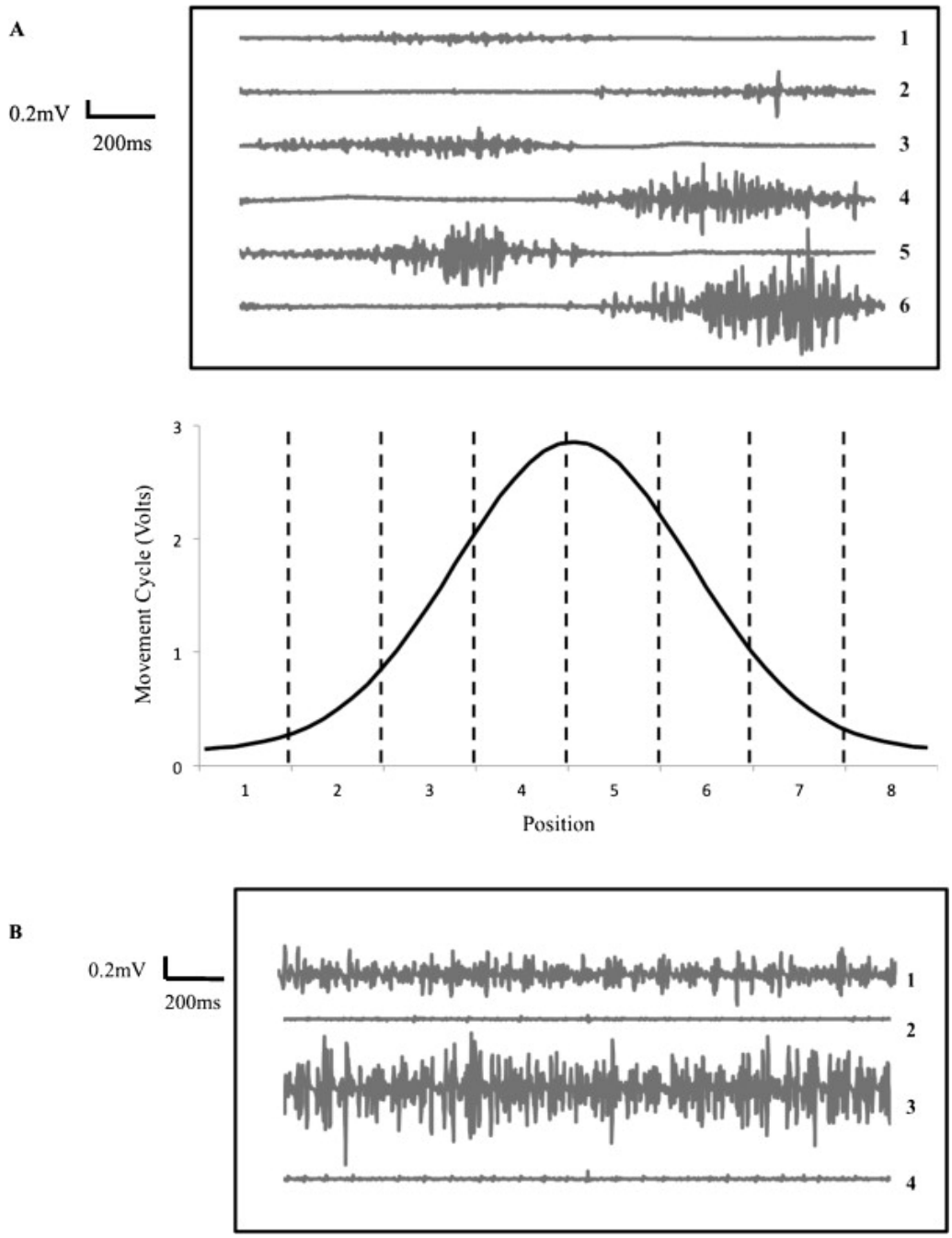

Figure 2.2. Raw electromyographic data of left wrist isotonic and isometric movement at different force levels. (A) EMG data from one participant collected during rhythmical wrist flexion and extension performed at a frequency of 0.5 Hz. A 2000 ms epoch is shown at each of the 3 levels of force: no force (1–2), low force (3–4) and high force (5–6). The flexors are represented by figures 1, 3 and 5 while the extensors are represented by figures 2, 4 and 6. EMG recordings are from the left flexor carpi radialis (FCR) and left extensor carpi radialis longus (ECR) (B) EMG data from one participant collected during isometric extension in neutral position. Low force of ECR (1) and high force of ECR (3). FCR activity is represented by (2) and (4)

during the low force and high force ECR contractions, respectively.

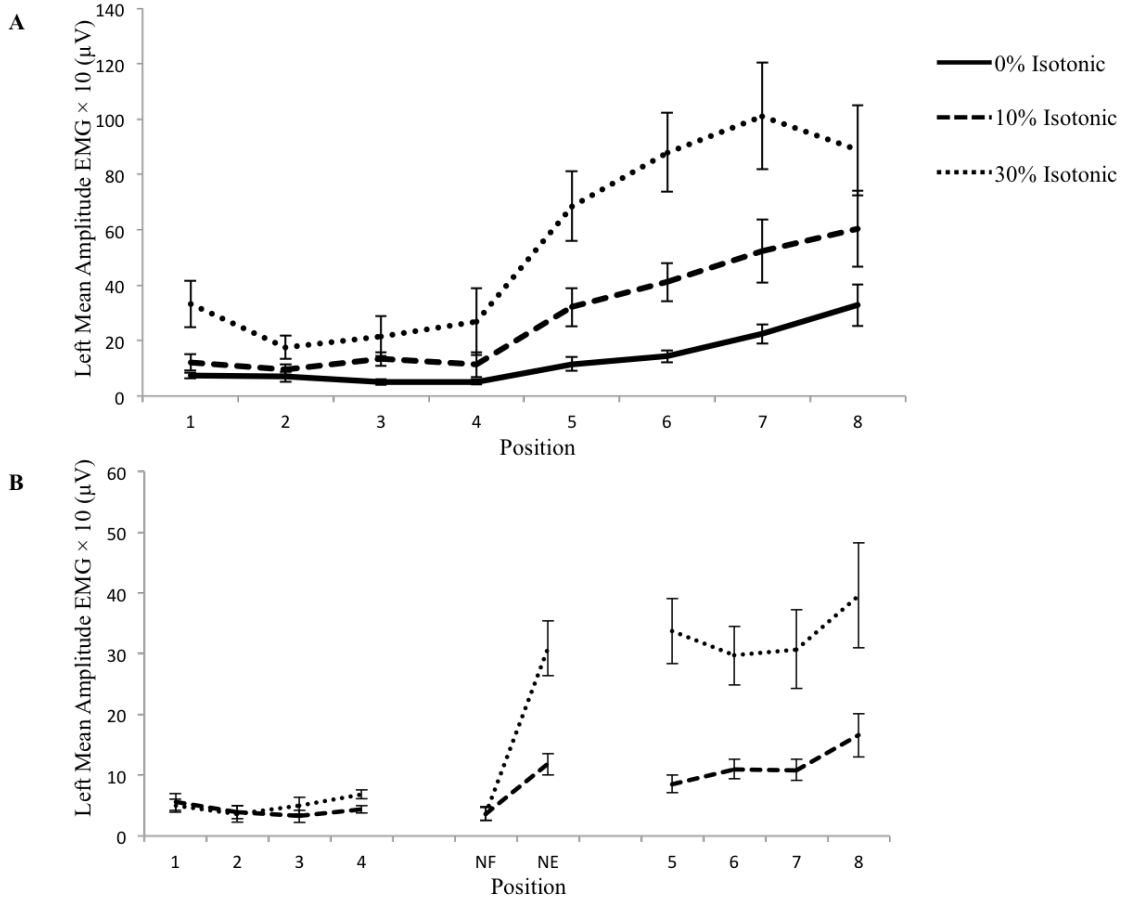

Figure 2.3. Electromyographic data of left wrist isotonic movement at increasing forces. (A) Average rectified EMG data (n=8) from the left ECR during isotonic movement at each level of force. (B) Average rectified EMG data (n=8) from the left ECR during isometric movement at each level of force. Error bars represent standard error of the mean.

Experimenters examined the EMG data in the resting right arm prior to analysis of the MEPs for all conditions to ensure the muscles were silent. Relaxation of the right ECR and FCR was defined as EMG activity below mean amplitude of 25 μV (Muellbacher et al., 2000). The mean amplitude (rectified) was calculated 50 ms prior to delivery of TMS for all isotonic movement conditions—in all subjects

at every phase position—to ensure right ECR was at rest. EMG profiles were acquired for right ECR, FCR and FDI.

The MEPs elicited in the right ECR during each phase of movement of the left arm were normalized with respect to the MEPs elicited during the resting trials. Two main analyses were performed for each experiment. Firstly, a two-way ANOVA was used to determine the effect of factors contraction level (i.e. no, low and high force for isotonic and low and high force for isometric) and phase position (1, 2, 3, 4, 5, 6, 7, 8 for isotonic and the addition of two neutral positions for isometric) on right resting ECR MEPs. *A priori* contrasts were used to test the hypothesis that facilitation of right ECR would be greatest during activation of the homologous muscles (extension vs. flexion) within each experiment. When the F value was significant, i.e. main effect of contraction level or phase position, Tukey's post hoc tests were used to determine the means by which they differed ($p < 0.05$). Lastly, a one-way ANOVA was conducted on each factor when an interaction was present.

2.4 **Results**

2.4.1 Electromyograms from left (active) and right (resting) arm

It is apparent from the graphs that the activity of left ECR is modulated based on the length of time spent in the extension phase (increased EMG activity of left ECR with further muscle shortening) (Fig. 2.3A and B). The one-way ANOVA revealed a main effect of phase position ($F_{7,49} = 16.27$, $p \leq 0.0001$ for no force; $F_{7,49} = 12.21$, $p \leq 0.0001$ for low force; $F_{7,49} = 17.25$, $p \leq 0.0001$ for high force) and pre-planned contrasts demonstrated a difference in EMG activity for position 5 and

8 for no force and low force conditions ($F_{7,49}$ = 38.07, $p \leq 0.0001$ for no force, $F_{7,49}$ = 11.88, $p \leq 0.001$ for low force and $F_{7,49}$ = 2.96, $p \leq 0.09$ for high force). It is clear from the analyses that there is increased motor drive (EMG activity) with further muscle shortening. Frequency analysis of the potentiometer data revealed the average frequency for the active and passive conditions was 0.498 ± 0.0006 and 0.499 ± 0.0005 Hz respectively. A two-tailed paired t-test revealed this was not significantly different (p = 0.43).

The average EMG activity for right (resting) ECR at dynamic no, low and high force was 1.17 ± 0.08 μV, 1.10 ± 0.06 μV and 2.52 ± 0.30 μV respectively. No significant differences were found between the conditions for EMG activity of all three muscles. Peak to peak amplitude was calculated for all the MEPs (n = 10) from the right resting ECR muscle for each position and for each subject, these were then averaged in order to carry out the analyses. Fig. 2.4 represents averaged raw MEP traces from one individual subject for each of the conditions.

2.4.2 Experiment 1: Effect of Isotonic Contraction

Fig. 2.5 illustrates right ECR MEPs averaged at each position during no, low and high force rhythmical isotonic contractions. Repeated-measures ANOVA revealed a main effect of contraction level ($F_{2,14}$ = 5.64; $p \leq 0.01$) and phase position ($F_{7,49}$ = 12.09; $p \leq 0.0001$), but no interaction between contraction level and phase position ($F_{14,98}$ = 0.83; $p < 0.6$) on MEPs from the resting right ECR. Post hoc analysis showed a significant difference between isotonic no force and high force ($p \leq 0.05$). Pre-planned contrasts of the specific phase positions within each level of force were completed to test the specific hypotheses. They indicated that CST excitability for

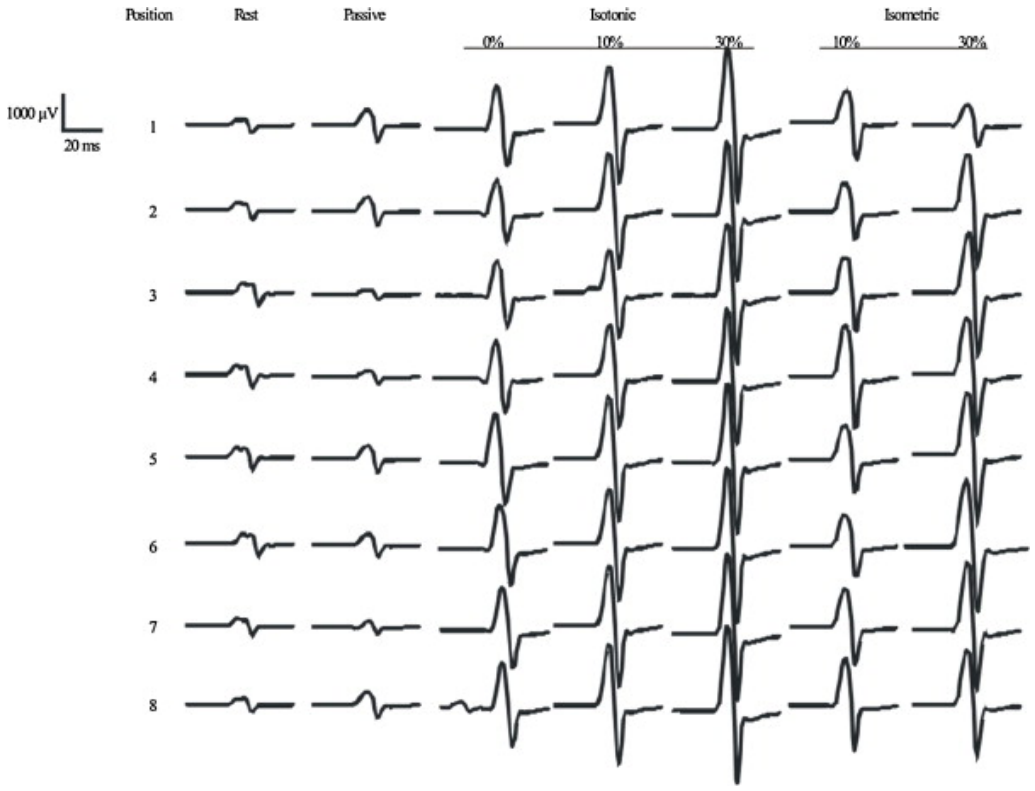

Figure 2.4. Effect of contralateral isotonic and isometric muscle activation on motor evoked potentials in the resting limb for one individual subject. Mean motor evoked potentials (MEPs) in the resting right ECR from one participant during rest, passive, isotonic and isometric conditions. Each trace (10 MEPs) corresponds to both the condition and one of eight phase positions.

extension phases (positions 5–8) is significantly higher than flexion phases (positions 1–4) ($F_{1,49}$ = 42.16, $p \leq 0.0001$ for no force; $F_{1,49}$ = 50.99, $p \leq 0.0001$ for low force; $F_{1,49}$ = 21.29, $p \leq 0.0001$ for high force). In order to test the hypotheses that within the extension phase the position of greatest muscle shortening (position 8) would have a greater MEP amplitude than the position of greatest muscle length (position 5), contrasts were run on these positions. From the results, the specific phase position during extension did not have an effect on CST excitability ($F_{1,49}$ = 3.38, $p \leq 0.07$ for no force; $F_{1,49}$ = 0.71, $p \leq 0.4$ for low force; $F_{1,49}$ = 0.11,

$p \leq 0.7$ for high force). Testing the hypothesis that CST excitability would decrease to the resting muscle when the non-homologous muscle was active against increasing forces was not performed statistically. Referring to Fig. 2.5 it is clear that contrary to our hypothesis the ECR MEP amplitudes increased with increased flexion forces.

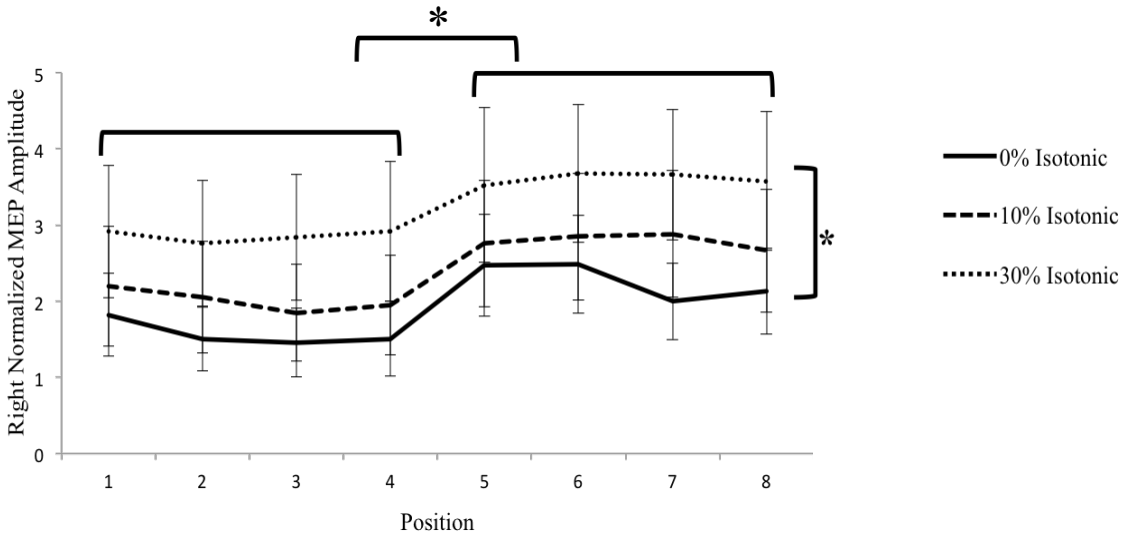

Figure 2.5. Mean effect of rhythmic movement of the contralateral wrist at increasing levels of force on the motor evoked potentials induced by TMS. The mean (n=8) motor evoked potentials (MEPs) in right (resting) ECR at each position in the movement cycle. Positions 1-4 represent the flexion phase while positions 5-8 represent the extension phase. All values are normalized with respect to a control condition (MEPs collected with the left limb at rest). Error bars represent standard error of the mean. * $p<0.05$

2.4.3 Experiment 2: Effect of Isometric Contraction

Fig. 2.6 shows right ECR MEPs averaged at each position during low and high force isometric contractions normalized to rest. Repeated-measures ANOVA revealed a main effect of phase position ($F_{9,63} = 3.36$, $p \leq 0.002$), however there was no significant effect of contraction level ($F_{1,7} = 3.81$, $p \leq 0.09$) and no interaction between contraction level and phase ($F_{9,63} = 1.28$, $p \leq 0.3$) on right ECR MEPs. Pre-

planned contrasts of the specific phases (flexion and extension) within each level of force indicated that like the isotonic contractions there was a significant increase in CST excitability when the homologous muscle was active ($F_{1,63}$ = 13.94, $p \leq 0.0004$ for low force; $F_{1,63}$ = 6.66, $p \leq 0.01$ for high force). Fig. 2.7 displays the similar phase effects for both contraction types. The flexion positions (1–4) and the extension positions (5–8) were collapsed for both the isotonic and isometric contractions in order to illustrate the phases (flexion and extension) within each contraction type for each level of force.

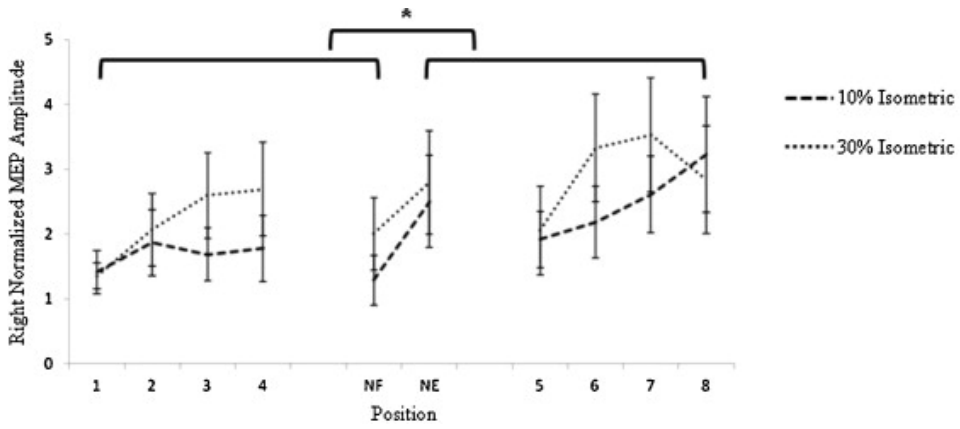

Figure 2.6. Mean effect of isometric contraction of the contralateral wrist at increasing levels of force on the motor evoked potentials induced by TMS. The mean (n = 8) motor evoked potentials (MEPs) in right (resting) ECR at each position in the movement cycle. Positions 1–4 represent the flexion phase while positions 5–8 represent the extension phase. Neutral flexion is labelled as NF while neutral extension is labelled as NE. All values are normalized with respect to a control condition (MEPs collected with the left limb at rest). Error bars represent standard error of the mean. *p < 0.05.

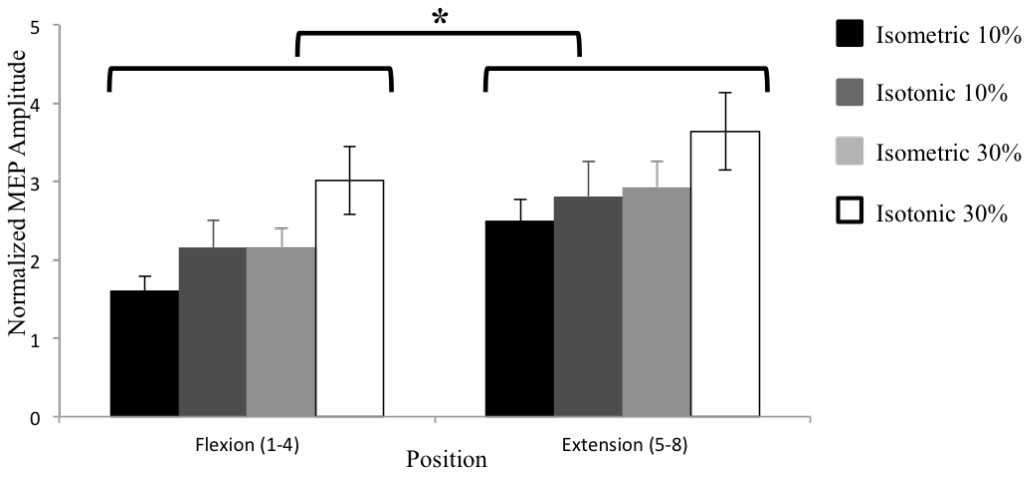

Figure 2.7. Main effect of extension and flexion conditions of the contralateral wrist on the motor evoked potentials induced by TMS. The mean (n = 8) motor evoked potentials (MEPs) in right (resting) ECR in each phase during both isometric and isotonic movement conditions. Positions 1–4 represent the flexion phase while positions 5–8 represent the extension phase. All values are normalized with respect to a control condition (MEPs collected with the left limb at rest). Error bars represent standard error of the mean. *$p < 0.05$.

2.4.4 Experiment 3: Effect of Passive Movement

Fig. 2.8 displays right ECR MEPs normalized to rest for both active rhythmical movement against no resistance and passive rhythmical movement. Repeated-measures ANOVA was run on raw MEPs for rest, passive movement and active movement (no force condition); this showed an effect of contraction level ($F_{2,14} = 4.58$, $p \leq 0.03$), phase ($F_{7,49} = 4.36$, $p \leq 0.0008$) and a contraction level × phase interaction ($F_{14,98} = 3.52$, $p \leq 0.0001$). A one-way ANOVA was run on the passive and active movement conditions to test if there was any effect of phase, which was not significant for the passive movement condition ($F_{7,49} = 1.73$, $p \leq 0.12$) but was significant for the active movement condition ($F_{7,49} = 7.68$, $p \leq 0.0001$).

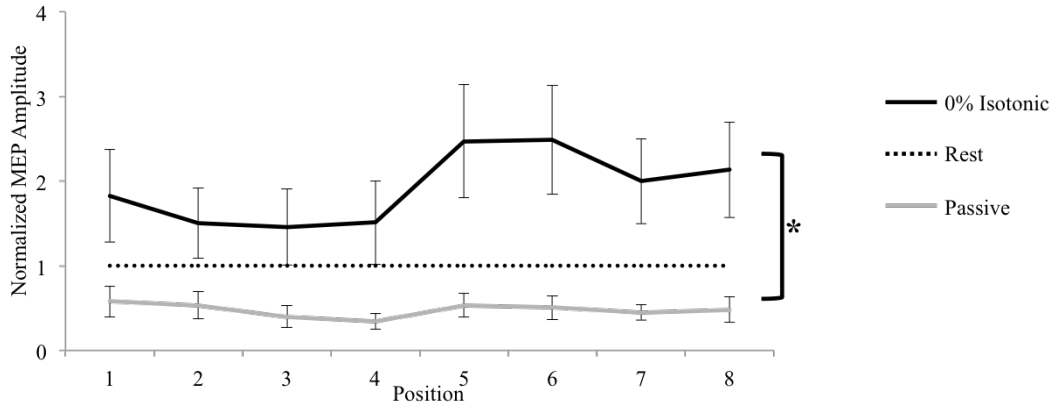

Figure 2.8. Mean effect of rhythmic movement of the contralateral wrist during both voluntary and passive movement conditions on the motor evoked potentials induced by TMS. The mean (n = 8) motor evoked potentials (MEPs) in right (resting) ECR at each position in the movement cycle. Positions 1–4 represent the flexion phase while positions 5–8 represent the extension phase. All values are normalized with respect to a control condition (MEPs collected with the left limb at rest). Error bars represent standard error of the mean. *p < 0.05.

2.5 Discussion

The focus of this study was to investigate ways in which the primary motor cortices interact during both rhythmical isotonic and isometric contractions at different load requirements. We investigated changes in corticomotor excitability in ipsi-M1 to an active hand undergoing both movement and force manipulations. Our findings demonstrate that during voluntary rhythmic flexion and extension of the left wrist, potentials evoked in the resting right ECR by TMS to ipsi-M1 are potentiated when the contralateral homologous muscle (extensors) is active; this enhancement of the MEPs is amplified as the required contraction force increases. In addition, MEPs are also increased (to a lesser degree) when the non-homologous muscle (FCR) is active (Fig. 2.5). Furthermore, we reveal that the positions within each phase do not have an effect on corticomotor excitability to the resting muscle,

rather this increase in excitability is solely phase-dependent (extension versus flexion). To our knowledge, this is the first study to investigate the effect of force during rhythmical isotonic contractions on the corticomotor excitability to the resting limb while also comparing the effect of position and contraction type.

2.5.1 Modulation of corticomotor excitability during voluntary contractions (isotonic and isometric)

Given that MEPs are recorded over the muscle in the periphery and TMS activates corticospinal neurons both directly and trans-synaptically, the MEP amplitude is reflective of excitability along the entire motor pathway to the muscle; this includes neurons in the motor cortex and spinal motor neurons (Rothwell, Thompson, Day, Boyd, & Marsden, 1991). This study highlights that corticomotor excitability induced in the resting ECR was enhanced with increased motor cortical output to the moving limb during both dynamic and static movement. Previously Carson et al. (Carson et al., 2004) found that MEPs evoked in right resting FCR were potentiated during the phases of movement in which the left FCR was most strongly engaged during rhythmical flexion and extension of the left wrist. These results lend support to the idea that there is increased cross-facilitation between the motor cortices during homologous muscle activation. Our results are in agreement with these findings and further show that excitability to the resting ECR goes up with increased motor cortical output to the moving limb. These results are in agreement with previous studies showing excitability of M1 increases with increased activity in the opposite hemisphere (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). This

was demonstrated here for both the isotonic and isometric contraction conditions. There was a significant effect of phase for the isometric conditions (low force and high force)—analogous to isotonic—when the homologous muscle was active. The effect of increasing the force of contraction was evident for the dynamic isotonic conditions more than the isometric (Fig. 2.7). The dynamic recruitment of other non-primary motor areas may contribute to this increased sensitivity. While MEPs were enhanced to the resting homologous muscle at low force, this contrasts the finding of Perez and Cohen (Perez & Cohen, 2008) who found a more prominent facilitation of MEPs in the left FCR between 30% and 10% isometric contraction force, but no difference was observed between rest and 10%. We observed changes even at low levels of force, therefore the overall balance of interhemispheric inhibition targeting ipsi-M1 may be reduced during rhythmical movements. This would have provided a constant corticomotor drive to the muscle, which may remove the inhibitory effect seen at low levels of force that has previously been reported. The influence of movement against low levels of force on ipsi-M1 have generated conflicting results in the literature (Hess et al., 1986; Liepert et al., 2001; Perez & Cohen, 2009; Perez & Cohen, 2008; Sohn, Jung, Kaelin-Lang, & Hallett, 2003; Stedman et al., 1998). These studies have reported facilitation, inhibition and no change in excitability of ipsi-M1.

Excitability of the active muscle (driven by contra-M1) correlates to changes in excitability in ipsi-M1. While we also hypothesized that excitability would increase with increased shortening of the muscle within that phase (measured by the 4 positions in each phase of movement), this did not have an effect. There is no

difference in the size of MEPs from the resting ECR when the homologous muscle is contracting across all positions within that movement phase. MEPs in the most lengthened and most shortened position during the extension phase of contraction in the isotonic conditions were not significantly different. This suggests that the amount of corticomotor drive to the active muscle is consistent across the movement phase, as evidenced by cross-facilitation to the resting muscle remaining consistent.

The cross-facilitation between M1s during the voluntary movement conditions was also not specific to homologous muscle activation. Increased activation of the resting muscle was seen when the non-homologous muscle was active which increased with further force output (see Fig. 2.5). Hortobagyi et al. (2003) delivered TMS to the cortical motor representation of right FCR (resting) while contractions were performed on the left side. They also reported that MEPs were enhanced with the strength of contraction in the left FCR, and comparably they also showed an increase when the extensors were active and a lesser increase when the ankle dorsiflexors were active at 75% MVC. They attributed this increase to a generalized increase in cortical excitability with strong voluntary contractions. However, we found increases in excitability even at lower levels of contraction force when the non-homologous muscle was active, which were graded based on the level of force. Afferent input to the muscle may also contribute to this increase in excitability during both homologous and non-homologous motor activation, however this is not consistent with our findings during the passive movement condition.

2.5.2 Modulation of spinal excitability during voluntary (isotonic) contractions and passive movement

Muscle spindles are sensitive to changes in muscle length and the velocity of the length change. Quick and tonic stretch of the muscle spindle is monitored by Ia afferents. Passive movement allows us to evaluate the effect of afferent input on corticomotor excitability. Previous studies have looked at the effect of passive movement on contra-M1 and have shown phasic modulation of MEP excitability (Carson et al., 1999; Coxon, Stinear, & Byblow, 2005; Lewis & Byblow, 2002; Lewis, Byblow, & Carson, 2001; Stinear & Byblow, 2002). This facilitation in contra-M1 likely originates in the somatosensory cortex; yet this modulation does not appear to transfer to ipsi-M1. Our findings show a depression of MEPs during the passive movement condition that is significantly below the voluntary rhythmical movement condition, consistent with the findings of Carson et al. (Carson & Riek, 2000).

2.5.3 Pathways mediating changes in spinal and cortical excitability

Potentials resulting from TMS reflect excitability of neurons in the motor cortex, although they are also influenced by subcortical activity and the state of spinal motor neurons. Carson and colleagues found that potentials elicited by stimulating the descending tracts at the level of the cervicomedullary junction are unaffected by rhythmic voluntary movement (Carson et al., 2004) which was supported by Hortobagyi and colleagues (2003) who found MEPs were unaffected by isometric contractions of the contralateral limb. One hypothesis is that this cortical facilitation is conveyed by transcallosal connections to the homotopic area of the contralateral motor cortex (Hanajima et al., 2001). Interhemispheric

inhibitory interactions between the primary motor cortices can be measured using a paired-pulse TMS technique, interhemispheric inhibition (IHI) (Ferbert et al., 1992; Uehara et al., 2013). IHI reflects transcallosal connections that are mediated by glutamatergic projections acting through $GABA_B$ interneurons (Chen, 2004; Di Lazzaro et al., 1999; Ferbert et al., 1992; Gerloff et al., 1998; Meyer et al., 1995; Rothwell et al., 1991). Perez and Cohen (2008) found decreases in IHI from contra-M1 to ipsi-M1 during unimanual right wrist flexion at 30% and 70% MVC compared to rest and 10% force. In addition, Uehara and colleagues (2013) have demonstrated that connectivity to M1 from the dorsal premotor cortex as well as long interval intracortical inhibition, reflecting local $GABA_B$ receptor inhibition, are modulated in the motor cortex ipsilateral to movement of the fingers. This suggests that there may be a decrease in inhibition acting between the motor cortices. This is opposed by evidence that increases in ipsi-M1 excitability was present during unimanual forceful contractions in patients that had agenesis of the corpus callosum, and no difference was found in comparison to healthy subjects (Meyer et al., 1995). This suggests that this interaction may at least be partially mediated at a subcortical level. In summary, these results suggest that there is a decrease of inhibitory interactions during these unimanual contractions. The neurophysiological mechanisms underlying the changes we have seen during rhythmical movements against increasing loads needs to be investigated further.

2.5.4 Significance of findings and clinical implications

We investigated ways in which the primary motor cortices interact during rhythmical movement at increasing load requirements. The present findings

indicate that cross facilitation between the active primary motor cortex and the resting primary motor cortex is dependent on the phase of contraction and the amount of force output, however it is not dependent on the position of the muscle within that phase of contraction. While cross-facilitation is enhanced by homologous muscle activation, the antagonist muscle also up-regulates cortical excitability. This contradicts previous findings, so future studies will be investigating this up-regulation of excitability of the non-homologous muscle during dynamic movements.

Bimanual movement training is proving to be an effective training method to enhance sensorimotor control of upper extremity musculature following brain injury due to stroke (Cauraugh, Lodha, Naik, & Summers, 2010; Lin, Chen, Chen, Wu, & Chang, 2010; McCombe Waller & Whitall, 2008; Stinear et al., 2008; Summers et al., 2007), however there is little understanding of the neurophysiological changes that drive behavioural improvements. Bilateral movement has been shown to enhance activity in the primary motor cortex of the affected hemisphere sub-acutely in individual patients with hemiparesis (Staines et al., 2001). The mechanisms responsible for cross-facilitation between the primary motor cortices are poorly understood in healthy individuals and are important in understanding the neurophysiology of every day upper limb movements which can then utilize in customizing rehabilitation protocols following brain injury.

It is well known that the contralesional M1 plays a critical role in the recovery of motor function on the affected side post-stroke. This research illustrates that both weak and strong rhythmical contractions up regulate excitability in the

resting hemisphere throughout the whole range of movement. Research in this area will help us understand interactions between the hemispheres and how we can use the unaffected side to influence neuroplasticity and functional changes on the affected side.

Chapter 3: Convergence of transcallosal sensorimotor information: effect of rhythmical movement, force and vibration on the contralateral resting limb

3.1 Overview

The goal of this study was to examine the effect of added sensory input on corticomotor projections to the contralateral resting limb during rest and rhythmical movement with force manipulations using transcranial magnetic stimulation (TMS). TMS-induced motor evoked potentials (MEPs) were recorded from the right extensor carpi radialis (ECR) while the left arm underwent unimanual manipulations. Rhythmical isotonic (0.5 Hz) flexion and extension movements of the left wrist were performed both with and without added force (20% maximum voluntary contraction) and muscle vibration (MV) to the left ECR muscle belly in a custom-made manipulandum. A frequency matched passive movement condition was also collected with and without vibration to the ECR muscle belly. TMS was delivered within both the flexion phase and extension phase during the movement conditions. Single pulse TMS was delivered along with paired-pulse TMS techniques to investigate the effect of intracortical inhibition (SICI and LICI). The potentials evoked by TMS in the right ECR were potentiated when the left ECR was engaged, this increased as force increased to the left wrist. Passive rhythmical movement did not influence excitability of the resting ECR. MV also did not influence excitability of the right ECR. Furthermore, there was no effect on intracortical inhibition. Our findings indicated that the increase in corticomotor drive during rhythmical isotonic movement is likely mediated by interhemispheric interactions between motor cortical areas. More research is required to progress our knowledge on the influence of afferent information during unimanual movement. Improving our understanding

of these cortical networks may influence future methods to enhance neuroplasticity through neurorehabilitation techniques.

3.2 Introduction

Transcallosal connections between homologous muscle groups during unilateral voluntary contractions have been well researched and shows both the contralateral and ipsilateral sensory and motor cortical areas are activated (Cramer et al., 1999; Muellenbacher, 2000; Stedman et al., 1998; Tinazzi and Zanette, 1998). Excitability changes seen in the resting contralateral homologous muscle using transcranial magnetic stimulation (TMS) are augmented with increasing contraction force during isometric contractions (i.e. Perez and Cohen, 2008, 2009). Voluntary rhythmical contractions also enhance excitability to the contralateral resting limb when the homologous muscle is active, which increases with increasing contraction force (Carson et al. 2004; Ibey and Staines, 2013). To date, the transcallosal influence of sensorimotor integration during rhythmical movement and force manipulations has not been researched.

It is well established in the literature that sensory input from one arm can modulate the excitability of motor cortical projections to muscles in the same limb. Proprioceptive input has a major impact on motor control at both the spinal and cortical level (Gandevia, 2001; Wiesendanger & Miles, 1982). Motor neuron firing rate may be reduced by 30% in the absence of afferent feedback (Macefield et al., 1993). Peripheral nerve electrical stimulation can modulate afferent input and induce neuroplastic changes in motor cortical areas (Kaelin-Lang et al., 2002;

Ridding et al., 2000); this suggests projections from the somatosensory cortex modulate motor cortical excitability.

In the motor cortex, various forms of deafferentation, including peripheral nerve lesions (Kolarik et al., 1994) and limb amputations (Chen et al., 1998; Qi et al., 2000) results in reorganization of the motor representation in the deafferented hemisphere (Cohen et al., 1991). Most reports concentrate on the effects of deafferentation on contralateral cortical representations, however decreased somatosensory input also elicits changes in the ipsilateral hemisphere. Acute deafferentation of a limb results in bilateral cortical reorganization. This may be driven by transcallosal connections between homologous muscle groups.

Many studies have used transcranial magnetic stimulation (TMS) to evaluate the impact of afferent sensory input on excitability of the human motor cortex (Rosenkranz & Rothwell, 2003). It is well known that sensory input from muscle vibration (MV) can modulate excitability of the muscles in the same limb (Claus et al., 1988; Kossev et al., 1999; Rosenkranz et al., 2000; Siggelkow et al., 1999). Modulation of afferent input through MV mainly excites 1a fibers of muscle spindle afferents (Burke et al., 1976; Gandevia, 1985; Martin & Park, 1997; Roll et al., 1989). This effect has been seen in both the hand and forearm muscles and appears to be somatotopically organized where excitability changes are seen in vibrated muscles and not in antagonistic muscles (Claus et al., 1988; Kossev et al., 1999; Rosenkranz & Rothwell, 2003; Siggelkow et al., 1999).

More recently research has explored whether manipulation of sensory input can influence the excitability of projections to muscles in the contralateral resting

limb. Enhancing sensory input through MV can have effects on excitability of corticospinal projections to the opposite limb (Kossev et al., 2001). Augmentation of MEPs did not reach significance in the homologous muscle, however MEPs in the contralateral antagonist were significantly reduced (Kossev et al., 2001). The facilitatory action of MV is most likely mediated via transcallosal pathways causing inhibition of cortical outputs to the contralateral antagonist. Previous research has demonstrated changes in excitability of short and long interval intracortical inhibition (SICI, LICI) during vibration suggesting that these effects may involve the GABAergic system which is thought to mediate these mechanisms (Rosenkranz & Rothwell, 2003; Ziemann et al., 1996). The cross excitability changes of muscle vibration during dynamic movement have not been researched.

Interhemispheric control is important in coordination of bimanual as well as unimanual voluntary movements (Geffen, Jones, & Geffen, 1994). Unilateral afferent inputs modulate excitability of both contralateral and ipsilateral motor cortical projections, which may become useful where abnormal interhemispheric inhibition is a potential therapeutic target. Here we will examine if there is an additive effect with muscle vibration during rhythmical movement with and without added force. In a previous study (Ibey & Staines, 2013) we established cross excitability changes during rhythmical isotonic contractions, and now we will see if additional sensory input enhances this effect. While there is an increase in excitability to the resting homologous muscle at rest (Kossev et al., 2001), we are unsure how the convergence of sensorimotor information will behave with movement, force and

sensory manipulations. We hypothesized that MV will increase excitability to the contralateral resting homologous muscles.

3.3 Materials and Methods

3.3.1 Participants

Nine right-handed healthy volunteers (6 females, 3 males) with an average age of 23.9 ± 4.6 participated in this study. Handedness was confirmed using the Edinburgh Inventory (Oldfield, 1971). All participants had no contraindications to TMS or any known neurological impairments. Participants gave their informed written consent to participate in the studies and completed a screening form for TMS. The Office of Research Ethics at the University of Waterloo approved all of the experimental procedures.

3.3.2 Experimental Setup

Participants were seated in an armchair with their forearms fully supported on an adjustable table. Arms were positioned 20-30° from their torso with both forearms in 90-100° of flexion. The right forearm rested in a neutral (semiprone) position during the entire experiment. The left forearm was secured semiprone in a custom-made manipulandum with the hand fastened midpalm thereby isolating wrist movement and eliminating grip (Figure 3.1). A rotating shaft and potentiometer was located above the wrist joint axis allowing for full extension and flexion range of motion. The rotating shaft is equipped with a wheel where resistance can be added using a fastener, providing tension in both directions of movement. The potentiometer monitored displacement of the wrist joint through

custom SIGNAL Software (Power 1401, Cambridge Electronic Design, Cambridge, UK) and was used to trigger TMS stimulation during active or passive wrist movements. A metronome provided auditory cuing for movements of the left wrist. The tones were presented to the right of the participant (opposite to the side of the TMS stimulation).

Figure 3.1. Experimental set-up. Overhead view of the experimental set-up where subjects were instructed to perform rhythmical flexion and extension at the left wrist while they were fixed in a custom made manipulandum. The right arm was at rest in all experimental conditions. TMS was over the left motor cortex targeting the right resting ECR motor representation.

At the start of the experiment the subject performed three trials of maximum voluntary isometric contractions for both wrist flexion and wrist extension. Maximum torque (MaxTorque) was used as an estimation of maximum force output (MaxForce). MaxForce was measured using a digital force/torque gauge (Chatillon Force Measurement, Greensboro, NC, USA) recording their MaxForce in Newtons. Subjects alternated between wrist flexion and extension MaxForce acquisition with

a rest period between each contraction (~20s). The MaxForce for both wrist flexion and extension was averaged and the lesser of the two was used to calculate 20% MaxForce for the experimental conditions (resistance on the manipulandum has to be the same in both directions). MaxForce was continually monitored using electromyography over extensor carpi radialis longus (ECR) and flexor carpi radialis (FCR) in SIGNAL Software (Power 1401, Cambridge Electronic Design, Cambridge, UK). We will term no added resistance as no force and 20% MaxForce as force condition.

3.3.3 Electromyography

Electromyographic (EMG) activity of ECR, FCR and first dorsal interosseous (FDI) was recorded from both upper limbs using two surface electrodes placed longitudinally over the muscle bellies with the ground electrode over the right styloid process of the ulna. EMG signals were amplified (1000x), filtered (bandpass 2-2500 Hz) (Intronix Technologies Corporation Model 2024F, Canada) and digitized at a sample frequency of 5 kHz by an analog-to-digital interface (Micro1401, Cambridge Electronics Design, Cambridge, UK). All EMG data was stored on a PC for off-line analysis.

3.3.4 Transcranial Magnetic Stimulation (TMS)

Motor evoked potentials (MEPs) were collected from the resting right arm by stimulating the left motor cortex. Single and paired pulse TMS was delivered using two custom built 50mm inner diameter figure-of-eight branding coils connected to two Magstim 200^2 stimulators (Magstim, Whitland, UK). Coil placement and orientation was continuously monitored using BrainSight (BrainSight 2; Rogue

Research, Montreal, QC, Canada), a TMS neuronavigation system that displays real-time coil placement and target location on an anatomical magnetic resonance image (MRI). The motor hotspot for the ECR in M1 was acquired by placing the stimulation coil on the scalp at a 45° angle to the mid-sagittal plane. The motor hotspot was determined to be the location in M1 that elicits an optimal MEP in the contralateral resting ECR. Resting motor threshold (RMT) was defined as the lowest stimulation intensity where potentials with peak-to-peak amplitude of a minimum of 50μV were evoked in at least five out of ten trials using the Magstim 200^2 stimulator (Magstim, Whitland, UK). TMS was triggered externally using SIGNAL Software and a Cambridge Electronic device (Power 1401, Cambridge Electronic Design, Cambridge, UK). MEPs, short-interval intracortical inhibition (SICI), and long-interval intracortical inhibition (LICI) were recorded for all conditions from the left motor cortex. For MEPs, 10 individual TMS pulses were applied over the left M1 and the intensity was 120% of RMT. For SICI, both the conditioning and test stimuli were applied over M1 with the same coil connected to a Magstim 200^2 stimulator operating via a Bistim module. SICI was performed with a subthreshold conditioning stimulus (CS) followed by a suprathreshold test stimulus (TS) to the M1 hotspot for ECR. The interstimulus interval (ISI) for SICI was 2.5ms to produce intracortical inhibition (Di Lazzaro et al., 2006; Kujirai et al., 1993). The CS was set at 80% of RMT for SICI. LICI uses a suprathreshold CS and TS of 120% RMT with an ISI of 100ms (Chen, 2004).

3.3.5 Muscle Vibration

Continuous muscle vibration (frequency 80Hz) was applied using a piezoelectric vibration device with a 1cm diameter probe. The muscle belly of the ECR was vibrated just distal to the lateral epicondyle. For each participant, their perceptual threshold was determined (lowest amplitude that they could feel the vibration) and the amplitude of vibration was set to two times that value. If there was illusory movement, the amplitude of the vibration was adjusted individually to be just below the threshold for perceiving illusory movement (Gilhodes, Roll, & Tardy-Gervet, 1986; Roll & Gilhodes, 1995; Roll et al., 1989).

3.3.6 Experimental Procedures

3.3.6.1 Effect of vibration on isotonic contraction

Participants were instructed to perform rhythmic flexion and extension of their left wrist, using their full range of motion, to the beat of an auditory metronome at a frequency of 0.5 Hz. The metronome ran at a frequency of 1 Hz so subjects could coordinate maximum flexion and extension with each beat. Each trial (~2 minutes) was performed separately for each condition (no force, force and passive). TMS was triggered within both flexion and extension phase of movement (1 target position within each movement phase). TMS was triggered 10 times in each direction for the single pulse and LICI conditions with vibration and without vibration. For the SICI conditions TMS was triggered 20 times in each direction (10 CS and 10 TS) with and without vibration. All the TMS conditions were pseudo-randomized. For the force condition the resistance was applied prior to the trial and EMG was monitored within SIGNAL Software (Power 1401, Cambridge Electronic

Design, Cambridge, UK). Resistance was re-measured at the end of each trial to ensure the resistance level remained consistent throughout the condition.

3.3.6.2 Effect of vibration on passive movement

Participants were secured semiprone in the same custom-made manipulandum used for the isotonic conditions with the hand fastened midpalm, thereby isolating wrist movement and eliminating grip. A researcher rhythmically moved the participants left wrist through flexion and extension to the beat of an auditory metronome at a frequency of 0.5 Hz. The metronome ran at a frequency of 1 Hz so the researcher could coordinate maximum flexion and extension with each beat. MEPs were elicited in the right ECR by triggering TMS at the two positions (using same procedure as isotonic conditions as per above). Only single pulse TMS was collected for this condition.

All conditions were pseudo-randomized with each participant. During all conditions experimenters monitored motor activity via EMG in the right resting limb and provided auditory feedback to the participant where necessary to ensure all muscles were silent.

3.3.7 Data Analysis

The EMG data in the resting right arm was examined prior to analysis of the MEPs for all conditions to ensure the muscles were silent. A three-way ANOVA determined the effect of factors condition [rest, no force, force], vibration (with and without) and phase (extension and flexion) on right resting ECR MEPs. *A priori* planned contrasts were conducted to test the hypothesis that facilitation of right ECR will be greatest during activation of the homologous muscles (extension vs.

flexion). When the F value was significant, i.e. main effect of condition, vibration or phase, Tukey's post hoc tests were used to determine the means by which they differed ($p < 0.05$). Additional ANOVAs were conducted when an interaction was present. For paired pulse measures CS amplitudes were expressed as a percent of TS amplitudes within each phase of the movement cycle and the average inhibition was calculated. Separate three-way repeated measures ANOVAs were carried out for each TMS condition. All data sets were tested for normality and were well modeled by a normal distribution.

3.4 **Results**

The average EMG activity for right (resting) ECR during the no force and force conditions was calculated. MEPs were only accepted from trials where the background ECR activity was < 10 μV. Peak to peak amplitude was calculated for all the MEPs (n=10) from the right resting ECR muscle for each condition and each phase for every subject, these were then averaged in order to carry out the analyses.

3.4.1 Effect of vibration on isotonic contraction

Figure 3.2 illustrates right resting ECR MEPs (single pulse) averaged within each phase (flexion vs. extension) during rest, no force and isotonic force conditions, both without vibration and with vibration. Repeated-measures ANOVA revealed a main effect of condition ($F_{2,16} = 13.05$, $p = 0.0004$) and phase ($F_{1,8} = 9.94$, $p = 0.0135$). There was no main effect of vibration ($F_{1,8} = 0.56$, $p = 0.4742$). There was an interaction between condition and phase ($F_{2,16} = 4.78$, $p = 0.0236$). Post hoc analysis showed a significant difference between all conditions and a significant

difference between each phase (flexion and extension)($p \leq 0.05$). Confirming previous results (Ibey & Staines, 2013), excitability was greatest when the homologous muscle was engaged. To test for the two-way interaction, a two-way ANOVA was performed with vibration collapsed across all conditions. Repeated-measures ANOVA again revealed a main effect of condition ($F_{2,16} = 13.05$, $p = 0.0004$) and phase ($F_{1,8} = 9.94$, $p = 0.0135$) and the interaction was still present between condition and phase ($F_{2,16} = 4.78$, $p = 0.0236$). To test the pre-planned hypothesis that there would be increased cross excitability in the vibration condition at rest, a contrast was run between vibration and no vibration at rest. They were not significantly different ($F_{1,8} = 4.12$, $p = 0.0769$).

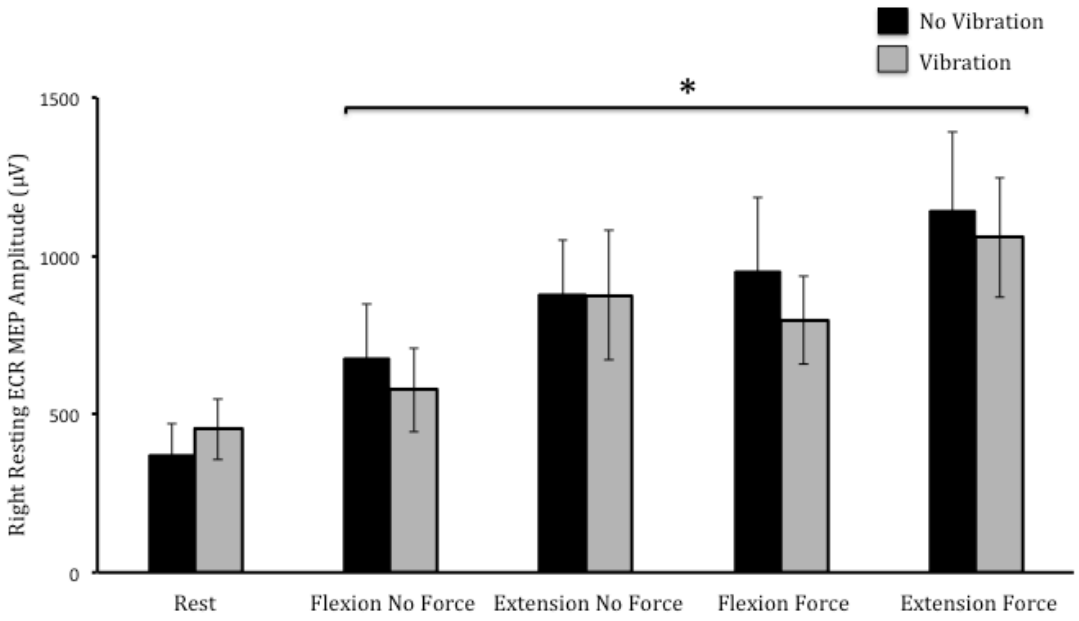

Figure 3.2. Mean effect of rhythmical movement of the contralateral wrist at two different force levels, with and without vibration, on the motor evoked potentials induced by TMS. The mean (n=10) motor evoked potentials (MEPs) in right (resting) ECR at each phase position with and without vibration. Error bars represent standard error of the mean * $p<0.05$.

Figure 3.3 represents right resting ECR MEPs during SICI averaged within each phase (flexion vs. extension) during rest, no force and isotonic force conditions, both without vibration and with vibration are displayed. MEPs are represented as the conditioned stimulus divided by the test stimulus. A three-way ANOVA showed no main effects of vibration ($F_{1,8} = 1.70$, $p = 0.2287$), condition ($F_{2,16} = 0.42$, $p = 0.6612$) or phase ($F_{1,8} = 0.39$, $p = 0.5485$) and no interactions were present. Figure 3.4 represents right resting ECR MEPs during LICI averaged within each phase (flexion vs. extension) during rest, no force and isotonic force conditions, both without vibration and with vibration are displayed. MEPs are represented as the conditioned stimulus divided by the test stimulus. A three-way ANOVA showed a main effect of condition ($F_{2,16} = 3.81$, $p = 0.0443$), however there was no main effect of vibration ($F_{1,8} = 0.00$, $p = 0.9625$) or phase ($F_{1,8} = 0.91$, $p = 0.3690$). Post-hoc analysis on type revealed no significant difference between conditions ($p<0.05$).

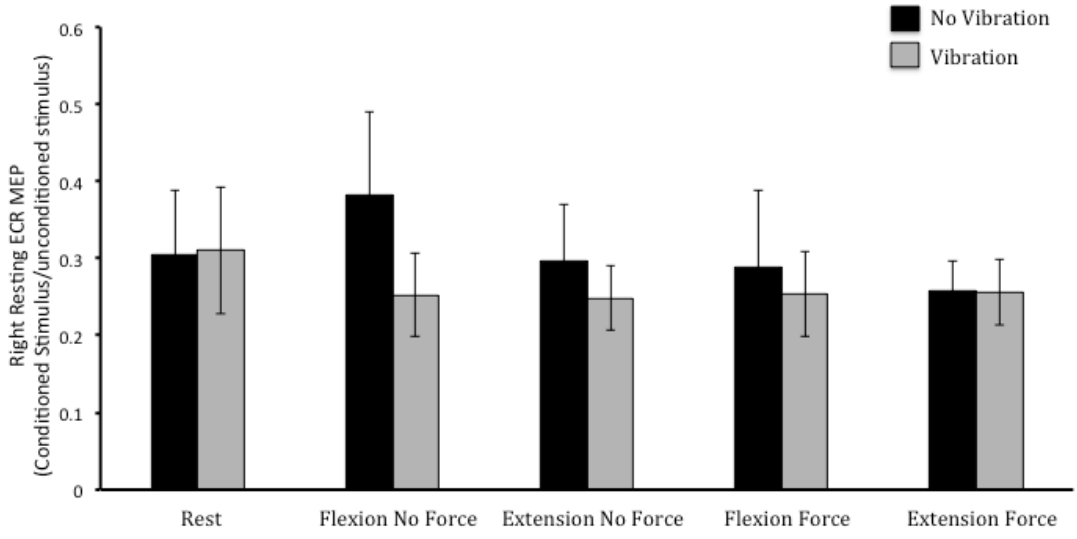

Figure 3.3. Mean effect of rhythmical movement of the contralateral wrist at two different force levels, with and without vibration, on SICI. The mean (n=10) motor evoked potentials (MEPs) in right (resting) ECR at each phase position with and without vibration are expressed as conditioned stimulus/test stimulus amplitude.

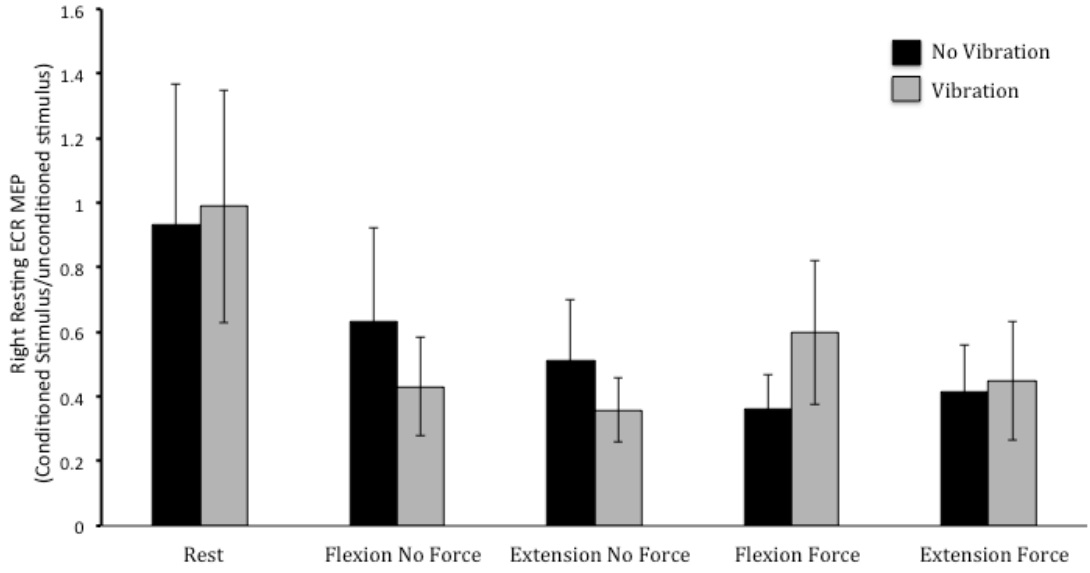

Figure 3.4. Mean effect of rhythmical movement of the contralateral wrist at two different force levels, with and without vibration, on LICI. The mean (n=10) motor evoked potentials (MEPs) in right (resting) ECR at each phase position with

and without vibration are expressed as conditioned stimulus/test stimulus amplitude.

3.4.2 Effect of vibration on passive movement

Figure 3.5 illustrates right resting ECR MEPs during passive movement with and without vibration to the left ECR muscle belly. A three-way ANOVA was completed, comparing rest, no force and passive movement conditions for both phase positions and with and without vibration. There was no main effect of vibration ($F_{1,8} = 0.56$, $p = 0.4742$), however there was a main effect of condition ($F_{2,16} = 13.05$, $p = 0.0004$) and phase ($F_{1,8} = 9.94$, $p = 0.0135$). There was also an interaction between type and phase ($F_{2,16} = 4.78$, $p = 0.0236$). Post hoc analysis showed a significant difference between the no force condition and the passive and rest condition, however no significant difference between passive and rest was found ($p<0.05$). There was no significant difference between passive rhythmical movement with no vibration and passive rhythmical movement with vibration, however there was a significant effect of phase in the vibration condition ($p = 0.023$). There was more excitability during the extension phase compared to the flexion phase.

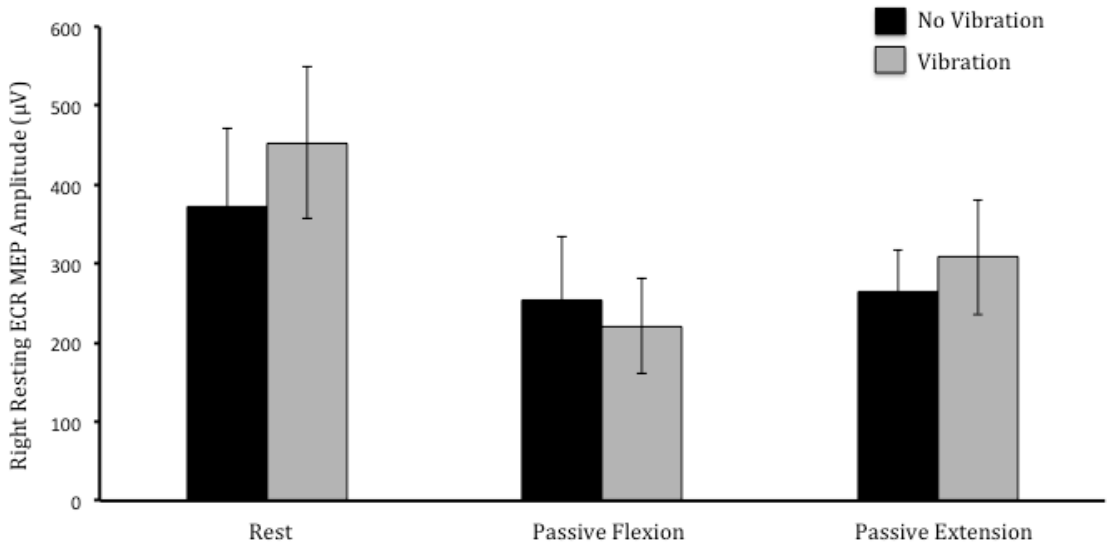

Figure 3.5. Mean effect of passive rhythmical movement of the contralateral wrist, with and without vibration. The mean (n=10) motor evoked potentials (MEPs) in right (resting) ECR at each phase position with and without vibration.

3.5 Discussion

The focus of this study was to investigate how the primary motor cortices interact with both sensory and motor manipulations during rhythmical movement. We looked at changes in corticomotor excitability in the hemisphere ipsilateral to the active hand that performed rhythmical isotonic movement (no force and force conditions) with and without added vibration to the ECR muscle belly on the active limb. Our findings demonstrate what we have found previously, voluntary rhythmic flexion and extension of the left wrist increases MEPs evoked in the resting right ECR when the homologous muscle (extensors) is active; this enhancement of the MEPs is amplified as the required contraction force increases. MEPs are also increased (to a lesser degree) when the non-homologous muscle (FCR) is active. Vibration to the ECR muscle belly did not have a significant effect on excitability

changes to the contralateral resting limb. Vibration also had no effect on intracortical inhibitory mechanisms (SICI and LICI). To our knowledge, this is the first study to investigate the effect of force and vibration during rhythmical isotonic contractions on the corticomotor excitability to the resting limb.

3.5.1 Effect of rhythmical isotonic movement on cortical excitability

TMS activates corticospinal neurons both directly and transynaptically, so the MEP amplitude measured over the muscle belly is reflective of excitability along the entire motor pathway, including neurons in the motor cortex and spinal motor neurons (Rothwell et al., 1991). This study confirms previous findings that corticomotor excitability induced in the right resting ECR was enhanced during isotonic rhythmical flexion and extension of the opposite limb. Excitability was greatest when the homologous muscle was active and went up with increased force of contraction. Carson and colleagues (Carson et al., 2004) previously found MEPs in the right resting FCR (target muscle) were potentiated during the phases of movement in which the left FCR was most strongly engaged during rhythmical flexion and extension of the left wrist. These results increased support for cross facilitation between the motor cortices during homologous muscle activation. Former work by our group took this research a step further and investigated the effect of added force during rhythmical movement (Ibey & Staines, 2013). Our results supported the work by Carson et al. (Carson et al., 2004) and further showed that excitability to the resting ECR increases with increased motor cortical output to the moving limb. Those results supported previous research that showed excitability of a resting muscle increased with increased contraction force of the

contralateral limb (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). MEPs were significantly higher than rest in both our no force and 10% MVC condition during rhythmical movement previously (Ibey & Staines, 2013) and this was also supported by the present results (Figure 3.2). This contrasts findings from Perez and Cohen (2008) who found more prominent facilitation at higher force conditions with no significance between rest and 10% MVC. Perez and Cohen (2008) found no difference at low force, however this was during isometric contractions. Therefore, the overall balance of interhemispheric inhibition may be reduced during rhythmical movements.

Rhythmical movements recruit many brain regions, including M1, somatosensory cortex (S1), premotor cortex, supplementary area, and cerebellum (Schaal, Sternad, Osu, & Kawato, 2004). The afferent input to the muscle during rhythmical movement may explain not only the increase in excitability at low levels of force, but also the increase seen when the non-homologous muscle is active (Figure 3.2 and (Ibey & Staines, 2013)). The excitability of the corticospinal pathway to the resting right ECR increased, to a lesser degree, when the non-homologous muscle was active and went up with increasing force of contraction. Hortobagyi and colleagues (2003) found an increase in excitability when the non-homologous muscle was active at higher levels of force during isometric contractions. The augmentation of MEPs in the resting right ECR during non-homologous muscle activation and at low levels of force during rhythmical movement may be attributed to afferent input, however the passive movement condition did not show an

increase in cortical excitability (Figure 3.5). Voluntary cortical drive appears to be required to drive these changes, which is why we attempted to upregulate afferent input with muscle vibration to test if this would further increase cortical excitability to the resting limb.

3.5.2 Modulation of Ia afferents with muscle vibration

Mechanical vibration of muscles or tendons is known to excite muscle spindles which activate Ia muscle afferents (Burke et al., 1976; Roll et al., 1989). There are excitatory projections of Ia muscle afferents to the somatosensory cortex (i.e. Jones & Porter, 1980). Further to this, animal research has shown topographical and functionally specific corticocortical excitatory connections between somatosensory areas and frontal motor areas such as the primary motor cortex (Murphy, Wong, & Kwan, 1974). Kossev and colleagues (1999) found an almost two-fold increase in MEP amplitude in response to TMS after low amplitude vibration of ECR at 80 Hz; this result has been confirmed by other groups while also showing nearby muscles display a suppression of excitability (Rosenkranz & Rothwell, 2003; Siggelkow et al., 1999). There is a frequency effect of vibration, where the strongest effect is normally seen around 75-80 Hz (Rosenkranz & Rothwell, 2003; Steyvers, Levin, Van Baelen, & Swinnen, 2003). Kossev and colleagues (1999) studied the effects of tendon vibration on corticospinal excitability by comparing MEP amplitude evoked by TMS and transcranial electrical stimulation (TES) which allows differentiation between intracortical and subcortical areas (Day et al., 1987; Rothwell, 1997). There was no effect with TES resulting in the assumption that the

excitability changes seen are driven cortically (Kossev et al., 1999). MV appears to be a good way to increase motor cortical activation in the contralateral cortex.

In the ipsilateral cortex augmentation of MEPs did not reach significance in the homologous muscle, however MEPs in the contralateral antagonist were significantly reduced (Kossev et al., 2001). Our results did show a significant difference between vibration and no vibration conditions at rest, however there was no effect of vibration in the isotonic contraction conditions. In fact, in the flexion phase when the discharge of muscle spindle afferents from the extensor group would be greatest, vibration conditions show a decrease in excitability compared to no vibration (not significantly different). While vibration did not show any added benefit to cross facilitation changes during rhythmical movement conditions, it still did increase excitability above resting conditions and may have some added benefit to long-term neuroplasticity that cannot be ruled out. There have been human studies that have confirmed the functional significance of both somatosensory afferents and motor intracortical circuits in inducing motor cortical plasticity (Ridding & Taylor, 2001; Stefan, Kunesch, Cohen, Benecke, & Classen, 2000). The relationship between motor cortical plasticity and somatosensory afferents requires further investigation.

3.5.3 Modulation of Ia afferents with passive rhythmical movement

Muscle spindles are sensitive to changes in both the muscle length and the velocity of the length of change. Quick and tonic stretch of the muscle spindle is monitored by Ia afferents. In humans, Ia afferent input can alter corticospinal excitability (Carson & Riek, 2000; Carson et al., 1999). Passive movement gives us a

means to evaluate the effect of afferent input on corticomotor excitability. Neuroimaging studies have shown M1 and SMA are involved in the processing of afferent signals during cyclic passive wrist and elbow movements which are predominantly mediated by Ia afferents (Lewis & Byblow, 2002). Previous studies have looked at the effect of passive movement on the contralateral M1 and have shown phasic modulation in MEP excitability (Carson et al., 1999; Coxon et al., 2005; Lewis & Byblow, 2002; Lewis et al., 2001; Stinear & Byblow, 2002) which does not appear to transfer to the ipsilateral M1. Our findings and past research show a depression in MEP amplitude in the ipsilateral cortex during passive movement, however the phasic modulation is present without significant differences between the phases (Figure 3.5) (Carson & Riek, 2000; Ibey & Staines, 2013). There was no significant difference between rhythmical passive movement conditions with and without vibration in this current study, however there was a significant difference between the flexion and extension phase during passive movement when the ECR muscle belly was vibrated. In this case excitability was higher during the extension phase of movement. It appears that there may be more interhemispheric inhibition during passive movement conditions resulting in no cross excitability, however this would need to be confirmed in a future study using interhemispheric inhibition protocols. In our experiment, excitability is lower than rest during the passive movement conditions, but increasing afferent input with vibration appears to have an effect on the target muscle.

3.5.4 Pathways medicating changes in spinal and cortical excitability

MEPs elicited from TMS reflect excitability of neurons in the motor cortex, however they are influenced by both subcortical activity and the state of the spinal motor neurons. Carson and colleagues (Carson et al., 2004) found potentials elicited by stimulating the descending tracts at the level of the cervicomedullary junction are unaffected by rhythmical movement. This has also been found with isometric contractions at increased level of force output (Hortobagyi et al., 2003). This cortical facilitation may be due to transcallosal connections to the homotopic area of the contralateral cortex (Hanajima et al., 2001). There may be a decrease in interhemispheric inhibition (IHI) between the motor cortices. IHI represents transcallosal connections that are mediated by glutamatergic projections acting through $GABA_B$ interneurons (i.e. Meyer et al., 1995). Perez and Cohen (2008) found decreases in IHI from the active to the resting hemisphere during unimanual wrist flexion at 30% and 70% MVC compared to rest and 10% MVC. Uehara and colleagues (2013) looked at rhythmical movements of the finger and found LICI was significantly modulated depending on the frequency, however no changes were seen in SICI or ICF. Our results did not show an effect of SICI. There was a main effect of type of condition, but there was not a significant effect with post hoc analysis.

Intracortical inhibition has been shown to be reduced for the vibrated hand muscle, whereas it is enhanced for adjacent non-vibrated muscles in the contralateral cortex (Binder, Kaya, & Liepert, 2009). Rosenkranz and Rothwell (2003) found amplitude of the MEP increased and SICI decreased to the vibrated muscle, while amplitude of the MEP decreased in the non-vibrated muscle and SICI increased. More research has investigated the effects of MV to the contralateral

hemisphere, however not as much is known about the neurophysiology in the ipsilateral hemisphere. Kossev and colleagues (2001) did find a non-significant augmentation of MEPs in the ipsilateral homologous muscle representation (and a significant decrease in the non-homologous muscle) during vibration. They looked at the same target muscle – ECR. Swayne and colleages (2006) on the other hand found a reduction in cortical excitability in the homologous muscle representation, but they were looking at the hand in this study. They also found an increase in SICI and IHI. There was no effect of vibration on either SICI or LICI in our study.

3.5.5 Significance of findings and clinical implications

We investigated ways in which the primary motor cortices interact during rhythmical movement at increasing load requirements with and without vibration applied to the muscle of interest. The present findings support previous findings that cross facilitation between the active and the resting M1 is dependent on the phase of contraction and the amount of force output. This research also supports previous research in our lab that excitability also increased when the non-homologous muscle was active during rhythmical movement. The addition of sensory input to the muscle of interest on the active side did not have any effect on the amount of cross facilitation to the resting limb.

Despite our lack of findings with the addition of sensory input, improvements in contralateral muscle strength can occur when muscle contractions are evoked by electrical stimulation (Hortobágyi, Scott, Lambert, Hamilton, & Tracy, 1999). Afferent input, which can be driven by crossed effects at cortical, subcortical, brainstem, propriospinal or segmental levels may drive these improvements in

motor function. Human studies have confirmed the functional significance of these connections, both somatosensory afferents and motor intracortical circuits, in inducing motor cortical plasticity (Ridding & Taylor, 2001; Stefan et al., 2000). Therefore, even though there were no additional excitability changes seen with muscle vibration during movement in this study, we cannot make any conclusions on the long-term effect of afferent input during motor tasks. The contralesional M1 plays a critical role in the recovery of motor function on the affected side post-stroke. Continuing to expand our knowledge on sensorimotor integration and the relationship between the hemispheres may help to improve neurorehabiliation and influence neuroplasticity post brain injury.

Chapter 4: Enhancing excitability of the primary motor cortex: the influence of somatotopy and convergence of multiple effectors

4.1 Overview

The aim of this study was to investigate interhemispheric interactions and somatotopic relationships influencing excitability of the left primary motor cortex (lM1) and to determine how coordination of multiple effectors influences lM1 excitability. Transcranial magnetic stimulation (TMS) induced motor evoked potentials (MEPs) were recorded from the resting right extensor carpi radialis (ECR) during two separate experiments. Experiment 1 examined the effect of somatotopy by having the participant perform separate unimanual isometric contractions at 10% maximum voluntary contraction (MVC) of different upper and lower limb muscles both ipsilateral (ipsi) and contralateral (contra) to the hemisphere of interest (lM1). Experiment 2 assessed the influence of convergence on lM1 excitability by having participants contract the right tibialis anterior (TA) contra to lM1 in conjunction with the homologous muscle (left ECR). The force of contraction of left ECR was kept constant at 10% MVC, while the right TA was tested at both 10% and 30% MVC. In Experiment 1, MEPs were potentiated in the right resting ECR when the homologous muscle was engaged, however no other muscles ipsi to lM1 had an effect on excitability. All muscles contra to lM1 (upper and lower limb) increased excitability to the same extent as the ipsilateral homologous muscle. In Experiment 2, results showed recruitment of multiple effectors augmented excitability more than the homologous muscle alone and the force of contraction of the lower extremity also had an influence on excitability changes. Our findings indicate that the increase in corticomotor drive seen during both Experiment 1 and 2 is likely mediated upstream of M1 in non-primary motor areas (i.e. SMA).

Improving our understanding of these cortical networks can be useful in the development of future neurorehabilitation methods.

4.2 Introduction

Many studies have looked at the distribution of callosal connections between motor representations within the primary motor cortex (M1). In the upper limb, transcallosal connections are more numerous between proximal musculature and this is also true in the supplementary motor area (SMA) (Gould et al., 1986; Rouiller et al., 1994). The literature has put more emphasis on the interhemispheric interactions and functional role of distal upper limb musculature, whereas less emphasis has been devoted to the interhemispheric interactions of proximal limb muscles.

Researchers have demonstrated activity-dependent changes in the primary motor cortex ipsilateral to the moving limb (Carson, 2005). Unilateral motor practice increases motor output to both the trained and untrained limb, a phenomenon known as cross education (Carroll et al., 2008). Many have hypothesized that cross education is mediated by transcallosal interactions between M1 regions. Excitability of corticomotor projections to the resting limb increase with contraction of the homologous muscle (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). This has been studied extensively with both isometric and isotonic contractions (Carson et al., 2004; Ibey & Staines, 2013). Both Ibey & Staines (2013) and Hortobagyi and colleagues (2003) found non-homologous muscle activation

also contributed to these cross excitability changes. This begs the question of whether somatotopy and spatial location of muscle representations have an influence on cross excitability changes in the forearm.

Excitability changes in the resting hemisphere exist when the contralateral homologous and non-homologous muscle are engaged, but also increase when the remote segment is active (remote effect). Motor excitability is increased in the upper limb as assessed by transcranial magnetic stimulation (TMS) during contraction of the lower limb (Hortobagyi et al., 2003). Voluntary rhythmic flexion and extension of the foot causes cyclic modulation of excitability of M1 to the resting forearm (Baldissera, Cavallari, & Leocani, 1998). Many thought the afferent signals generated by the foot movement influenced spinal excitability at the level of the cervical spine; however, cyclic modulation of the H-reflex in the resting forearm was not related to movement, and it was temporally bound to activation of the foot movers (Cerri, Borroni, & Baldissera, 2003). Excitability changes during activation of the remote segment are likely driven at a central level.

The homuncular sequence of body representations has held true in depicting locations of gross segments, such as upper and lower limb (Grafton, Woods, Mazziotta, & Phelps, 1991). However, debate surrounds the depiction of smaller within-limb representations, for example fingers, elbow and shoulder within the upper limb. The debate involves whether within limb segments overlap with each other, or maintain somatotopically-defined locations. Functional magnetic resonance image (fMRI) studies have shown that somatotopic gradients exist in M1 despite the overlap of representations. The overlap within M1 is believed to allow

for multi-joint coordination, while maintaining homuncular order is thought to contribute to fine individual muscle control (Plow et al., 2010). Further to this, within the primary motor cortex there are no known anatomical connections between the arm and leg muscle representations (Brown et al., 1991; Huntley & Jones, 1991). Common input to these motor regions likely arises from secondary motor areas, rather than horizontal connectivity within the primary motor cortex (Murthy & Fetz, 1996). Fink and colleagues (Fink, Frackowiak, Pietrzyk, & Passingham, 1997) found arm and leg regions to overlap considerably within secondary motor areas (dorsal and ventral premotor cortices and supplementary motor area). The question of how somatotopy and contraction of different upper and lower limb muscles influences corticomotor excitability of an individual forearm muscle is not yet known. Understanding how these corticomotor projections are influenced by nearby muscle activity will provide a better understanding of the neural mechanisms influencing unimanual movement and contributing to modulation of motor cortical excitability.

The goal of this study was to investigate interhemispheric interactions and somatotopic relationships influencing excitability of the left M1 and to determine how coordination of multiple effectors influences M1 excitability. This was broken down into two experiments. The first experiment explored interhemispheric interactions and somatotopic relationships during both ipsilateral and contralateral isometric contractions. The second experiment studied whether movement of the homologous muscle with another effector (lower extremity on the same side as the resting muscle) increases excitability of the resting muscle more than just the

homologous muscle alone (convergence of primary and non-primary motor regions). We hypothesized MEPs would be enhanced during ipsilateral activation of the homologous muscle (ECR) and its functional antagonist (FCR in this case) and would decrease as a function of spatial position during contralateral activation. Further, it was hypothesized that the recruitment of dual effectors (i.e. hand-foot) would increase M1 excitability more than one effector due to the convergence of transcallosal pathways and non-primary motor cortices.

4.3 Materials and Methods

4.3.1 Participants

Experiment 1 and 2 were collected on separate days. Twelve right-handed healthy volunteers (7 male, 5 female) with an average age of 24.9 ± 2.43 years participated in Experiment 1 and eight right-handed healthy volunteers (4 male, 4 female) with an average age of 25.3 ± 3.70 years participated in Experiment 2. Handedness was confirmed using the Edinburgh Inventory (Oldfield, 1971). All participants had no contraindications to TMS or any known neurological impairments. Participants gave their informed written consent to participate in the studies and completed a screening form for TMS. The Office of Research Ethics at the University of Waterloo approved all of the experimental procedures.

4.3.2 Experimental Setup

In both experiments subjects were examined at rest and in tasks that required isometric contractions. The left hemisphere extensor carpi radialis (ECR) motor representation was tested for all participants. In both experiments the

muscle of interest (right ECR muscle) was monitored to ensure it remained quiescent during all conditions. Muscle relaxation of the forearm was continuously monitored using custom LabVIEW software (National Instruments, Austin, TX, USA). Subjects were given auditory feedback by the experimenters who monitored the electromyogram (EMG) signal. Muscle relaxation was confirmed using quantitative off-line analysis.

Participants were seated in a custom armchair with their forearms and feet fully supported. Arms were positioned 20-30° from their torso with both elbows in 90-100° of flexion. Knees were positioned in approximately 70-80° of flexion. Subjects were seated with their arms and feet in a custom-built manipulandum (Figure 4.1). At the beginning of each experiment subjects performed three trials of maximum voluntary isometric contractions for each movement. Maximum force output was measured in LabVIEW (National Instruments, Austin, TX, USA). The average was calculated and percent maximum voluntary contraction was determined for each condition.

4.3.3 Electromyography

Electromyographic (EMG) activity of ECR, FCR, bicep, deltoid and first dorsal interosseus (FDI) was recorded from both upper limbs and TA was also collected bilaterally using bipolar surface electrodes placed longitudinally over the muscle bellies in Experiment 1. Experiment 2 recorded EMG activity from the ECR, FCR and FDI bilaterally and the right TA using bipolar surface electrodes. EMG signals were amplified (1000X), filtered (bandpass 1-200 Hz) and recorded using a SynAmps^2

amplifier and Neuroscan 4.3 (Compumedics, Charlotte, NC, USA) with a sampling rate of 1000 Hz. All EMG data was stored on a PC for off-line analysis.

Figure 4.1. **Experimental set-up for Experiment 1 and 2**. Overhead view of the experimental set-up where subjects were instructed to perform isometric contractions [both ipsilateral (left upper and lower limb) and contralateral (right upper and lower limb) to stimulation] while they were secured in manipulandums. The right ECR (muscle of interest) was at rest in all experimental conditions. Please note: during Experiment 1 more custom build manipulandums were required to secure the various upper and lower extremity muscles that were tested.

4.3.4 Transcranial Magnetic Stimulation (TMS)

Motor evoked potentials (MEPs) were collected from the resting right arm by stimulating the left motor cortex with a MagPro x100 stimulator (Medtronic, Minneapolis, MN, USA) through a figure-eight coil (loop diameter 10 cm; model MC-B70) using a biphasic waveform. Coil placement and orientation was continuously monitored using Brainsight (Brainsight 2; Rogue Research, Montréal, QC, Canada), a TMS neuronavigation system that displays real-time coil placement and target

location on an anatomical magnetic resonance image (MRI). The coil was oriented tangential to the scalp with the handle pointing posterior and 45° away from the midline to activate the corticospinal system.

TMS was delivered over the optimal location to elicit a response in the ECR muscle of the right arm. Resting motor threshold (RMT) was defined as the lowest stimulation intensity where potentials with peak-to-peak amplitude of a minimum of 50 μV were evoked in at least five out of ten trials (Rossini & Rossi, 2007). TMS was triggered externally using a customized LabVIEW program (National Instruments, Austin, TX, USA) at an intensity of 120% of RMT. Along with single pulse TMS, short-interval intracortical inhibition (SICI) was also recorded for all conditions from the left motor cortex. The intensity was 120% of RMT for single pulse conditions. For SICI, there is a subthreshold conditioning stimulus (CS) followed by a suprathreshold test stimulus (TS) to the M1 hotspot for ECR. The interstimulus interval (ISI) for SICI was 2.5 ms to produce intracortical inhibition (Di Lazzaro et al., 2006; Kujirai et al., 1993). The CS was set at 80% and the TS at 120% of RMT for SICI.

4.3.5 Experimental Procedures

4.3.5.1 Experiment 1

The aim of Experiment 1 was to investigate corticomotor excitability of ECR during isolated isometric contractions of both contralateral and ipsilateral upper and lower limb muscles. In this experiment, ipsilateral and contralateral are referring to the muscle location relative to the hemisphere of interest (Figure 4.1). Five muscle groups were collected bilaterally [tibialis anterior (TA), first dorsal

interosseus (FDI), flexor carpi radialis (FCR), bicep (B), deltoid (D)] and one muscle group (ECR) was collected unilaterally only on the left representing the homologous muscle. Subjects performed isometric contractions during each condition at 10% MVC. Muscles were tested individually and a rest condition was also collected. For each condition 20 MEPs were collected from the right resting ECR while participants held an isometric contraction of each individual muscle. Conditions were pseudo-randomized. Subjects monitored their contraction on a computer screen placed in front of them for the visual condition. They also performed a non-visual condition where they received feedback verbally from an experimenter on their level of contraction.

4.3.5.2 Experiment 2

The aim of Experiment 2 was to investigate corticomotor excitability of ECR during the convergence of multiple effectors. Subjects were examined at rest and during six movement conditions: (1) Isometric contraction of left ECR at 10% MVC, (2) Isometric contraction of right TA at 10% MVC, (3) Isometric contraction of right TA at 30% MVC, (4) Isometric contraction of right soleus at 30% MVC, (5) Isometric contraction of left ECR at 10% MVC + Isometric contraction of right TA at 10% MVC, and (6) Isometric contraction of left ECR at 10% + Isometric contraction of right TA at 30%. For each condition 20 single pulse and SICI conditioned MEPs were collected from the right resting ECR. All experimental conditions were pseudo-randomized. Subjects monitored their raw EMG contraction on a computer screen placed in front of them.

4.3.6 Data Analysis

Experimenters examined the EMG data in the resting right arm prior to analysis of the MEPs for all conditions to ensure the muscles were silent. Relaxation of the right ECR and FCR was defined as EMG activity below the mean amplitude of 10 μV. The mean amplitude (rectified) was calculated 100 ms prior to delivery of TMS for all isometric conditions to ensure the muscle of interest (right ECR) was at rest. EMG profiles were also acquired for all the movement conditions for each muscle being collected at the time to calculate the average percent MVC for each condition.

The MEPs elicited in the right ECR during each condition were normalized with respect to the MEPs elicited during the resting trials. Firstly, a one-way ANOVA was used to determine the effect of spatial specificity in Experiment 1 on right resting ECR MEPs. Separate one-way ANOVAs were run for the muscles ipsilateral to M1 of interest and contralateral to M1 of interest. *A priori* contrasts were used to test the hypothesis that facilitation of ECR would be greatest during homologous muscle activation and would also be enhanced during activation of the non-homologous functional antagonist (FCR). Secondly, a one-way ANOVA was used to determine the effect of condition in Experiment 2 on right resting ECR MEPs. There was no difference in Experiment 1 between vision and no-vision MEPs, so these were grouped together. For SICI conditions, unconditioned MEPs are expressed as a percent of conditioned MEPs and the average inhibition was calculated. All data sets were tested for normality and were well modeled by a normal distribution.

4.4 **Results**

EMG activity from the right resting ECR during all conditions along with the average percent MVC for each condition is represented in Table 1 and 2 for Experiments 1 and 2 respectively. Peak to peak amplitude was calculated for all the MEPs (n=20) from the right resting ECR muscle for each condition and for each subject. These were then averaged in order to carry out the analyses.

Table 4.1. EMG activity from the right resting ECR and from all active muscles during their respective conditions during Experiment 1.

	Rest	DF	FDI	ECR	FCR	Bicep	Deltoid
AVG % MVC							
Ipsilateral		13±0.8	7.6±1.4	11.1±1.4	11.0±2.0	10.4±1.4	10.6±0.9
Contralateral		13.2±1.1	13.2±1.5		12.3±1.9	9.7±1.5	10.9±1.0
AVG ECR Activity (μV)							
Ipsilateral	3.6±0.5	3.3±0.4	3.8±0.5	7.2±1.9	3.3±0.6	3.0±0.4	3.0±0.4
Contralateral		3.2±0.5	7.6±2.0		4.7±0.4	5.0±1.7	3.7±0.5

Table 4.2. EMG activity from the right resting ECR and from all active muscles during their respective conditions during Experiment 2.

						L ECR 10 + R DF 30		L ECR 10 + R DF 10	
	Rest	**R PF 30**	**R DF 30**	**R DF 10**	**L ECR 10**	**L ECR 10**	**R DF 30**	**L ECR 10**	**R DF 10**
AVG % MVC		34.9 ±3.7	35.7 ±3.7	14.9 ±1.3	10.5± 1.0	10.8± 0.8	34.8 ±3.7	11.0± 1.0	16.4 ±2.1
AVG ECR Activity (μV)	1.1± 0.1	2.5± 0.7	2.3± 0.4	1.9± 0.5	1.8±0 .3	3.4±0.7		2.1±0.3	

4.4.1 Experiment 1

Figure 4.2 illustrates peak to peak amplitude of right ECR MEPs averaged during each isometric condition carried out individually by muscles on the left

(ipsilateral to hemisphere stimulated). A one-way ANOVA revealed a main effect of condition ($F_{6,66} = 2.76$; $p \leq 0.01$) on MEPs from the resting right ECR. Post hoc analysis showed a significant difference between rest and contraction of the homologous muscle (ECR) ($p \leq 0.05$). There was no significant difference between rest and any other upper or lower limb ipsilateral muscle. Pre-planned contrasts were run between rest and the homologous muscle (ECR) and between rest and the non-homologous direct antagonist (FCR). There was a significant difference in excitability of the resting ECR muscle between rest and homologous muscle activation ($F_{1,66} = 13.76$; $p \leq 0.0004$), however there was not a significant difference between rest and the non-homologous muscle (FCR) ($F_{1,66} = 0.84$; $p \leq 0.36$).

Figure 4.3 illustrates right ECR MEPs averaged during each isometric contraction carried out by muscles on the right (contralateral to stimulated hemisphere), on the same side as the muscle of interest (right ECR). A one-way ANOVA demonstrated there was no effect of condition ($F_{5,55} = 2.19$; $p \leq 0.069$) on MEPs from the resting right ECR. From Figure 4.3 one can see that excitability changes of the resting contralateral ECR was comparable between ipsilateral ECR contraction (homologous muscle) and any isometric contraction on the contralateral side. A one-way ANOVA was run on these conditions (all contralateral isometric contraction conditions and ipsilateral ECR). The ANOVA revealed no effect of condition ($F_{5,55} = 0.19$; $p \leq 0.97$) meaning contraction of any contralateral muscle (on the same side of the muscle of interest) increased excitability of the resting ECR to the same extent as the homologous muscle.

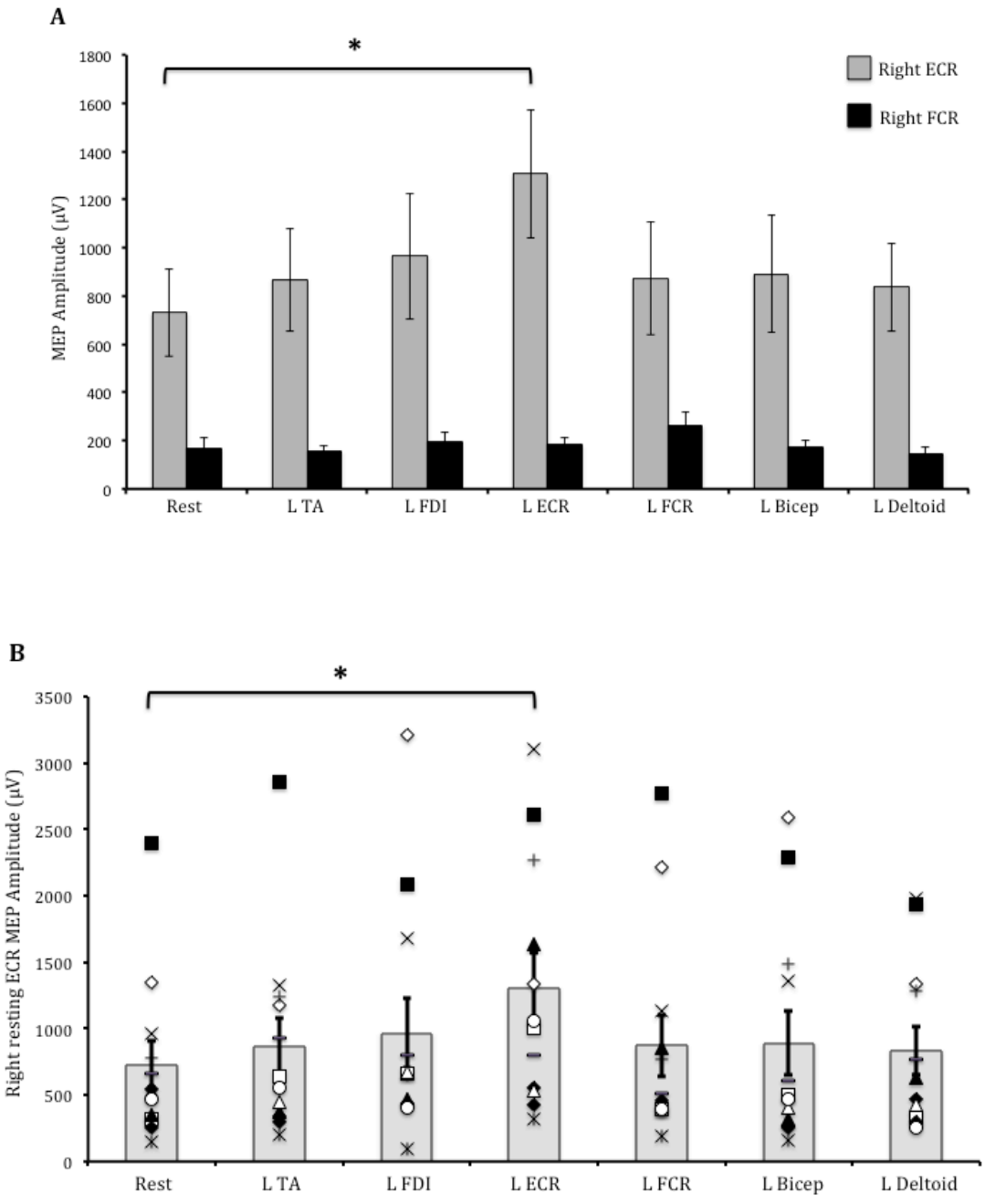

Figure 4.2. Mean effect of isometric contraction of ipsilateral muscles on the motor evoked potentials induced by TMS. The mean (n=12) motor evoked potentials (MEPs) in right (resting) ECR & FCR during contraction of ipsilateral muscles (A). Raw MEPs are represented and individual data is displayed for the right (resting) ECR in (B) for each condition. Error bars represent standard error of the mean. * $p<0.05$

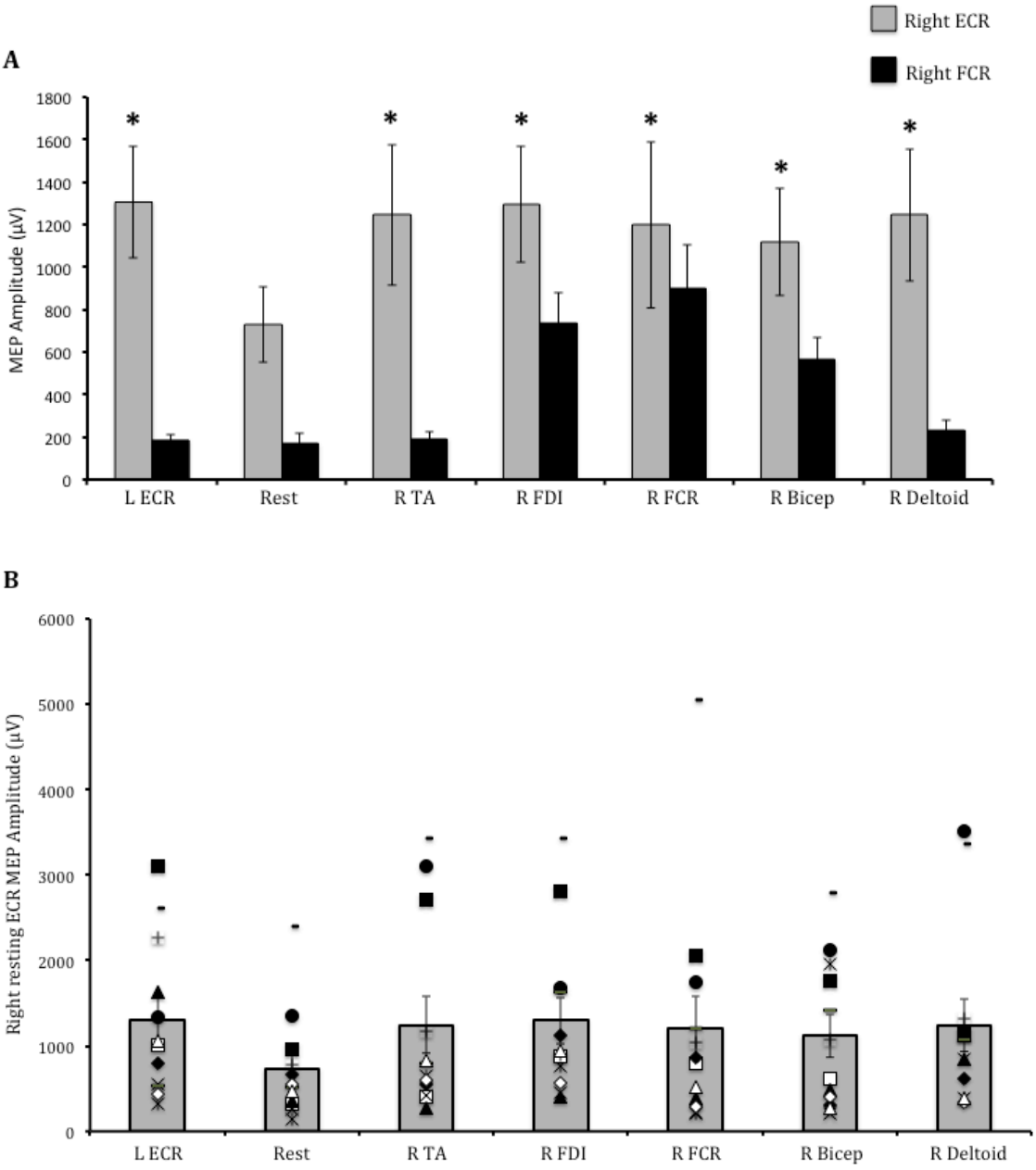

Figure 4.3. **Mean effect of isometric contraction of contralateral muscles and ipsilateral ECR on the motor evoked potentials induced by TMS**. The mean (n=12) motor evoked potentials (MEPs) in right (resting) ECR & FCR during contraction of contralateral muscles and ipsilateral ECR (A). Raw MEPs are represented and individual data is displayed for the right (resting) ECR in (B) for each condition. Error bars represent standard error of the mean. * $p<0.05$

4.4.2 Experiment 2

Figure 4.4 represents the magnitude of MEPs in the right resting ECR during isometric contractions of the homologous muscle (left ECR) and lower extremity muscles on the same side as the muscle of interest. Figure 4.5 illustrates the effect of homologous muscle activation compared to the combination of homologous muscle and recruitment of a lower extremity muscle at two different levels of force on excitability of M1 to the resting right ECR. A one-way ANOVA was run on all conditions (excluding right PF 30%) and revealed a main effect of condition ($F_{5,35}$ = 7.25; $p \leq 0.0001$). Post hoc analysis showed a difference between rest and all of DF at 10%, DF at 30%, DF at 10% + ECR at 10%, and DF at 30% + ECR at 10% ($p \leq 0.05$). Pre-planned contrasts showed no significant difference between L ECR 10% and combination of DF at 10% + ECR at 10% ($F_{1,35} = 0.88$; $p \leq 0.35$), however there was a significant difference between L ECR 10% and combination of DF at 30% + ECR at 10% ($F_{1,35} = 8.87$; $p \leq 0.005$). Contrast between L ECR at 10% and R DF at 10% confirmed what was found in Experiment 1; there was no significant difference between homologous muscle contraction and contralateral muscle contraction ($F_{1,35}$ = 0.65; $p \leq 0.43$). Contrast between L ECR at 10% and R DF at 30% was significant, revealing force of contraction of lower extremity appears to have an effect on resting ECR excitability ($F_{1,35} = 3.94$; $p \leq 0.05$).

Figure 4.6 represents the SICI data for each condition. There was no effect of the experimental conditions in Experiment 2 on SICI.

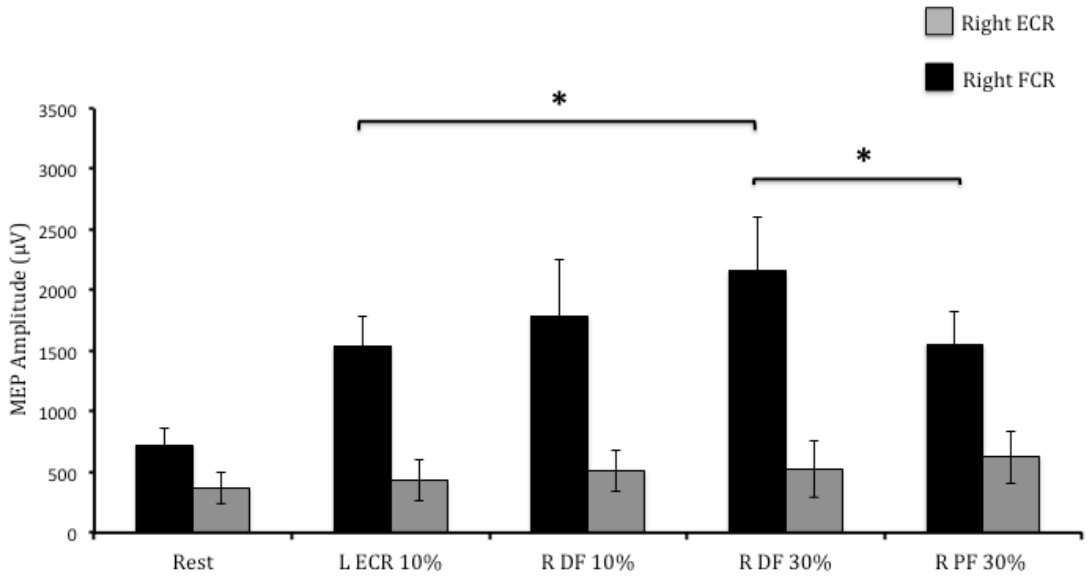

Figure 4.4. **Mean effect of isometric contraction of muscles individually on the motor evoked potentials induced by TMS**. The mean (n=8) motor evoked potentials (MEPs) in right (resting) ECR and FCR during contraction of contralateral lower extremity muscles and ipsilateral ECR. Raw MEPs are represented. Error bars represent standard error of the mean. * $p<0.05$

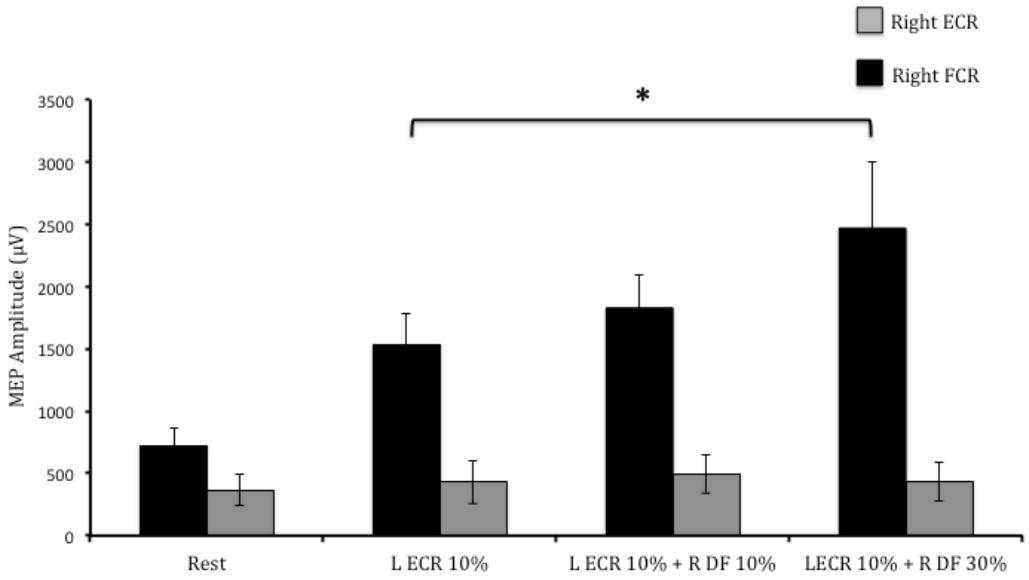

Figure 4.5. **Mean effect of isometric contraction of muscles individually and at the same time on the motor evoked potentials induced by TMS**. The mean (n=8) motor evoked potentials (MEPs) in right (resting) ECR and FCR during contraction of contralateral lower extremity muscles and ipsilateral ECR. Raw MEPs are represented. Error bars represent standard error of the mean. * $p<0.05$

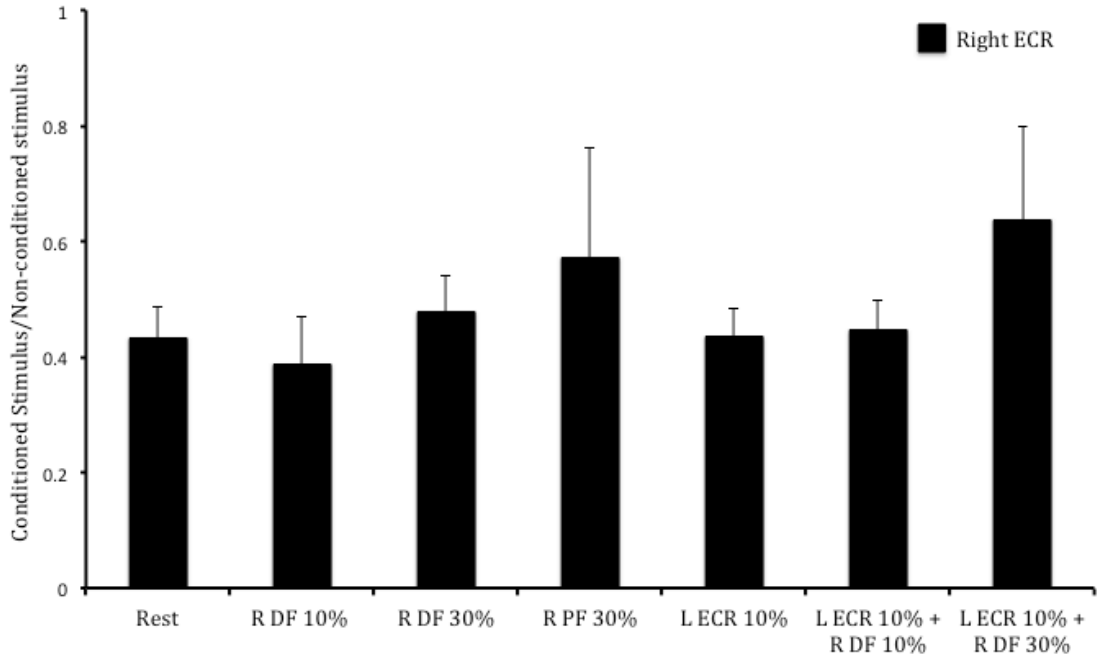

Figure 4.6. **Modulation of SICI during isometric contraction of muscles individually and at the same time**. The mean (n=8) unconditioned single pulse amplitudes at 120% RMT are compared to conditioned stimulus amplitudes. Error bars represent standard error of the mean.

4.5 Discussion

The aim of these studies was to investigate interhemispheric interactions and somatotopic relationships influencing corticomotor excitability within M1 and how recruitment of multiple effectors may influence M1 excitability. Our findings suggest that there does not appear to be an effect of somatotopy or spatial location within or between M1, rather any contralateral muscle (contralateral to TMS stimulation) appears to increase excitability of the muscle of interest (ECR) to the same extent as the ipsilateral homologous muscle. Further, recruitment of multiple effectors (homologous muscle and contralateral lower extremity) augments excitability more than homologous muscle alone and force of contraction also

influences excitability changes. To our knowledge, this is the first study that has investigated somatotopy and spatial location of muscle representations bilaterally and its influence on excitability in one muscle group. It is also the first study that has looked at the convergence of the homologous muscle group with a distal effector.

4.5.1 Effect of somatotopy and spatial location of muscles on M1 excitability

There is debate surrounding whether within limb segments overlap with each other, or maintain somatotopically defined locations. Overlap of various motor representations has been attributed to bidirectional horizontal connections (Huntley & Jones, 1991), convergence of corticospinal input from multiple locations (Donoghue et al., 1992) and divergence of corticospinal output to multiple segments (McKiernan et al., 1998; Shinoda, Zarzecki, & Asanuma, 1979). FMRI studies have shown that somatotopic gradients exist in M1 despite the overlap of representations. The overlap within M1 is believed to allow for multi-joint coordination, while maintaining homuncular order is thought to contribute to individual movement control (Plow et al., 2010). Within the left upper limb we tested FDI, FCR, ECR (homologous muscle), bicep and deltoid. Our results show that at a low level of contraction (10% MVC) there was no significant cross excitability influence on the right resting ECR other than the homologous muscle which has been extensively studied. Past research has reported an increase in excitability with contraction of the non-homologous muscle (Hortobagyi et al., 2003; Ibey & Staines, 2013). In these studies the increase in excitability was seen at higher levels of contraction force in the isometric conditions than was used in this experiment, and also occurred during isotonic movement. We kept the level of contraction low to

eliminate co-contraction of surrounding muscles and isolate the muscles of interest. Within the right upper limb we tested FDI, FCR, bicep and deltoid. Our results show that at 10% MVC all the muscles significantly increased excitability of the right resting ECR, further this increase in excitability was not significantly different than the increase seen with the homologous muscle activation. This non-specific increase in excitability with any upper limb muscle may be attributed to a generalized up regulation of M1 excitability through horizontal connections, convergence or divergence mechanisms within M1.

However, this also may be credited to a non-primary motor region upstream, which is likely what influenced the excitability changes seen during leg contraction. Right TA contraction (10% MVC) increased excitability of the right resting ECR to the same extent as all the right upper limb muscles tested. The remote effect has been extensively studied within the literature. Within M1 there are no known anatomical connections between the arm and leg muscle representations (Brown et al., 1991; Huntley & Jones, 1991). Common input to these motor regions likely arises from secondary motor areas, rather than horizontal connectivity within M1 (Murthy & Fetz, 1996). Arm and leg regions overlap in secondary motor areas (Fink et al., 1997). Previous research that demonstrates the remote effect in the upper limb with lower limb contraction has shown this with a high level of MVC (>70%) (Hortobagyi et al., 2003; Tazoe et al., 2007). With this high level of contraction the remote effect may be due to an increase in excitability of the motoneuron pool in the muscle of interest, rather than the remote effect. Since we were able to get an

increase with a much lower contraction level (% MVC) and the muscle tested was quiescent, a secondary motor area is likely mediating this interaction.

4.5.2 Effect of convergence of multiple effectors

Previous research has looked at convergence of multiple effectors with activation of upper and lower limb muscles contralateral to the hemisphere tested (Byblow et al., 2007), however to our knowledge no research has paired the remote effector (in our case the right TA) with the homologous muscle (left ECR) to see the effect this has on the resting upper limb muscle. By doing this we can explore convergence of primary and non-primary motor regions. Cross excitability changes with homologous muscle activation have been extensively studied (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). What we do not know is how activation of the distal lower limb muscle (representing the remote effect) influences the excitability of M1 during contraction of the homologous muscle. M1 excitability is higher when the isodirectional movement is performed, i.e. if the forearm is fully pronated, MEPs will be larger in ECR during dorsiflexion (Baldissera & Borroni, 2002; Borroni et al., 2004; Cerri et al., 2003). We tested both plantar flexors and dorsiflexors in order to confirm this. MEPs were larger in the right ECR during dorsiflexion. There was also an influence of force, MEPs were larger in the 30% MVC TA condition compared to the 10% MVC TA condition. Contraction of the homologous muscle in conjuction with the remote effector increased excitability more than the homologous muscle alone. There does appear to be an influence of convergence of primary and non-primary motor areas on M1 excitability.

4.5.3 Pathways mediating changes in cortical excitability during homologous muscle activation and remote effector

These studies were single pulse MEP experiments exploring corticospinal excitability changes during isolated low force isometric contractions. We did not explore the neural correlates influencing these excitability changes in this current study. The increases in excitability with homologous muscle contraction are likely due to transcallosal connections M1-M1; this has been studied extensively in the literature. Perez and Cohen (2008) found decreases in interhemispheric inhibition from the contralateral to the ipsilateral M1 during unimanual wrist flexion at higher levels of contraction force (30% and 70% MVC) compared to rest and low force (10%). Their low force condition was not associated with changes in MEP excitability, while we did see an increase at this low level of force, a decrease in interhemispheric inhibition may be mediating this interaction. Other studies have also found a decrease in intracortical inhibition during ipsilateral homologous muscle activation (Perez & Cohen, 2009; Uehara et al., 2013).

Past research of the remote effector has found that voluntary rhythmic flexion and extension of the foot causes cyclic modulation of the H-reflexes in the resting forearm (Baldissera et al., 1998). Many thought the afferent signals generated by the foot movement influenced spinal excitability at the level of the cervical spine, however cyclic modulation of the H-reflex in the resting forearm was not related to movement, it was temporally bound to activation of the foot movers (Cerri et al., 2003). Excitability changes during activation of the remote segment are likely driven at a central level. The facilitation seen during TA contraction may be

due to SMA-M1 connections. Byblow and colleagues (Byblow et al., 2007) found SMA conditioning during dorsiflexion and plantarflexion relative to resting conditions facilitated ECR MEPs.

4.5.4 Significance of findings and clinical implications

The present studies looked at the influence of spatial location of muscle representations with respect to somatotopy as well as the influence of the coordination of multiple effectors on M1 excitability. It has been established in the literature that unilateral voluntary muscle contractions in one arm yields excitability changes in the contralateral homologous muscle in the resting limb. The remote effector has also been well researched and produces excitability changes in the resting upper limb. What we did not know was the influence of coordinating a contraction of these two effectors at the same time to see their influence on M1 excitability. There does appear to be an increase in excitability with the convergence of multiple effectors, however we do not know what neurophysiological mechanisms are mediating this interaction.

We know that bimanual movement training can be an effective training method to enhance sensorimotor control of the upper limb musculature following brain injury (Cauraugh et al., 2010; Lin et al., 2010; McCombe Waller & Whitall, 2008; Stinear et al., 2008; Summers et al., 2007). What is not clear is the benefit of lower limb activity on upper limb function. We have little understanding of the neurophysiological changes that drive behavioural improvements in this population. Improving our understanding of the neurophysiology of everyday movements can help us to customize future rehabilitation interventions.

Chapter 5: Increased corticospinal output associated with the convergence of multiple effectors: a TMS study

5.1 Overview

The objective of this study was to investigate the neural mechanisms contributing to changes in the primary motor cortex (M1) during isometric contraction of multiple effectors. This study investigated the influence of both primary and non-primary motor areas on excitability changes in the resting M1. Transcranial magnetic stimulation (TMS) induced motor evoked potentials (MEPs) were recorded from the resting right extensor carpi radialis (ECR) during three conditions: (1) Isometric contraction of left wrist extensors at 10% MVC, (2) Isometric contraction of right dorsiflexors at 30% MVC, (3) Isometric contraction of left wrist extensors at 10% MVC + right dorsiflexors at 30% MVC. Single pulse TMS was delivered along with paired-pulse TMS techniques to investigate the effect of intracortical inhibition and facilitation [short interval intracortical inhibition (SICI), long interval intracortical inhibition (LICI) and intracortical facilitation (ICF)]. We also investigated interhemispheric inhibition (IHI) at both 10 ms and 40 ms interstimulus intervals. MEPs were potentiated in the right resting ECR during all three conditions (above rest), however there was no statistical difference in excitability between conditions. There was also no effect of the conditions on SICI, LICI, IHI10 or IHI40. There was however a decrease in ICF when the homologous muscle was active. Our findings reproduced excitability changes seen previously by our group. The increase in corticomotor drive is likely mediated upstream of M1 in non-primary motor areas (i.e. SMA), however this will have to be investigated in a future study. Improving our understanding of these cortical networks can be useful in the development of future neurorehabilitation techniques.

5.2 Introduction

Many activities of daily living require the precise coordination of activity in both the upper and lower limbs; this includes walking, playing musical instruments and playing sports. Many previous studies have investigated coordination of the upper and lower limbs and have shown when contralateral limbs are coordinated (i.e. left hand and right foot) comparable accuracy and stability is observed, regardless of whether the limbs are moved in the same (iso-) or opposite (noniso-) direction (Hiraga, Summers, & Temprado, 2004, 2005; Meesen, Wenderoth, Temprado, Summers, & Swinnen, 2006). In contrast, movements of the arm and leg on the same side of the body favour isodirectional movements (Baldissera, Cavallari, & Civaschi, 1982; Kelso & Jeka, 1992; Swinnen, Dounskaia, Verschueren, Serrien, & Daelman, 1995; Swinnen, 2002).

The remote effect is the term given to excitability changes that occur in the upper limb due to activity of the lower limb. Motor excitability is increased in the upper limb during contraction of the lower limb, which has been assessed with transcranial magnetic stimulation (TMS) (Hortobagyi et al., 2003). Voluntary rhythmic flexion and extension of the foot also causes cyclic modulation of H-reflexes in the resting forearm (Baldissera et al., 1998). Excitability of corticomotor projections to the resting limb increase with contraction of the remote effect, but also increase with contraction of the homologous muscle (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). Excitability changes associated with homologous muscle

activation have been studied extensively with both isometric and isotonic contractions (Carson et al., 2004; Ibey & Staines, 2013).

More recently TMS has been used to probe the neural correlates associated with the changes observed in both isodirectional and nonisodirectional coordination of the hand and foot. Byblow and colleagues (Byblow et al., 2007) used paired pulse TMS to study short interval intracortical inhibition (SICI) in resting forearm extensors during oscillatory dorsiflexion and plantar-flexion of the ipsilateral foot. They found that SICI was selectively reduced during dorsiflexion compared to plantar flexion. Byblow and colleagues suggested that lower M1 inhibition during dorsiflexion might facilitate isodirectional movement of the hand and foot. Reduced cortical inhibition in the hand muscles as a result of discrete (Sohn et al., 2005) and phasic (Tazoe et al., 2007) dorsiflexion movement of the ipsilateral foot has also been observed during measurements of silent period duration. Fujiyama and colleagues (Fujiyama et al., 2012) found that coordination of contralateral limbs (right hand, left foot) resulted in decreased corticospinal inhibition compared to coordination of ipsilateral limbs (right hand, right foot). Borroni and colleagues (Borroni et al., 2004) found that H-reflex excitability modulations in the upper limb remained phase linked to muscle contractions, not movement, of the lower limb. Force signals generated by Golgi tendon organs during movement of the foot muscles may in fact reach the hand motor area, modulating its excitability (McIntyre et al., 1984).

The convergence of information from the primary motor cortex (homologous muscle recruitment) and non-primary motor cortex has not been explored to our

knowledge. Within M1 there is no overlap or neurophysiological connections between arm and leg muscle representations (Brown et al., 1991; Huntley & Jones, 1991). Excitability changes arising in upper limb M1 representations during lower limb contraction is likely due to secondary motor areas where arm and leg regions overlap significantly, rather than horizontal connections in M1. Byblow and colleagues (2007) explored the functional connectivity between secondary and primary motor areas during foot movement. They found that upper limb corticomotor excitability and SICI were altered by movement conditions involving leg muscle activation and connections between SMA-M1 appeared to facilitate forearm corticospinal excitability in a non-specific manner (Byblow et al., 2007). Past research has demonstrated modulation of GABA-mediated inhibitory networks in upper limb M1 representations during foot movements (Baldissera & Borroni, 2002; Borroni et al., 2004). Previous studies have also found a reduction in upper limb corticomotor excitability when conditioning SMA at rest (Civardi et al., 2001), however lower limb dorsiflexion and plantarflexion appear to cause facilitation relative to rest. It has been speculated that this is due to the role of SMA in stabilizing posture during the coordination of hand and foot movements (Cerri et al., 2003).

To our knowledge no one has explored the neural mechanisms underlying changes in M1 during the convergence of both primary and non-primary motor regions. Ibey and Staines (*in preparation*) have previously found that excitability increases not only occur with homologous muscle activation and the remote effector, but there is also an increase when both contract together. The aim of this

study was to examine the neural mechanisms that contribute to the increase in excitability of M1 with recruitment of primary and non-primary motor areas. We explored intracortical inhibitory and excitatory networks (SICI, LICI, ICF) and interhemispheric inhibition (IHI) (M1-M1). We hypothesized that intracortical inhibition of the resting forearm muscle representation would be reduced by both ankle contraction and wrist contraction (individually and in combination). We also hypothesized there would be a reduction in interhemispheric inhibition.

5.3 **Materials and Methods**

5.3.1 Participants

Fifteen right-handed healthy volunteers (6 male, 9 female) with an average age of 26.9 ± 4.59 years participated in this study. Handedness was confirmed using the Edinburgh Inventory (Oldfield, 1971). All participants had no contraindications to TMS or any known neurological impairments. Participants gave their informed written consent to participate in the studies and completed a screening form for TMS. The Office of Research Ethics at the University of Waterloo approved all of the experimental procedures.

5.3.2 Experimental Setup

Subjects were examined at rest and in tasks that required isometric contractions. The left hemisphere extensor carpi radialis (ECR) motor representation was tested for all participants. The muscle of interest (right ECR muscle) was monitored to ensure it remained quiescent during all conditions. Muscle relaxation of the forearm was continuously monitored using custom

LabVIEW software (National Instruments, Austin, TX, USA). Subjects were given auditory feedback by the experimenters who monitored the electromyogram (EMG) signal. Muscle relaxation was confirmed using quantitative off-line analysis.

Participants were seated in a custom armchair with their forearms and feet fully supported. Arms were positioned 20-30° from their torso with both elbows in 90-100° of flexion. Knees were positioned in approximately 70-80° of flexion. Subjects were seated with their arms and feet in a custom-built manipulandum (Figure 5.1). At the beginning of each experiment subjects performed three trials of maximum voluntary isometric contractions for each movement. Maximum force output was measured in LabVIEW (National Instruments, Austin, TX, USA). The average was calculated and percent maximum voluntary contraction was determined for each condition.

Figure 5.1. Experimental Setup. Overhead view of the experimental set-up where subjects were instructed to perform isometric contractions of their left wrist extensors and/or right dorsiflexors while they were secured in manipulandums. The right ECR (muscle of interest) was at rest in all experimental conditions.

5.3.3 Electromyography

Electromyographic (EMG) activity of ECR, FCR and first dorsal interosseous (FDI) was recorded from both upper limbs and tibialis anterior (TA) of the right lower limb using bipolar surface electrodes placed longitudinally over the muscle bellies with the ground electrode over the right styloid process of the ulna. EMG signals were amplified (1000x), filtered (bandpass 2-2500 Hz) (Intronix Technologies Corporation Model 2024F, Canada) and digitized at a sample frequency of 5 kHz by an analog-to-digital interface (Micro1401, Cambridge Electronics Design, Cambridge, UK). All EMG data was stored on a PC for off-line analysis.

5.3.4 Transcranial Magnetic Stimulation (TMS)

Motor evoked potentials (MEPs) were collected from the resting right arm by stimulating the left motor cortex. Single and paired pulse TMS were delivered using two custom built 50 mm inner diameter figure-of-eight branding coils connected to two Magstim 200^2 stimulators (Magstim, Whitland, UK). Coil placement and orientation was continuously monitored using Brainsight (Brainsight 2; Rogue Research, Montreal, QC, Canada), a TMS neuronavigation system that displays real-time coil placement and target location on an anatomical magnetic resonance image (MRI). The motor hotspot for the ECR in M1 was acquired by placing the stimulation coil on the scalp at a 45° angle to the mid-sagittal plane. The motor hotspot was determined to be the location in M1 that elicits the largest MEP in the contralateral resting ECR (right). Resting motor threshold (RMT) was defined as the lowest stimulation intensity where potentials with peak-to-peak amplitude of a minimum

of 50 μV are evoked in at least five out of ten trials using the MagStim 200^2 stimulator (Magstim, Whitland, UK). TMS was triggered externally using SIGNAL Software and a Cambridge Electronic device (Power 1401, Cambridge Electronic Design, Cambridge, UK). Right ECR MEPs were recorded during single pulse, short-interval intracortical inhibition (SICI), intracortical facilitation (ICF) and long-interval intracortical inhibition (LICI) for all conditions from the left motor cortex stimulation. For MEPs, 10 individual TMS pulses were applied over the left M1 and the intensity was 120% of RMT. For SICI and ICF, both the conditioning and test stimuli were applied over M1 with the same coil connected to a Magstim 200^2 stimulator operating via a Bistim module. SICI and ICF were performed with a subthreshold conditioning stimulus (CS) followed by a suprathreshold test stimulus (TS) to the M1 hotspot for ECR. The interstimulus interval (ISI) for SICI and ICF was 3 and 12 ms respectively to produce intracortical inhibition and facilitation (Di Lazzaro et al., 2006; Kujirai et al., 1993). The CS was set at 80% of RMT for SICI and ICF. LICI uses a suprathreshold CS and TS of 120% RMT with an ISI of 100 ms (Chen, 2004). For testing interhemispheric inhibition (IHI) pairs of magnetic stimuli were delivered with two separate figure of eight coils. The TS was over the ECR hotspot in the left motor cortex (as per above) and the CS will be delivered with the other coil over the ECR hotspot in the right motor cortex. The ISI for testing IHI was 10 ms and 40 ms to produce short and long IHI (SIHI and LIHI respectively) (Chen, 2004; Chen et al., 2003; Ferbert et al., 1992; Ibey et al., 2015; Nelson et al., 2009; Perez & Cohen, 2008).

5.3.5 Experimental Procedures

Subjects were examined at rest and during three movement conditions: (1) Isometric contraction of left ECR at 10% MVC, (2) Isometric contraction of right TA at 30% MVC, and (3) Isometric contraction of left ECR at 10% MVC + Isometric contraction of right TA at 30% MVC. The left hemisphere was tested for all subjects. In all conditions the muscle of interest was the right resting ECR. All experimental conditions were pseudorandomized. Ten trials were collected of each TMS condition (Single pulse, SICI, LICI, ICF, IHI10, IHI40) for each movement condition.

5.3.6 Data Analysis

Experimenters examined the EMG data in the resting right arm prior to analysis of the MEPs for all conditions to ensure the muscles were silent. Relaxation of the right ECR and FCR was defined as EMG activity below the mean amplitude of 10 μV. The mean amplitude (rectified) was calculated 100 ms prior to delivery of TMS for all isometric conditions to ensure the muscle of interest (right ECR) was at rest. EMG profiles were also acquired for all the movement conditions for each muscle being collected at the time to calculate the average percent MVC for each condition.

A one-way ANOVA with factor Condition ((1) Isometric contraction of left ECR at 10% MVC, (2) Isometric contraction of right TA at 30% MVC, and (3) Isometric contraction of left ECR at 10% MVC + Isometric contraction of right TA at 30% MVC) was used to analyze the TMS protocols separately. SICI, LICI and ICF were expressed using the formula conditioned stimulus (CS)/test stimulus (TS). For the M1-M1 data the MEPs were also expressed as a percentage (CS/TS) and also

analyzed using a one-way ANOVA. Pre-planned contrasts were used to test hypotheses. Post-hoc analyses (Tukey correction method) were used to investigate any other differences between the conditions when a main effect was present. Significance was set at $p \leq 0.05$. All data sets were tested for normality and were well modeled by a normal distribution.

5.4 **Results**

EMG activity from the right resting ECR during all conditions along with the average percent MVC for each condition was calculated. The right resting ECR had an average activity of 6.06 μV ± 1.57 across all subjects and conditions. The average activity of the left ECR and right TA during active conditions (single and dual) across all subjects and conditions was 11.65% ± 3.35 MVC and 31.31% ± 5.37 MVC respectively. Peak to peak amplitude was calculated for all the MEPs (n=10) from the right resting ECR muscle for each condition and for each subject, these were then averaged in order to carry out the analyses.

Figure 5.2 illustrates the mean right resting ECR MEPs for each condition: isometric contractions of left ECR at 10% MVC, right TA at 30% MVC and left ECR at 10% in conjunction with right TA at 30%. A one-way ANOVA revealed a main effect of condition ($F_{3,42} = 5.16$; $p = 0.0040$). Post-hoc analysis showed all conditions were significantly different from rest, however there were no differences between isometric conditions ($p < 0.05$).

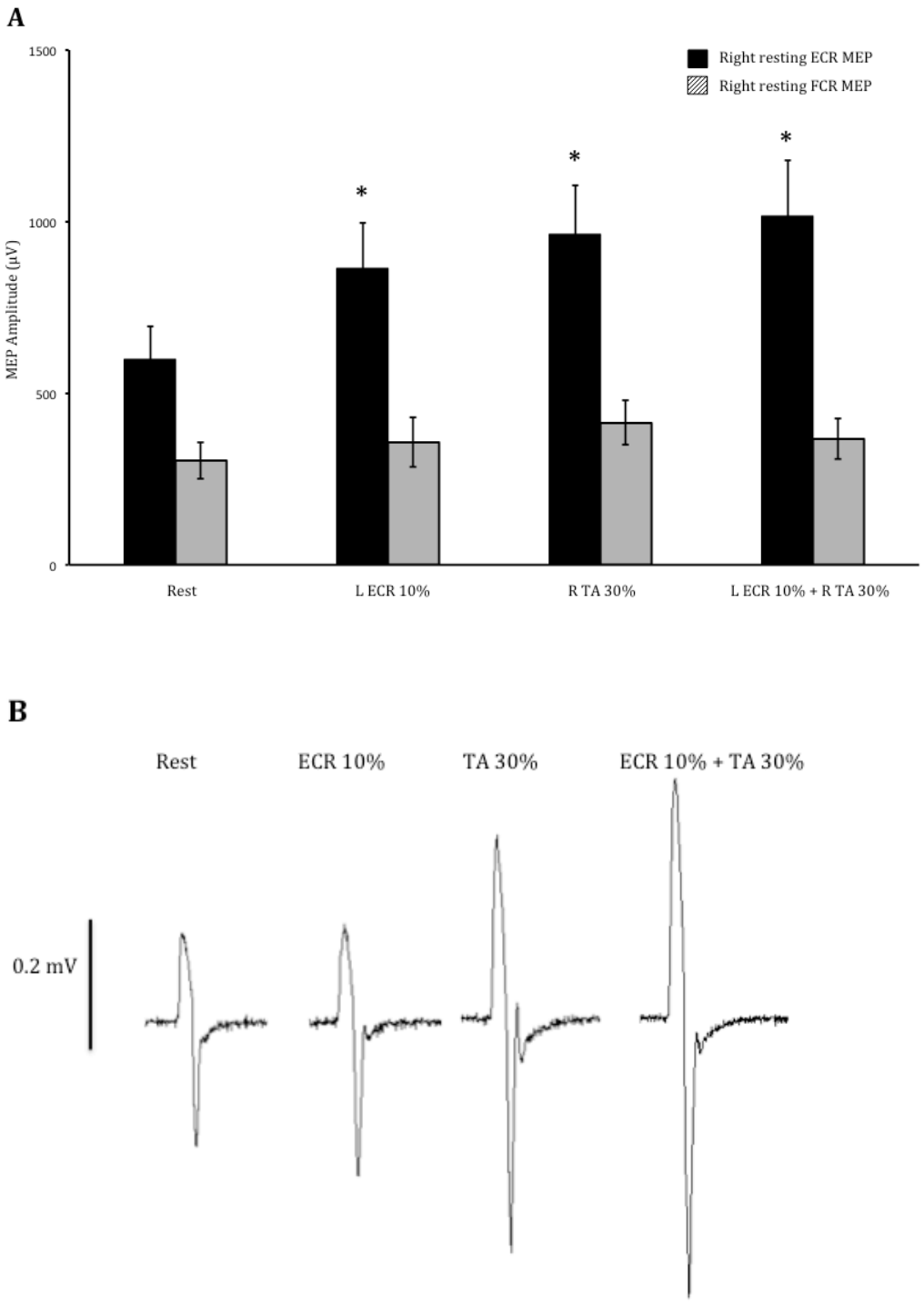

Figure 5.2. Mean effect of isometric contractions of the homologous muscle and a distal effector on motor evoked potentials induced by TMS. The mean (n=15) motor evoked potentials in right resting ECR and right resting FCR during isometric contraction of left ECR, right TA and left ECR + right TA together (A). Raw MEPs are represented in both (A) and (B). Data from one participant is shown in (B). Error bars represent standard error of the mean * $p<0.05$.

Figure 5.3 represents the data from SICI, LICI and ICF during each of the isometric conditions (isometric contractions of left ECR at 10% MVC, right TA at 30% MVC and left ECR at 10% in conjunction with right TA at 30%). A one-way ANOVA was run on each of these TMS conditions. There was no effect of condition on SICI ($F_{3,33}$ = 0.36; p = 0.7815) or LICI ($F_{3,42}$ = 2.32; p = 0.0889). There was an effect of condition on ICF ($F_{3,33}$ = 3.23; p = 0.0346). Post-hoc analysis showed isometric contraction of left ECR at 10% was significantly different from all other conditions. As shown in Figure 5.3, MEPs during ICF in the right resting ECR are decreased during the left ECR contraction.

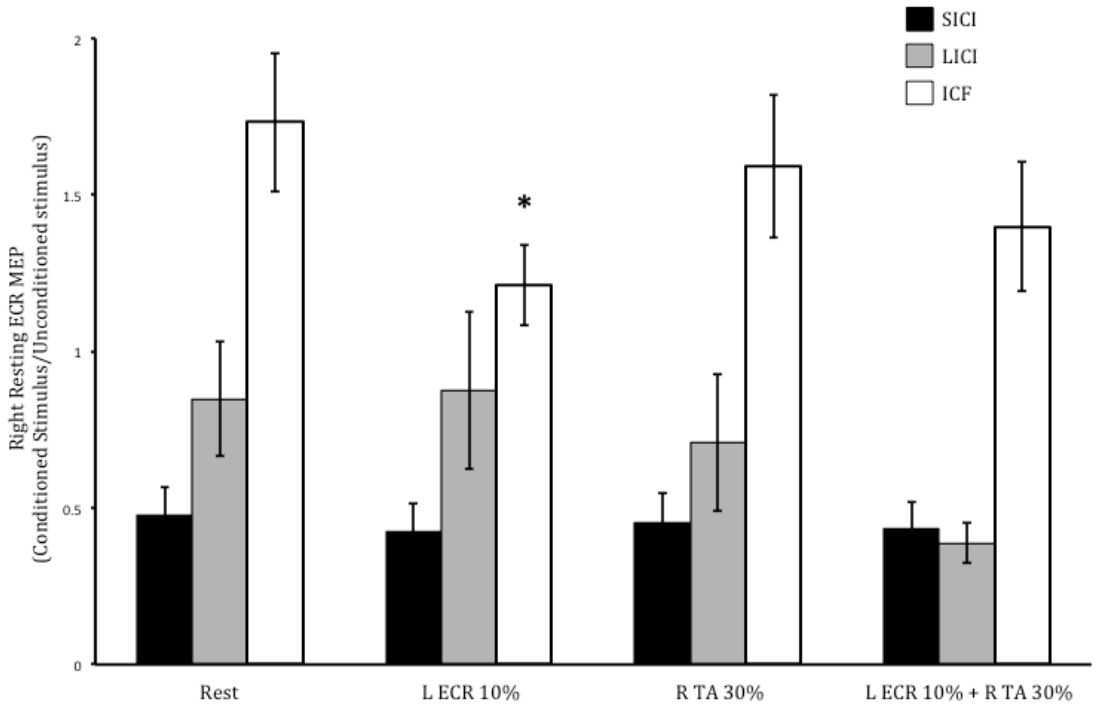

Figure 5.3. Mean effect of isometric contractions of the homologous muscle and a distal effector on inhibitory and excitatory mechanisms induced by TMS. The mean (n=12 SICI and ICF, n=15 LICI) motor evoked potentials in right resting ECR (expressed as conditioned stimulus over test stimulus) during isometric contraction of left ECR, right TA and left ECR + right TA together. Error bars represent standard error of the mean * p<0.05.

Interhemispheric inhibition at 10 ms and 40 ms across all conditions can be seen in Figure 5.4. A one-way ANOVA was run on each of these TMS conditions, IHI10 and IHI40, there was no main effect of condition in either condition [IHI 10 ($F_{3,27}$ = 1.52; p = 0.2328), IHI40 ($F_{3,27}$ = 0.35; p = 0.7917)].

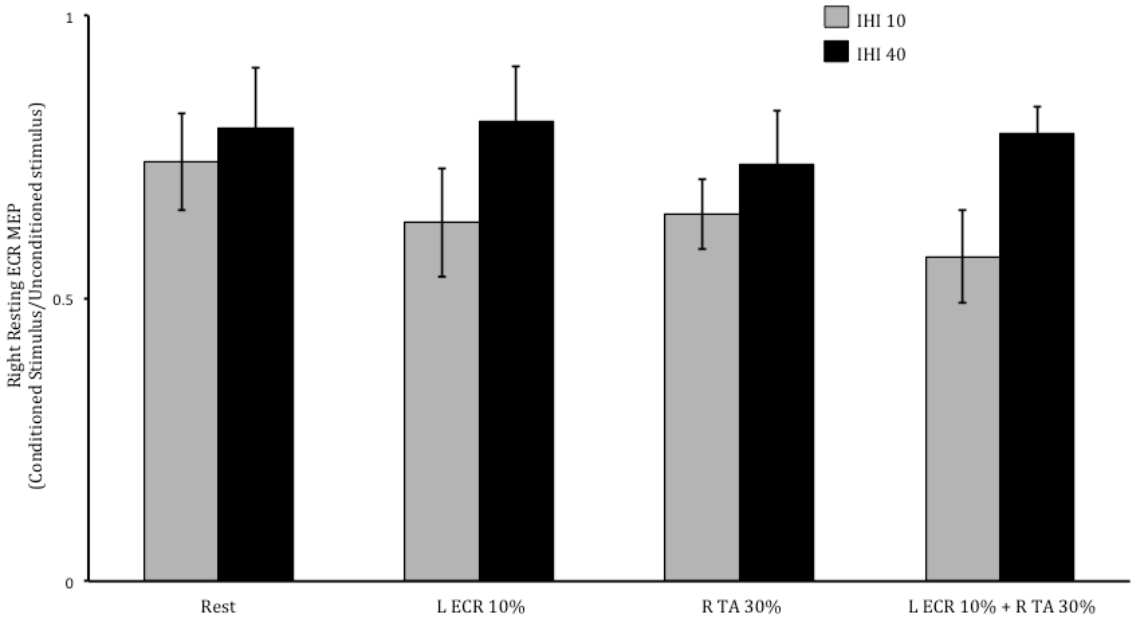

Figure 5.4. Mean effect of isometric contractions of the homologous muscle and a distal effector on interhemispheric inhibition induced by TMS. The mean (n=10) motor evoked potentials in right resting ECR (expressed as conditioned stimulus over test stimulus) during isometric contraction of left ECR, right TA and left ECR + right TA together. Error bars represent standard error of the mean.

5.5 Discussion

The aim of this study was to investigate the neural mechanisms contributing to changes in the primary motor cortex (M1) during the convergence of activity from primary and non-primary motor regions. Specifically, we investigated changes in corticomotor excitability to the right resting ECR muscle during isometric contractions of both the homologous muscle (left wrist extensors) and the remote

segment (right dorsiflexors). Our findings replicated the increase in corticomotor excitability that we have found previously in our lab, however we did not see any effect of the conditions on SICI, LICI, IHI10 or IHI40. There was a significant decrease in ICF during contraction of the homologous muscle group. To our knowledge, this is the first study to investigate the neural correlates involved in convergence of the homologous muscle and the remote segment on the same side as the muscle of interest.

5.5.1 Effect of convergence of homologous muscle and remote effector

The convergence of information from the primary motor cortex (homologous muscle recruitment) and non-primary motor cortex has not been explored to our knowledge. Using transcranial magnetic stimulation we are able to activate corticospinal neurons directly and transynaptically, and the size of the MEP is influenced by excitability of the neurons in both the motor cortex and the motoneuron pool (Rothwell et al., 1991). Cross excitability changes with homologous muscle activation have been studied significantly in the literature (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2009; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). Many research groups have also studied the remote effect and its influence on upper limb excitability. This effect is strongest when the isodirectional movement is performed (Baldissera & Borroni, 2002; Borroni et al., 2004; Cerri et al., 2003). Our current study did find a significant increase in excitability across all conditions in comparison to rest. There was no significant difference between activation of the homologous muscle, remote effector and the combination of the two. Past studies have attributed the increase in

excitability of the remote effector being due to a general increase in motor cortical excitability with strong voluntary contractions, however our effect was seen at a very low MVC in the remote segment (30%) as compared to past research (70%) (Hortobagyi et al., 2003). Byblow and colleagues (2007) did not indicate at what percent of MVC the participants activate their lower limb DF and plantarflexors. They stated that for the phasic contraction the resistance torque was programmed to obtain full range of motion and for the isometric contraction the subjects generated enough force to overcome the same torque. We kept our MVC low due to past study by our group that showed excitability changes at this low contraction and we wanted to avoid fatigue.

5.5.2 Effect of fatigue of the homologous muscle or remote effector on upper limb excitability

Fatigue can have a significant effect on both corticospinal excitability and intracortical inhibition. Unilateral fatiguing exercise affects the motor area innervating the exercising muscle and also the homologous non-exercised muscle (Bonato et al., 1996). There have been reports of a decrease in ICF in the ipsilateral muscles and decreases in ICI after unilateral fatiguing exercise (Bäumer, Münchau, Weiller, & Liepert, 2002; Takahashi et al., 2009). Takahashi and colleagues (2011) found muscle fatigue induced by exercising the lower limb has an effect on both SICI and corticospinal projections to muscles of the non-exercised upper limb. Since there are no apparent connections between the arm and leg within or between M1, it is more likely that these interactions occur outside of M1 rather than due to somatotopic spread. We kept the contraction levels low in this experiment in order

to avoid the element of fatigue from influencing our results (10% MVC in the left ECR and 30% in the right DF). Conditions were pseudo-randomized to try to avoid this from happening and MEPs were analyzed at the start and end of each trial.

5.5.3 Neural correlates mediating changes in cortical excitability during homologous muscle and remote effector contraction

More recently TMS has been used to probe the neural correlates associated with the changes observed in coordination of the hand and foot. Byblow and colleagues (2007) used paired pulse TMS to study short interval intracortical inhibition (SICI) in resting forearm extensors during oscillatory dorsiflexion and plantar-flexion of the ipsilateral foot. They found that SICI was selectively reduced during dorsiflexion compared to plantar-flexion. We did not see an effect on SICI during DF contraction, however we are unsure if our study was conducted at the same contraction level. Reduced cortical inhibition in the hand muscles as a result of discrete (Sohn et al., 2005) and phasic (Tazoe et al., 2007) dorsiflexion movement of the ipsilateral foot has also been observed during measurements of silent period duration. We did not see a difference during SICI or LICI in this study, however there was a trend towards greater LICI during the dual contraction condition. Byblow and colleagues (2007) did look at the effect of lower limb activation on ECR excitability, however they did not pair this with contraction of the upper limb. Their group explored the functional connectivity between secondary and primary motor areas during foot movement. They found that upper limb corticomotor excitability and SICI were altered by movement conditions involving leg muscle activation and connections between SMA-M1 appeared to facilitate forearm corticospinal

excitability in a non-specific manner (Byblow et al., 2007). Past research has demonstrated modulation of GABA-mediated inhibitory networks in upper limb M1 representations during foot movements (Baldissera & Borroni, 2002; Borroni et al., 2004). Previous studies have also found a reduction in upper limb corticomotor excitability when conditioning SMA at rest (Civardi et al., 2001), however lower limb dorsiflexion and plantarflexion appear to cause facilitation relative to rest.

5.5.4 Significance of findings and clinical implications

The present study investigated the neural correlates that may be influencing excitability changes seen during activation of the homologous muscle and remote effector. We did reproduce past results in our lab where there was an increase in excitability of the upper limb motor representation with contraction of the homologous muscle and remote effector. This increase in excitability remained with the coordination of both contractions together. We did not find an effect on intracortical inhibition or facilitation mechanisms during this coordination of effectors. We also did not observe a change in interhemispheric inhibition. More research will have to be done to investigate the neural correlates mediating these interactions. Improving our understanding of factors affecting both upper and lower limb coordination may help to customize future rehabilitation interventions post brain injury.

Chapter 6: General Discussion

6.1 Summary of Findings

This thesis examined the effect of different sensorimotor manipulations on corticospinal excitability to a resting proximal upper limb muscle. We investigated different properties modulating excitability of the resting muscle during rhythmical movement of the contralateral limb, including force, position, phase, and the addition of sensory input to the moving limb. We also explored the effect of somatotopy and spatial location within the motor cortex on excitability changes during both contralateral and ipsilateral muscle contractions of upper and lower limb muscles. Lastly we explored whether there is an effect of convergence of multiple effectors on M1 excitability changes – looking at the simultaneous influence of primary and non-primary motor regions. Increasing our understanding of interhemispheric interactions and the influence of non-primary motor regions on excitability changes in M1 can have future application on neuroplasticity.

Throughout this thesis the muscle of interest was always the right ECR, which was always at rest. Chapters 2-4 explored different factors that may modulate excitability of that resting muscle. In Chapter 2, our findings demonstrated that during rhythmic flexion and extension of the left wrist, there was an augmentation of the right resting ECR MEPs when the contralateral homologous muscle was active (extensors) with no effect of position within the phase, which was amplified as the force of contraction increased. While there was a significant phase effect (biggest amplification of excitability during the extension phase), there was also an increase in MEPs when the non-homologous muscle (wrist flexor) was active across all isotonic contractions. There was no significant difference between isometric and

isotonic contraction conditions. In Chapter 3 we added to the procedure in the previous experiment by investigating the effect of muscle vibration on the extensors on corticomotor projections to the contralateral resting limb during rest and rhythmical movement with added force manipulations. We replicated the findings of Chapter 2, however there was no difference between conditions with or without muscle vibration applied to the contracting ECR. There was however a cross excitability effect of vibration during the resting condition. In Chapter 4 we did not find an effect of somatotopy or spatial location of motor representations on M1 excitability during contralateral contractions. Interestingly, any contralateral muscle contraction increased excitability of the resting ECR to the same extent as its homologous muscle. We also looked at the effect of convergence of information from both the ipsilateral homologous muscle and a distal effector on the contralateral side. Recruitment of multiple effectors augmented excitability more than the homologous muscle alone. Further, the force of contraction of the distal effector also had an effect on the excitability changes. Chapter 5 sought out to explore the neural mechanisms contributing to the excitability changes seen with multiple effectors in Chapter 4, however no changes were seen in the measures we explored.

6.2 Contribution to existing literature

6.2.1 Cross excitability between homologous muscles (M1-M1)

For years researchers have known that voluntary contractions of one limb give rise to excitability changes in the cortical representation of homologous muscles of the opposite limb. This area has been extensively studied with isometric

contractions in both distal muscles of the hand and more recently in proximal arm muscles (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2008; Stedman et al., 1998; Stinear et al., 2001). Carson and colleagues also explored these cross excitability changes during continuous rhythmical movement and found it is in fact phase dependent (Carson et al., 2004). They (Carson et al., 2004) reported that the average EMG obtained from the moving arm was 21.4% MVC and 1.28% MVC for two different positions for FCR and 33.5% MVC 4.04% MVC for ECR. In that study contraction level was not controlled and they moved at a high frequency (2 Hz). What was yet to be studied was the effect of rhythmical movement at increasing force requirements and whether or not there is an influence of position in the movement cycle and if the phase relationship remained the same. Rhythmical contractions involve the convergence of multiple brain regions including M1, premotor cortex, SMA, S1, cerebellum and basal ganglia (Kim, Eliassen, & Sanes, 2005; Sadato et al., 1996; Witt, Laird, & Meyerand, 2008). The different positions within each phase tested the effect of somatosensory input (Ia muscle spindle activity) during the movement and there was no effect of position. Somatosensory input did not appear to be driving the excitability changes seen during the rhythmical contraction conditions. This was also supported by the decrease in corticospinal excitability during the passive rhythmical condition. Isotonic rhythmical contractions at different force levels also increase the amount of motor drive from that hemisphere. We looked at three different levels of MVC – no added external force, 10% and 30% MVC. We found there to be a significant effect of condition and phase (strongest when the homologous muscle was engaged), but

what was interesting was the significant increase in excitability when the non-homologous muscle (FCR) was active. The manipulandum we used in this study was able to set the force level in each direction, so when the non-homologous muscle was active the antagonistic muscle (ECR) was virtually silent. The spread of cross excitability from the non-homologous muscle has been seen in other studies, however this was at high levels of contraction forces (Hortobagyi et al., 2003). We cannot conclude from our study if these significant increases are due to the (1) rhythmical continuous nature of this task, (2) spread of excitability through horizontal connections to the antagonistic muscle, or (3) upstream non-primary motor areas.

We also looked at both isometric and isotonic contraction conditions to see if there would be a difference in cross excitability with these two different contraction types and we did not find there to be a significant difference between these conditions. Liepert and colleagues (2001) have found that tonic contractions facilitate excitability more than phasic contractions. Both of our conditions were held continuously, so we did not feel this would affect the excitability changes. Past studies have found that corticospinal and spinal excitability is significantly less in the lengthening contractions compared with shortening (Abbruzzese et al., 1994; Sekiguchi et al., 2001; Sekiguchi et al., 2003). However, in a more proximal muscle group Uematsu and colleagues (2010) and Howatson and colleagues (2011) both found that lengthening contractions produced greater increases in excitability when compared to shortening and isometric contractions. Our rhythmical isotonic conditions were concentric (shortening) in both directions and were never eccentric

contractions. There may be a larger influence of eccentric contractions in proximal muscle groups, however there was no difference between isometric and concentric with our experimental manipulations.

Cross facilitation is also augmented by the strength of the contraction (Hortobagyi et al., 2003; Muellbacher et al., 2000; Perez & Cohen, 2008; Stinear et al., 2001). At low levels of force (under 30% MVC) excitability is not consistently amplified (Liepert et al., 2001; Muellbacher et al., 2000; Stinear et al., 2001). We found significant increases in excitability in all contraction conditions (significantly above rest) – rhythmical contraction with no added force, 10% and 30% MVC, as well as isometric 10% and 30% MVC. There may be a difference depending on location of the muscle – i.e. distal vs. proximal muscles – which lends support to differences in transcallosal connections between the hemispheres.

We did explore SICI and LICI in the rhythmical conditions (no force and 20% MVC). There was no effect of our conditions on SICI and LICI. The excitability changes we demonstrated during rhythmical contractions in both the homologous and non-homologous muscle are likely due to interhemipsheric interactions between M1.

6.2.2 Effect of muscle vibration on cross excitability during motor task

The effect of muscle vibration on excitability of the contralateral M1 has been investigated with TMS (Rosenkranz & Rothwell, 2003). Muscle vibration has a significant effect on excitability of the vibrated muscle (Claus et al., 1988; Kossev et al., 1999; Rosenkranz et al., 2000; Siggelkow et al., 1999). Not many studies have investigated the cross excitability effect of muscle vibration between the

hemispheres. Past research has found increases in excitability to the contralateral resting homologous muscle (Claus et al., 1988; Kossev et al., 1999), however these studies were all performed at rest. Our study (Chapter 3) did investigate cross excitability during muscle vibration at rest, but also explored if there was an effect of increased afferent information during the same isotonic rhythmical contractions we performed in Chapter 2 (we had a no force and a 20% MVC condition this time). We did find there to be a trending increase in cross excitability of muscle vibration at rest, however there was no effect during the movement manipulations. In the previous study there was no effect of position within phase, which would be activating muscle spindles. This study looked to see if there was an effect of up-regulating somatosensory input during voluntary muscle contraction, since previous research has shown cross excitability changes with muscle vibration. Without motor drive there may be a convergence of this sensory information onto the resting M1, however during voluntary contraction there is no added contribution of vibration to excitability changes. Rather, there may be an inhibitory effect of somatosensory information converging onto M1 when the task is not relevant. This task did not require the use of that information, subjects simply had to rhythmically move to a metronome against increasing force manipulations. We cannot rule out whether vibration may have an added benefit to neuroplasticity mechanisms during motor tasks/manipulations, however it did not have an acute benefit on M1 excitability.

6.2.3 Impact of somatotopy and spatial location of muscle representation

We found a significant increase in excitability of M1 during non-homologous muscle activation during both isometric and isotonic contractions (Chapter 2). In Chapter 4 (Experiment 1) we had subjects perform low force isometric contractions of both ipsilateral and contralateral upper and lower limb muscles to see how they influenced excitability of the ECR M1 representation. There was only a significant increase in the homologous muscle on the ipsilateral side, yet all contralateral muscles increased excitability to the same extent. There was no significant difference between excitability increases during homologous muscle activation or any contralateral muscle. There was no spread of excitability to the ECR muscle during any contractions – we monitored EMG levels and eliminated any conditions with co-contraction. This non-specific increase in excitability during all contralateral muscle contractions may be due to horizontal connections, convergence or divergence within M1. This may also be attributed to areas upstream – non-primary motor areas. It is possible that the increases seen with contralateral upper limb contractions may be due to a different mechanism than lower limb contractions. There are no known anatomical connections between the arm and leg muscle representations in M1 (Brown et al., 1991; Huntley & Jones, 1991). The increases seen during lower limb contraction are likely originating upstream, i.e. SMA (Byblow et al., 2007). Lower limb contraction increased ECR corticospinal excitability to the same extent as upper limb contractions; this may indicate there is a common driver in a non-primary motor region contributing to these changes. We need to investigate the influence of these non-homologous muscles in more detail to

better understand factors that can influence excitability of a resting motor representation and lead to mechanisms influencing plasticity.

6.2.4 Excitability changes in the upper limb during contraction of lower limb (i.e. remote effect)

Previous studies have demonstrated the remote effect increases corticospinal excitability from the upper limb motor areas (Hortobagyi et al., 2003; Tazoe et al., 2007). Our studies (Chapter 4 and 5) demonstrated an increase in excitability during both plantar-flexion and dorsiflexion, with the biggest increase being during TA activation (dorsiflexion). This was no surprise since past research has shown the biggest increases in excitability are during the iso-directional movement to the resting limb. Many studies on the remote effect have only found an effect at high contraction forces, however we found a difference in excitability at both 10% and 30% MVC. Our contractions were kept lower to avoid the influence of fatigue, though there was no sign of fatigue during analysis of the MEPs. There are many studies that have looked at the influence of fatigue on upper limb MEPs during lower limb contraction (Bonato et al., 1996). The magnitude of the remote effect is greater during fatiguing remote muscle contraction in comparison to non-fatiguing contraction (Tazoe et al., 2009). It is unlikely that this would explain the increases in excitability that we found at low contraction forces.

Byblow and colleagues (Byblow et al., 2007) explored the functional connectivity between secondary and primary motor areas during foot movement. They found that upper limb corticomotor excitability and SICI were altered by movement conditions involving leg muscle activation and connections between

SMA-M1 appeared to facilitate forearm corticospinal excitability in a non-specific manner (Byblow et al., 2007). Since this may be mediated cortically, the increase in motor drive may be converging onto the resting upper limb representation and influencing the difference in excitability between contraction levels. Further to this, past studies have found force signals generated by Golgi tendon organs during movement of the foot muscles may in fact reach the hand motor area, modulating its excitability (McIntyre et al., 1984). We explored the effect of lower limb contraction on SICI, LICI and ICF in a resting upper limb motor region and found no effect of our condition (TA contraction at 30%) on intracortical inhibitory and excitatory interneurons. Many regions may be contributing to the remote effect, however it is clear from past research that it is at least partially mediated at a cortical level where there is convergence of this information onto the CST of the upper limb representation.

6.2.5 Influence of convergence of primary and non-primary motor cortices during motor tasks

In both Chapters 4 and 5 we looked at the influence of converging information from primary and non-primary motor regions by having participants simultaneously contract the ipsilateral homologous muscle (left ECR) at a low level contraction (10% MVC) with the contralateral remote effector (right TA). There was a significant increase in M1 excitability during the dual contraction conditions, however we were unable to find the neural mechanisms responsible for this increase. The increase in excitability during homologous muscle activity is likely M1-M1, however the increase in excitability during lower limb contraction is likely

mediated upstream in non-primary motor areas (Brown et al., 1991; Fink et al., 1997; Huntley & Jones, 1991; Murthy & Fetz, 1996). How the convergence of these regions affects excitability of M1 is not yet known. During dual contracting there was a non-significant increase in LICI. There may be an increase in intracortical inhibition doing the dual effector condition. This could mean that there is not a benefit of lower limb contraction and this may not have a beneficial influence on neuroplasticity mechanisms. More studies will need to explore the mechanisms contributing to this interaction.

6.3 Conclusions

In this thesis we have demonstrated different sensorimotor manipulations that increase excitability of a resting motor representation within M1. This research adds to the body of cross excitability research and further informs us that these excitability changes are not limited to the homologous muscle or to non-homologous muscles at high levels of contraction force. We demonstrated augmentation of motor drive to the resting M1 representation during rhythmical isotonic and isometric contractions. These studies displayed the phase relationship (high excitability increases during homologous muscle activation), however the non-homologous muscle also increased excitability of M1. Vibration to the moving muscle did not increase corticospinal excitability during rhythmical isotonic contractions. Further, there did not appear to be an effect of somatotopy or spatial location of muscle representations on differences in M1 excitability – rather, there was a general increase in excitability of M1 during any contralateral contraction that

matched excitability changes during homologous muscle activation. Contraction of multiple effectors (homologous muscle and distal effector), which in theory is recruiting primary and non-primary motor areas, also increased M1 excitability. The mechanisms by which these interactions occur are not clear. These findings suggest that cross excitability studies, which may inform new mechanisms to target neuroplasticity, should not be limited to the homologous muscle. Cross education research as well as neurorehabilitation research may benefit from using other nodes to target excitability changes in M1.

6.4 Limitations

There are some general limitations across these studies that we will address. We did not control for testing time, caffeine consumption, exercise, gender, or hormonal levels. All of these factors have an affect on corticospinal excitability. We can use TMS to examine the role of the different sensory and motor manipulations on changes in M1 excitability, however the variability in the neurophysiological response is high. This variability can influence our interpretations of the results and our ability to know which behaviours are influencing plasticity changes. Gender, activity level/fitness, time of day, age, attention, genetics, and history of synaptic activity all influence resting excitability of M1. There are also pharmacological influences that will affect excitability of M1 and individuals also have variability in their inhibitory circuitry (for review see Ridding & Ziemann, 2010). Anatomical differences also contribute to differences in stimulation intensities, i.e. skull thickness and neuronal fiber orientation. These factors can also interact with each

other, adding to the complexity of interpreting the response. Therefore the variability that is reported in these studies likely arises from the stimulation parameters, inter-individual variability and the interaction of the two.

We used transcranial magnetic stimulation to test excitability changes across all studies in this thesis. TMS reflects corticomotor excitability changes along the whole neuroaxis – the brain and the spinal cord. We did not distinguish between cortical, subcortical and spinal excitability in these studies and therefore cannot conclude that the CST excitability changes are due to cortical neural correlates. Further, TMS does not have good spatial resolution. There may have been spread of excitability to both muscle representations within M1 and to neighbouring cortical regions (i.e. S1, premotor cortices, etc.). TMS also does not distinguish which neuronal pools are contributing to the excitability changes we are measuring. That being said, all conditions would have the same limitations and are being compared against each other, so these factors shouldn't have been an issue in interpreting the results.

In Chapters 3 and 5 we tested intracortical inhibitory and excitatory mechanisms (SICI, LICI, ICF) and interhemispheric inhibition (IHI10 and IHI40) using paired pulse and dual coil TMS. These mechanisms have been extensively studied during resting conditions, however not as much literature has been produced on movement conditions and manipulations. In resting conditions the aim is to keep the testing stimulus amplitude at ~1 mV for distal muscles and ~0.5 mV for proximal muscles. Due to the high variability of MEPs during contraction conditions it was very difficult to adjust TMS amplitudes during both the isotonic

and isometric contraction conditions. We decided to keep the CS and TS amplitudes consistent, but it is possible that these were not optimal to see both increases and decreases in excitability during these manipulations. In addition, while these manipulations are supposed to be probing both GABA and glutamatergic receptor activity we cannot be sure that these mechanisms hold true during voluntary contraction in proximal muscles at the ISIs used.

Lastly, because we only used TMS as our assessment tool we cannot conclude the effect our conditions had on other areas of the cortex that may have contributed to our findings. We would need to use high resolution imaging techniques to determine the effect of the manipulations on other areas of the cortex.

6.5 Future directions & clinical applications

6.5.1 Exploring the neural correlates mediating these interactions

Future research will need to explore the neural correlates mediating the excitability changes we have demonstrated within this thesis. Intracortical and interhemispheric interactions have been studied during rhythmical movement and isometric contractions in the past, although the cross excitability changes during rhythmical isotonic movement have not been studied at increasing levels of force in the proximal muscles. Looking at contributions upstream of M1 from premotor cortices or SMA will increase our understanding of the non-primary motor areas that may be coordinating excitability changes in M1. These areas upstream of M1 may be contributing to the decreased inhibition during non-homologous muscle activation, and may be mediating excitability changes during homologous muscle

activation with a distal effector. Past research has found that the bilateral interactions between SMA may be a predictor of the transfer effect leading to cross education. The next step in this series of experiments would be to do a continuous theta burst stimulation (cTBS) study. We could test these manipulations (isotonic contractions and multiple effectors) pre- and post-cTBS to the SMA. If the SMA was a large contributor to this effect, excitability should decrease post-cTBS.

6.5.2 Investigate mechanisms of plasticity

It is well known that neuroplasticity can change the structure and/or function of the central nervous system in many patient populations. Using paired associative stimulation to investigate potential plasticity changes with the interventions in this thesis that affected excitability of M1 may demonstrate the potential utility these protocols have on neuroplasticity.

6.5.3 Determining if there is an effect on cross education

Cross education research typically focuses on the upper limb. Training paradigms that involve more than one limb may lead to greater excitability changes in the resting M1 and therefore may effect the transfer for cross education changes. More cross education research should explore the convergence of information from different cortical regions – i.e. S1, SMA, premotor cortices. We know that this transfer effect is training specific between muscle groups, however the simultaneous recruitment of other regions may up regulate or down regulate this interaction. We need to better understand how the selective convergence of information from multiple brain areas can increase the induction of plasticity.

6.5.4 Clinical applicability

One goal of neurorehabilitation after a brain injury (i.e. a stroke) is to restore or improve motor function. A large number of individuals who have experienced a stroke can regain the ability to perform basic activities of daily living after rehabilitation therapy. Various interventions and strategies, like constraint induced therapy and bimanual motor training, are used to promote functional recovery. The goal is to promote neuroplasticity and long-term cortical reorganization post brain injury. Unfortunately, there is currently little understanding of the neurophysiological changes that drive these behavioural improvements. Continuing to improve our understanding of the neurophysiology of everyday upper limb movements will help us to have a more clear understanding of the changes post brain injury. With this we can design better rehabilitation protocols, which may improve motor function.

Letter of Copyright Permission

ELSEVIER LICENSE
TERMS AND CONDITIONS

Mar 06, 2017

This Agreement between Robyn J Ibey ("You") and Elsevier ("Elsevier") consists of your license details and the terms and conditions provided by Elsevier and Copyright Clearance Center.

License Number	4063071498280
License date	Mar 06, 2017
Licensed Content Publisher	Elsevier
Licensed Content Publication	Behavioural Brain Research
Licensed Content Title	Corticomotor excitability changes seen in the resting forearm during contralateral rhythmical movement and force manipulations: A TMS study
Licensed Content Author	R.J. Ibey,W.R. Staines
Licensed Content Date	15 November 2013
Licensed Content Volume	257
Licensed Content Issue	n/a
Licensed Content Pages	10
Start Page	265
End Page	274
Type of Use	reuse in a thesis/dissertation
Portion	full article
Format	both print and electronic
Are you the author of this Elsevier article?	Yes
Will you be translating?	No
Order reference number	
Title of your thesis/dissertation	Factors influencing bilateral interactions in the human motor cortex: investigating transcallosal sensorimotor networks
Expected completion date	Apr 2017
Estimated size (number of pages)	200
Elsevier VAT number	GB 494 6272 12
Requestor Location	Robyn J Ibey University of Waterloo Department of Kinesiology 200 University Ave W

References

Abbruzzese, G., Morena, M., Spadavecchia, L., & Schieppati, M. (1994). Response of arm flexor muscles to magnetic and electrical brain stimulation during shortening and lengthening tasks in man. *The Journal of Physiology*, *481*(Pt 2), 499–507.

Aizawa, H., Mushiake, H., Inase, M., & Tanji, J. (1990). An output zone of the monkey primary motor cortex specialized for bilateral hand movement. *Experimental Brain Research*, *82*(1), 219–221.

Amassian, V. E., Cracco, R. Q., & Maccabee, P. J. (1989). Focal stimulation of human cerebral cortex with the magnetic coil: a comparison with electrical stimulation. *Electroencephalography and Clinical Neurophysiology*, *74*(6), 401–16.

Angelucci, A., Levitt, J. B., Walton, E. J. S., Hupe, J.-M., Bullier, J., & Lund, J. S. (2002). Circuits for local and global signal integration in primary visual cortex. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *22*(19), 8633–8646.

Ashe, J. (1997). Force and the motor cortex. *Behavioural Brain Research*, *87*(2), 255–269.

Baldissera, F., & Borroni, P. (2002). Excitability changes in human corticospinal projections to forearm muscles during voluntary movement of ipsilateral foot. *The Journal of Physiology, 539*(Pt 3), 903–911.

Baldissera, F., Cavallari, P., & Civaschi, P. (1982). Preferential coupling between voluntary movements of ipsilateral limbs. *Neuroscience Letters*, *34*(1), 95–100.

Baldissera, F., Cavallari, P., & Leocani, L. (1998). Cyclic modulation of the H-reflex in

a wrist flexor during rhythmic flexion-extension movements of the ipsilateral foot. *Experimental Brain Research*, *118*(3), 427–30.

Barker, A. T., Jalinous, R., & Freeston, I. L. (1985). Non-invasive magnetic stimulation of human motor cortex. *Lancet*, *1*(8437), 1106–7.

Bäumer, T., Münchau, A., Weiller, C., & Liepert, J. (2002). Fatigue suppresses ipsilateral intracortical facilitation. *Experimental Brain Research*, *146*(4), 467–473.

Beck, S., & Hallett, M. (2011). Surround inhibition in the motor system. *Experimental Brain Research*, *210*(2), 165–172.

Beck, S., Richardson, S. P., Shamim, E. A., Dang, N., Schubert, M., & Hallett, M. (2008). Short intracortical and surround inhibition are selectively reduced during movement initiation in focal hand dystonia. *The Journal of Neuroscience*, *28*(41), 10363–9.

Beck, S., Schubert, M., Richardson, S. P., & Hallett, M. (2009). Surround inhibition depends on the force exerted and is abnormal in focal hand dystonia. *Journal of Applied Physiology*, *107*(5), 1513–8.

Binder, C., Kaya, A. E., & Liepert, J. (2009). Vibration prolongs the cortical silent period in an antagonistic muscle. *Muscle & Nerve*, *39*(6), 776–780.

Bonato, C., Zanette, G., Manganotti, P., Tinazzi, M., Bongiovanni, G., Polo, A., & Fiaschi, A. (1996). “Direct” and “crossed” modulation of human motor cortex excitability following exercise. *Neuroscience Letters*, *216*(2), 97–100.

Borroni, P., Cerri, G., & Baldissera, F. (2004). Excitability changes in resting forearm muscles during voluntary foot movements depend on hand position: a neural

substrate for hand-foot isodirectional coupling. *Brain Research*, *1022*(1–2), 117–25.

Brasil-Neto, J. P., Cohen, L. G., Pascual-Leone, A., Jabir, F. K., Wall, R. T., & Hallett, M. (1992). Rapid reversible modulation of human motor outputs after transient deafferentation of the forearm: a study with transcranial magnetic stimulation. *Neurology*, *42*(7), 1302–1306.

Brinkman, J., & Kuypers, H. G. (1973). Cerebral control of contralateral and ipsilateral arm, hand and finger movements in the split-brain rhesus monkey. *Brain : A Journal of Neurology*, *96*(4), 653–674.

Brown, P., Day, B. L., Rothwell, J. C., Thompson, P. D., & Marsden, C. D. (1991). Intrahemispheric and interhemispheric spread of cerebral cortical myoclonic activity and its relevance to epilepsy. *Brain : A Journal of Neurology*, *114* (Pt 5), 2333–51.

Burke, D., Hagbarth, K. E., Löfstedt, L., & Wallin, B. G. (1976). The responses of human muscle spindle endings to vibration of non-contracting muscles. *The Journal of Physiology*, *261*(3), 673–93.

Byblow, W. D., Coxon, J. P., Stinear, C. M., Fleming, M. K., Williams, G., Müller, J. F. M., & Ziemann, U. (2007). Functional connectivity between secondary and primary motor areas underlying hand-foot coordination. *Journal of Neurophysiology*, *98*(1), 414–22.

Carroll, T. J., Barton, J., Hsu, M., & Lee, M. (2009). The effect of strength training on the force of twitches evoked by corticospinal stimulation in humans. *Acta Physiologica*, *197*(2), 161–173.

Carroll, T. J., Herbert, R. D., Munn, J., Lee, M., & Gandevia, S. C. (2006). Contralateral effects of unilateral strength training: evidence and possible mechanisms. *Journal of Applied Physiology*, *101*(5), 1514–22.

Carroll, T. J., Lee, M., Hsu, M., & Sayde, J. (2008). Unilateral practice of a ballistic movement causes bilateral increases in performance and corticospinal excitability. *Journal of Applied Physiology*, *104*(6), 1656–64.

Carson, R. G. (2005). Neural pathways mediating bilateral interactions between the upper limbs. *Brain Research Reviews*, *49*(3), 641–62.

Carson, R. G., & Riek, S. (2000). Musculo-skeletal constraints on corticospinal input to upper limb motoneurones during coordinated movements. *Human Movement Science*, *19*(4), 451–474.

Carson, R. G., Riek, S., & Bawa, P. (1999). Electromyographic activity, H-reflex modulation and corticospinal input to forearm motoneurones during active and passive rhythmic movements. *Human Movement Science*, *18*(2–3), 307–343.

Carson, R. G., Riek, S., Mackey, D. C., Meichenbaum, D. P., Willms, K., Forner, M., & Byblow, W. D. (2004). Excitability changes in human forearm corticospinal projections and spinal reflex pathways during rhythmic voluntary movement of the opposite limb. *The Journal of Physiology*, *560*(Pt 3), 929–940.

Carson, R. G., Welsh, T. N., & Pamblanco-Valero, M. A. (2005). Visual feedback alters the variations in corticospinal excitability that arise from rhythmic movements of the opposite limb. *Experimental Brain research*, *161*(3), 325–334.

Castro-Alamancos, M. A., & Borrel, J. (1995). Functional recovery of forelimb response capacity after forelimb primary motor cortex damage in the rat is due

to the reorganization of adjacent areas of cortex. *Neuroscience*, *68*(3), 793–805.

Cauraugh, J. H., Lodha, N., Naik, S. K., & Summers, J. J. (2010). Bilateral movement training and stroke motor recovery progress: a structured review and meta-analysis. *Human Movement Science*, *29*(5), 853–70.

Cernacek, J. (1961). Contralateral motor irradiation--cerebral dominance. Its changes in hemiparesis. *Archives of Neurology*, *4*, 165–172.

Cerri, G., Borroni, P., & Baldissera, F. (2003). Cyclic h-reflex modulation in resting forearm related to contractions of foot movers, not to foot movement. *Journal of Neurophysiology*, *90*(1), 81–8.

Chen, R. (2004). Interactions between inhibitory and excitatory circuits in the human motor cortex. *Experimental Brain research*, *154*(1), 1–10.

Chen, R., Corwell, B., Yaseen, Z., Hallett, M., & Cohen, L. G. (1998). Mechanisms of cortical reorganization in lower-limb amputees. *The Journal of Neuroscience*, *18*(9), 3443–50.

Chen, R., Tam, A., Butefisch, C., Corwell, B., Ziemann, U., Rothwell, J. C., & Cohen, L. G. (1998). Intracortical inhibition and facilitation in different representations of the human motor cortex. *Journal of Neurophysiology*, *80*(6), 2870–2881.

Chen, R., Yung, D., & Li, J. Y. (2003). Organization of ipsilateral excitatory and inhibitory pathways in the human motor cortex. *Journal of Neurophysiology*, *89*(3), 1256–1264.

Chiappa, K. H., Cros, D., Day, B., Fang, J. J., Macdonell, R., & Mavroudakis, N. (1991). Magnetic stimulation of the human motor cortex: ipsilateral and contralateral facilitation effects. *Electroencephalography and Clinical Neurophysiology*

Supplement, *43*, 186–201.

Cisek, P., Crammond, D. J., & Kalaska, J. F. (2003). Neural activity in primary motor and dorsal premotor cortex in reaching tasks with the contralateral versus ipsilateral arm. *Journal of Neurophysiology*, *89*(2), 922–942.

Civardi, C., Cantello, R., Asselman, P., & Rothwell, J. C. (2001). Transcranial magnetic stimulation can be used to test connections to primary motor areas from frontal and medial cortex in humans. *NeuroImage*, *14*(6), 1444–53.

Claus, D., Mills, K. R., & Murray, N. M. (1988). The influence of vibration on the excitability of alpha motoneurones. *Electroencephalography and Clinical Neurophysiology*, *69*(5), 431–6.

Cohen, L. (1971). Synchronous bimanual movements performed by homologous and non-homologous muscles. *Perceptual and Motor Skills*, *32*, 639–644.

Cohen, L. G., Bandinelli, S., Topka, H. R., Fuhr, P., Roth, B. J., & Hallett, M. (1991). Topographic maps of human motor cortex in normal and pathological conditions: mirror movements, amputations and spinal cord injuries. *Electroencephalography and Clinical Neurophysiology Supplement*, *43*, 36–50.

Collins, D. F., & Prochazka, A. (1996). Movement illusions evoked by ensemble cutaneous input from the dorsum of the human hand. *The Journal of Physiology*, *496*(Pt 3), 857–71.

Coxon, J. P., Stinear, J. W., & Byblow, W. D. (2005). Amplitude of muscle stretch modulates corticomotor gain during passive movement. *Brain Research*, *1031*(1), 109–117.

Dai, T. H., Liu, J. Z., Sahgal, V., Brown, R. W., & Yue, G. H. (2001). Relationship

between muscle output and functional MRI-measured brain activation. *Experimental Brain Research*, *140*(3), 290–300.

Daskalakis, Z. J., Christensen, B. K., Fitzgerald, P. B., Roshan, L., & Chen, R. (2002). The mechanisms of interhemispheric inhibition in the human motor cortex. *The Journal of Physiology*, *543*(Pt 1), 317–326.

Day, B. L., Rothwell, J. C., Thompson, P. D., Dick, J. P., Cowan, J. M., Berardelli, A., & Marsden, C. D. (1987). Motor cortex stimulation in intact man. 2. Multiple descending volleys. *Brain : A Journal of Neurology*, 1191–209.

DeFelipe, J., & Fariñas, I. (1992). The pyramidal neuron of the cerebral cortex: Morphological and chemical characteristics of the synaptic inputs. *Progress in Neurobiology*, *39*(6), 563–607.

Dettmers, C., Fink, G. R., Lemon, R. N., Stephan, K. M., Passingham, R. E., Silbersweig, D., … Frackowiak, R. S. (1995). Relation between cerebral activity and force in the motor areas of the human brain. *Journal of Neurophysiology*, *74*(2), 802–815.

Di Lazzaro, V., Oliviero, A., Meglio, M., Cioni, B., Tamburrini, G., Tonali, P., & Rothwell, J. C. (2000). Direct demonstration of the effect of lorazepam on the excitability of the human motor cortex. *Clinical Neurophysiology*, *111*(5), 794–799.

Di Lazzaro, V., Oliviero, A., Profice, P., Insola, A., Mazzone, P., Tonali, P., & Rothwell, J. C. (1999). Direct demonstration of interhemispheric inhibition of the human motor cortex produced by transcranial magnetic stimulation. *Experimental Brain research*, *124*(4), 520–524.

Di Lazzaro, V., Pilato, F., Dileone, M., Ranieri, F., Ricci, V., Profice, P., … Ziemann, U.

(2006). GABAA receptor subtype specific enhancement of inhibition in human motor cortex. *The Journal of Physiology*, *575*(Pt 3), 721–6.

Donchin, O., Gribova, A., Steinberg, O., Bergman, H., & Vaadia, E. (1998). Primary motor cortex is involved in bimanual coordination. *Nature*, *395*(6699), 274–278.

Donchin, O., Gribova, A., Steinberg, O., Mitz, A. R., Bergman, H., & Vaadia, E. (2002). Single-unit activity related to bimanual arm movements in the primary and supplementary motor cortices. *Journal of Neurophysiology*, *88*(6), 3498–3517.

Donoghue, J. P., Leibovic, S., & Sanes, J. N. (1992). Organization of the forelimb area in squirrel monkey motor cortex: representation of digit, wrist, and elbow muscles. *Experimental Brain Research*, *89*(1), 1–19.

Dum, R. P., & Strick, P. L. (1991). The origin of corticospinal projections from the premotor areas in the frontal lobe. *The Journal of Neuroscience*, *11*(3), 667–689.

Dum, R. P., & Strick, P. L. (2002). Motor areas in the frontal lobe of the primate. *Physiology & Behavior*, *77*(4–5), 677–682.

Evarts, E. V. (1968). Relation of pyramidal tract activity to force exerted during voluntary movement. *Journal of Neurophysiology*, *31*(1), 14–27.

Evarts, E. V, Fromm, C., Kroller, J., & Jennings, V. A. (1983). Motor Cortex control of finely graded forces. *Journal of Neurophysiology*, *49*(5), 1199–1215.

Farthing, J. P. (2009). Cross-education of strength depends on limb dominance: implications for theory and application. *Exercise and Sport Sciences Reviews*, *37*(33), 179–187.

Farthing, J. P., Borowsky, R., Chilibeck, P. D., Binsted, G., & Sarty, G. E. (2007). Neuro-

physiological adaptations associated with cross-education of strength. *Brain Topography*, *20*(2), 77–88.

Ferbert, A., Priori, A., Rothwell, J. C., Day, B. L., Colebatch, J. G., & Marsden, C. D. (1992). Interhemispheric inhibition of the human motor cortex. *The Journal of Physiology*, *453*, 525–546.

Fink, G. R., Frackowiak, R. S., Pietrzyk, U., & Passingham, R. E. (1997). Multiple nonprimary motor areas in the human cortex. *Journal of Neurophysiology*, *77*(4), 2164–2174.

Fling, B. W., Benson, B. L., & Seidler, R. D. (2013). Transcallosal sensorimotor fiber tract structure-function relationships. *Human Brain Mapping*, *34*(2), 384–95.

Fujiyama, H., Hinder, M. R., Schmidt, M. W., Garry, M. I., & Summers, J. J. (2012). Age-related differences in corticospinal excitability and inhibition during coordination of upper and lower limbs. *Neurobiology of Aging*, *33*(7), 1484.e1-14.

Fulton, J. F. (1935). A note on the definition of the “motor” and “premotor” areas. *Brain*, *58*(2), 311–316.

Gandevia, S. C. (1985). Illusory movements produced by electrical stimulation of low-threshold muscle afferents from the hand. *Brain : A Journal of Neurology*, *108* (Pt 4), 965–81.

Gandevia, S. C. (2001). Spinal and Supraspinal Factors in Human Muscle Fatigue. *Physiol Rev*, *81*(4), 1725–1789.

Geffen, G. M., Jones, D. L., & Geffen, L. B. (1994). Interhemispheric control of manual motor activity. *Behavioural Brain Research*, *64*(1–2), 131–40.

Gerloff, C., Cohen, L. G., Floeter, M. K., Chen, R., Corwell, B., & Hallett, M. (1998). Inhibitory influence of the ipsilateral motor cortex on responses to stimulation of the human cortex and pyramidal tract. *The Journal of Physiology*, *510*(Pt 1), 249–259.

Gerloff, C., Corwell, B., Chen, R., Hallett, M., & Cohen, L. G. (1998). The role of the human motor cortex in the control of complex and simple finger movement sequences. *Brain : A Journal of Neurology*, *121*(Pt 9), 1695–1709.

Gilhodes, J. C., Roll, J. P., & Tardy-Gervet, M. F. (1986). Perceptual and motor effects of agonist-antagonist muscle vibration in man. *Experimental Brain Research*, *61*(2), 395–402.

Gould, H. J., Cusick, C. G., Pons, T. P., & Kaas, J. H. (1986). The relationship of corpus callosum connections to electrical stimulation maps of motor, supplementary motor, and the frontal eye fields in owl monkeys. *The Journal of Comparative Neurology*, *247*(3), 297–325.

Grafton, S. T., Woods, R. P., & Mazziotta, J. C. (1993). Within-arm somatotopy in human motor areas determined by positron emission tomography imaging of cerebral blood flow. *Experimental Brain Research*, *95*(1), 172–6.

Grafton, S. T., Woods, R. P., Mazziotta, J. C., & Phelps, M. E. (1991). Somatotopic mapping of the primary motor cortex in humans: activation studies with cerebral blood flow and positron emission tomography. *Journal of Neurophysiology*, *66*(3), 735–43.

Hall, E. J., Flament, D., Fraser, C., & Lemon, R. N. (1990). Non-invasive brain stimulation reveals reorganized cortical outputs in amputees. *Neuroscience*

Letters, *116*(3), 379–86.

Hallett, M. (2003). Chapter 13 Surround inhibition. *Supplements to Clinical Neurophysiology*, *56*(C), 153–159.

Hallett, M. (2007). Transcranial magnetic stimulation: a primer. *Neuron*, *55*(2), 187–99.

Hanajima, R., Ugawa, Y., Machii, K., Mochizuki, H., Terao, Y., Enomoto, H., … Kanazawa, I. (2001). Interhemispheric facilitation of the hand motor area in humans. *The Journal of Physiology*, *531*(Pt 3), 849–859.

Hellebrandt, F. A. (1951). Cross education; ipsilateral and contralateral effects of unimanual training. *Journal of Applied Physiology*, *4*(2), 136–44.

Hess, C. W., Mills, K. R., & Murray, N. M. (1986). Magnetic stimulation of the human brain: facilitation of motor responses by voluntary contraction of ipsilateral and contralateral muscles with additional observations on an amputee. *Neuroscience Letters*, *71*(2), 235–240.

Hess, C. W., Mills, K. R., & Murray, N. M. (1987). Responses in small hand muscles from magnetic stimulation of the human brain. *The Journal of Physiology*, *388*, 397–419.

Hess, G., Aizenman, C. D., & Donoghue, J. P. (1996). Conditions for the induction of long-term potentiation in layer II/III horizontal connections of the rat motor cortex. *Journal of Neurophysiology*, *75*(5), 1765–78.

Hess, G., & Donoghue, J. P. (1994). Long-term potentiation of horizontal connections provides a mechanism to reorganize cortical motor maps. *Journal of Neurophysiology*, *71*(6), 2543–2547.

Hess, G., & Donoghue, J. P. (1996). Long-term potentiation and long-term depression of horizontal connections in rat motor cortex. *Acta Neurobiologiae Experimentalis*, *56*(1), 397–405.

Hinder, M. R., Schmidt, M. W., Garry, M. I., Carroll, T. J., & Summers, J. J. (2011). Absence of cross-limb transfer of performance gains following ballistic motor practice in older adults. *Journal of Applied Physiology*, *110*(1), 166–75.

Hinder, M. R., Schmidt, M. W., Garry, M. I., & Summers, J. J. (2010). Unilateral contractions modulate interhemispheric inhibition most strongly and most adaptively in the homologous muscle of the contralateral limb. *Experimental Brain research, 205*(3), 423-33.

Hiraga, C. Y., Summers, J. J., & Temprado, J. J. (2004). Attentional costs of coordinating homologous and non-homologous limbs. *Human Movement Science*, *23*(3–4), 415–430.

Hiraga, C. Y., Summers, J. J., & Temprado, J. J. (2005). Effects of attentional prioritisation on the temporal and spatial components of an interlimb circle-drawing task. *Human Movement Science*, *24*(5–6), 815–832.

Hopf, H. C., Schlegel, H. J., & Lowitzsch, K. (1974). Irradiation of Voluntary Activity to the Contralateral Side in Movements of Normal Subjects and Patients with Central Motor Disturbances. *European Neurology*, *12*(3), 142–147.

Hortobágyi, T., Dempsey, L., Fraser, D., Zheng, D., Hamilton, G., Lambert, J., & Dohm, L. (2000). Changes in muscle strength, muscle fibre size and myofibrillar gene expression after immobilization and retraining in humans. *The Journal of Physiology*, *524*(1), 293–304.

Hortobágyi, T., Richardson, S. P., Lomarev, M., Shamim, E., Meunier, S., Russman, H., … Hallett, M. (2011). Interhemispheric plasticity in humans. *Medicine and Science in Sports and Exercise*, *43*(7), 1188–99.

Hortobágyi, T., Scott, K., Lambert, J., Hamilton, G., & Tracy, J. (1999). Cross-education of muscle strength is greater with stimulated than voluntary contractions. *Motor Control*, *3*(2), 205–19.

Hortobagyi, T., Taylor, J. L., Petersen, N. T., Russell, G., & Gandevia, S. C. (2003). Changes in segmental and motor cortical output with contralateral muscle contractions and altered sensory inputs in humans. *Journal of Neurophysiology*, *90*(4), 2451–2459.

Howatson, G., Taylor, M. B., Rider, P., Motawar, B. R., Mcnally, M. P., Solnik, S., … Hortobagyi, T. (2011). Ipsilateral motor cortical responses to TMS during lengthening and shortening of the contralateral wrist flexors. *European Journal of Neuroscience*, *33*(5), 978–990.

Huntley, G. W., & Jones, E. G. (1991). Relationship of intrinsic connections to forelimb movement representations in monkey motor cortex: a correlative anatomic and physiological study. *Journal of Neurophysiology*, *66*(2), 390–413.

Ibey, R. J., Bolton, D. A. E., Buick, A. R., Staines, W. R., & Carson, R. G. (2015). Interhemispheric inhibition of corticospinal projections to forearm muscles. *Clinical Neurophysiology*, *126*(10), 1934–1940.

Ibey, R. J., & Staines, W. R. (2013). Corticomotor excitability changes seen in the resting forearm during contralateral rhythmical movement and force manipulations: A TMS study. *Behavioural Brain Research*, *257*, 265–274.

Imamizu, H., & Shimojo, S. (1995). The locus of visual-motor learning at the task or manipulator level: implications from intermanual transfer. *Journal of Experimental Psychology*, *21*(4), 719–33.

Irlbacher, K., Brocke, J., Mechow, J. V, & Brandt, S. A. (2007). Effects of GABA(A) and GABA(B) agonists on interhemispheric inhibition in man. *Clinical Neurophysiology*, *118*(2), 308–16.

Jacobs, K. M., & Donoghue, J. P. (1991). Reshaping the cortical motor map by unmasking latent intracortical connections. *Science*, *251*(4996), 944–7.

Jeka, J. J., & Kelso, J. A. (1995). Manipulating symmetry in the coordination dynamics of human movement. *Journal of Experimental Psychology. Human Perception and Performance*, *21*(2), 360–74.

Jones, E. G., & Porter, R. (1980). What is area 3a? *Brain Research*, *203*(1), 1–43.

Kaelin-Lang, A., Luft, A. R., Sawaki, L., Burstein, A. H., Sohn, Y. H., & Cohen, L. G. (2002). Modulation of human corticomotor excitability by somatosensory input. *The Journal of Physiology*, *540*(Pt 2), 623–33.

Kammer, T., Beck, S., Erb, M., & Grodd, W. (2001). The influence of current direction on phosphene thresholds evoked by transcranial magnetic stimulation. *Clinical Neurophysiology*, *112*(11), 2015–21.

Kelso, J. A., & Jeka, J. J. (1992). Symmetry breaking dynamics of human multilimb coordination. *Journal of Experimental Psychology. Human Perception and Performance*, *18*(3), 645–68.

Kim, J. A., Eliassen, J. C., & Sanes, J. N. (2005). Movement quantity and frequency coding in human motor areas. *Journal of Neurophysiology*, *94*(4), 2504–11.

Kleinschmidt, A., Nitschke, M. F., & Frahm, J. (1997). Somatotopy in the human motor cortex hand area. A high-resolution functional MRI study. *The European Journal of Neuroscience*, *9*(10), 2178–86.

Kobayashi, M., Hutchinson, S., Schlaug, G., & Pascual-Leone, A. (2003). Ipsilateral motor cortex activation on functional magnetic resonance imaging during unilateral hand movements is related to interhemispheric interactions. *NeuroImage*, *20*(4), 2259–2270.

Kolarik, R. C., Rasey, S. K., & Wall, J. T. (1994). The consistency, extent, and locations of early-onset changes in cortical nerve dominance aggregates following injury of nerves to primate hands. *The Journal of Neuroscience*, *14*(7), 4269–88.

Kossev, A., Siggelkow, S., Kapels, H., Dengler, R., & Rollnik, J. D. (2001). Crossed effects of muscle vibration on motor-evoked potentials. *Clinical Neurophysiology*, *112*(3), 453–6.

Kossev, A., Siggelkow, S., Schubert, M., Wohlfarth, K., & Dengler, R. (1999). Muscle vibration: different effects on transcranial magnetic and electrical stimulation. *Muscle & Nerve*, *22*(7), 946–8.

Kujirai, T., Caramia, M. D., Rothwell, J. C., Day, B. L., Thompson, P. D., Ferbert, A., … Marsden, C. D. (1993). Corticocortical inhibition in human motor cortex. *The Journal of Physiology*, *471*, 501–519.

Kwan, H. C., Mackay, W. A., Murphy, J. T., & Wong, Y. C. (1978). An intracortical microstimulation study of output organization in precentral cortex of awake primates. *Journal de Physiologie*, *74*(3), 231–3.

Laszlo, J. I., Baguley, R. A., & Bairstow, P. J. (1970). Bilateral transfer in tapping skill

in the absence of peripheral information. *Journal of Motor Behavior*, *2*(4), 261–71.

Lee, M., & Carroll, T. J. (2007). Cross education: possible mechanisms for the contralateral effects of unilateral resistance training. *Sports Medicine*, *37*(1), 1–14.

Lee, M., Gandevia, S. C., & Carroll, T. J. (2009). Unilateral strength training increases voluntary activation of the opposite untrained limb. *Clinical Neurophysiology*, *120*(4), 802–808.

Lewis, G. N., & Byblow, W. D. (2002). Modulations in corticomotor excitability during passive upper-limb movement: Is there a cortical influence? *Brain Research*, *943*(2), 263–275.

Lewis, G. N., Byblow, W. D., & Carson, R. G. (2001). Phasic modulation of corticomotor excitability during passive movement of the upper limb: effects of movement frequency and muscle specificity. *Brain Research*, *900*(2), 282–294.

Liepert, J., Dettmers, C., Terborg, C., & Weiller, C. (2001). Inhibition of ipsilateral motor cortex during phasic generation of low force. *Clinical Neurophysiology*, *112*(1), 114–121.

Liepert, J., Tegenthoff, M., & Malin, J. P. (1995). Changes of cortical motor area size during immobilization. *Electroencephalography and Clinical Neurophysiology*, *97*(6), 382–6.

Lin, K. -c., Chen, Y. -a., Chen, C. -l., Wu, C. -y., & Chang, Y. -f. (2010). The Effects of Bilateral Arm Training on Motor Control and Functional Performance in Chronic Stroke: A Randomized Controlled Study. *Neurorehabilitation and*

Neural Repair, *24*(1), 42–51.

Liu, J., Morel, A., Wannier, T., & Rouiller, E. M. (2002). Origins of callosal projections to the supplementary motor area (SMA): a direct comparison between pre-SMA and SMA-proper in macaque monkeys. *The Journal of Comparative Neurology*, *443*(1), 71–85.

Macefield, V. G., Gandevia, S. C., Bigland-Ritchie, B., Gorman, R. B., & Burke, D. (1993). The firing rates of human motoneurones voluntarily activated in the absence of muscle afferent feedback. *The Journal of Physiology*, *471*, 429–43.

Magnus, C. R. A., Barss, T. S., Lanovaz, J. L., & Farthing, J. P. (2010). Effects of cross-education on the muscle after a period of unilateral limb immobilization using a shoulder sling and swathe. *Journal of Applied Physiology*, *109*(6), 1887–94.

Martin, B. J., & Park, H. S. (1997). Analysis of the tonic vibration reflex: influence of vibration variables on motor unit synchronization and fatigue. *European Journal of Applied Physiology and Occupational Physiology*, *75*(6), 504–11.

Matsunami, K., & Hamada, I. (1981). Characteristics of the ipsilateral movement-related neuron in the motor cortex of the monkey. *Brain Research*, *204*(1), 29–42.

Mayston, M. J., Harrison, L. M., & Stephens, J. A. (1999). A neurophysiological study of mirror movements in adults and children. *Annals of Neurology*, *45*(5), 583–594.

McCombe Waller, S., & Whitall, J. (2008). Bilateral arm training: why and who benefits? *NeuroRehabilitation*, *23*(1), 29–41.

McIntyre, A. K., Proske, U., & Rawson, J. A. (1984). Cortical projection of afferent information from tendon organs in the cat. *The Journal of Physiology*, *354*, 395–

406.

McKiernan, B. J., Marcario, J. K., Karrer, J. H., & Cheney, P. D. (1998). Corticomotoneuronal postspike effects in shoulder, elbow, wrist, digit, and intrinsic hand muscles during a reach and prehension task. *Journal of Neurophysiology*, *80*(4), 1961–80.

Meesen, R. L. J., Wenderoth, N., Temprado, J. J., Summers, J. J., & Swinnen, S. P. (2006). The coalition of constraints during coordination of the ipsilateral and heterolateral limbs. *Experimental Brain Research*, *174*(2), 367–375.

Merton, P. A., & Morton, H. B. (1980). Stimulation of the cerebral cortex in the intact human subject. *Nature*, *285*(5762), 227.

Meyer, B.-U., Röricht, S., von Einsiedel, H. G., Kruggel, F., & Weindl, A. (1995). Inhibitory and excitatory interhemispheric transfers between motor cortical areas in normal humans and patients with abnormalities of the corpus callosum. *Brain*, *118*(2), 429–440.

Muellbacher, W., Facchini, S., Boroojerdi, B., & Hallett, M. (2000). Changes in motor cortex excitability during ipsilateral hand muscle activation in humans. *Clinical Neurophysiology*, *111*(2), 344–349.

Munn, J., Herbert, R. D., & Gandevia, S. C. (2004). Contralateral effects of unilateral resistance training: a meta-analysis. *Journal of Applied Physiology*, *96*(5), 1861–6.

Murphy, J. T., Wong, Y. C., & Kwan, H. C. (1974). Distributed feedback systems for muscle control. *Brain Research*, *71*(2–3), 495–505.

Murthy, V. N., & Fetz, E. E. (1996). Oscillatory activity in sensorimotor cortex of

awake monkeys: synchronization of local field potentials and relation to behavior. *Journal of Neurophysiology*, *76*(6), 3949–67.

Nelson, A. J., Hoque, T., Gunraj, C., Ni, Z., & Chen, R. (2009). Bi-directional interhemispheric inhibition during unimanual sustained contractions. *BMC Neuroscience*, *10*, 31.

Nudo, R. J., Jenkins, W. M., Merzenich, M. M., Prejean, T., & Grenda, R. (1992). Neurophysiological correlates of hand preference in primary motor cortex of adult squirrel monkeys. *The Journal of Neuroscience*, *12*(8), 2918–47.

Nudo, R. J., & Milliken, G. W. (1996). Reorganization of movement representations in primary motor cortex following focal ischemic infarcts in adult squirrel monkeys. *Journal of Neurophysiology*, *75*(5), 2144–9.

Nudo, R. J., Milliken, G. W., Jenkins, W. M., & Merzenich, M. M. (1996). Use-dependent alterations of movement representations in primary motor cortex of adult squirrel monkeys. *The Journal of Neuroscience*, *16*(2), 785–807.

Oldfield, R. C. (1971). The assessment and analysis of handedness: the Edinburgh inventory. *Neuropsychologia*, *9*(1), 97–113.

Parlow, S. E., & Kinsbourne, M. (1989). Asymmetrical transfer of training between hands: implications for interhemispheric communication in normal brain. *Brain and Cognition*, *11*(1), 98–113.

Pearce, A. J., Hendy, A., Bowen, W. A., & Kidgell, D. J. (2013). Corticospinal adaptations and strength maintenance in the immobilized arm following 3 weeks unilateral strength training. *Scandinavian Journal of Medicine & Science in Sports*, *23*(6), 740–8.

Penfield, W., & Rasmussen, T. (1950). The Cerebral Cortex of Man. A Clinical Study of Localization of Function. *Academic Medicine*, *25*(5), 375.

Perez, M. A., & Cohen, L. G. (2008). Mechanisms underlying functional changes in the primary motor cortex ipsilateral to an active hand. *The Journal of Neuroscience*, *28*(22), 5631–5640.

Perez, M. A., & Cohen, L. G. (2009). Scaling of motor cortical excitability during unimanual force generation. *Cortex*, *45*(9), 1065–1071.

Picard, N., & Strick, P. L. (1996). Motor areas of the medial wall: A review of their location and functional activation. *Cerebral Cortex*, *6*(3), 342-53.

Picard, N., & Strick, P. L. (2001). Imaging the premotor areas. *Current Opinion in Neurobiology*, *11*(6), 663-72.

Plow, E. B., Arora, P., Pline, M. A., Binenstock, M. T., & Carey, J. R. (2010). Within-limb somatotopy in primary motor cortex--revealed using fMRI. *Cortex*, *46*(3), 310–21.

Qi, H. X., Stepniewska, I., & Kaas, J. H. (2000). Reorganization of primary motor cortex in adult macaque monkeys with long-standing amputations. *Journal of Neurophysiology*, *84*(4), 2133–47.

Ralston, D. D., & Ralston, H. J. (1985). The terminations of corticospinal tract axons in the macaque monkey. *The Journal of Comparative Neurology*, *242*(3), 325–337.

Rasmussen, T., & Penfield, W. (1947). The human sensorimotor cortex as studied by electrical stimulation. *Federation Proceedings*, *6*(1 Pt 2), 184.

Reitz, M., & Müller, K. (1998). Differences between `congenital mirror movements'

and `associated movements' in normal children: a neurophysiological case study. *Neuroscience Letters*, *256*(2), 69–72.

Ridding, M. C., Brouwer, B., Miles, T. S., Pitcher, J. B., & Thompson, P. D. (2000). Changes in muscle responses to stimulation of the motor cortex induced by peripheral nerve stimulation in human subjects. *Experimental Brain Research*, *131*(1), 135–43.

Ridding, M. C., & Taylor, J. L. (2001). Mechanisms of motor-evoked potential facilitation following prolonged dual peripheral and central stimulation in humans. *The Journal of Physiology*, *537*(Pt 2), 623–31.

Roll, J. P., & Gilhodes, J. C. (1995). Proprioceptive sensory codes mediating movement trajectory perception: human hand vibration-induced drawing illusions. *Canadian Journal of Physiology and Pharmacology*, *73*(2), 295–304.

Roll, J. P., Vedel, J. P., & Ribot, E. (1989). Alteration of proprioceptive messages induced by tendon vibration in man: a microneurographic study. *Experimental Brain Research*, *76*(1), 213–22.

Rosenkranz, K., Altenmüller, E., Siggelkow, S., & Dengler, R. (2000). Alteration of sensorimotor integration in musician's cramp: impaired focusing of proprioception. *Clinical Neurophysiology*, *111*(11), 2040–5.

Rosenkranz, K., & Rothwell, J. C. (2003). Differential effect of muscle vibration on intracortical inhibitory circuits in humans. *The Journal of Physiology*, *551*(Pt 2), 649–60.

Rossini, P. M., & Rossi, S. (2007). Transcranial magnetic stimulation. *Neurology*, *68*(7), 484–488.

Rothwell, J. C. (1997). Techniques and mechanisms of action of transcranial stimulation of the human motor cortex. *Journal of Neuroscience Methods*, *74*(2), 113–122.

Rothwell, J. C., Thompson, P. D., Day, B. L., Boyd, S., & Marsden, C. D. (1991). Stimulation of the Human Motor Cortex through the Scalp. *Experimental Physiology*, *76*(2), 159–200.

Rouiller, E. M., Babalian, A., Kazennikov, O., Moret, V., Yu, X. H., & Wiesendanger, M. (1994). Transcallosal connections of the distal forelimb representations of the primary and supplementary motor cortical areas in macaque monkeys. *Experimental Brain research*, *102*(2), 227–243.

Rudiak, D., & Marg, E. (1994). Finding the depth of magnetic brain stimulation: a re-evaluation. *Electroencephalography and Clinical Neurophysiology/ Evoked Potentials*, *93*(5), 358–371.

Sadato, N., Ibanez, V., Deiber, M.-P., Campbell, G., Leonardo, M., & Hallett, M. (1996). Frequency-Dependent Changes of Regional Cerebral Blood Flow During Finger Movements. *Journal of Cerebral Blood Flow & Metabolism*, *16*(1), 23–33.

Sanes, J. N., & Donoghue, J. P. (1997). Static and dynamic organization of motor cortex. *Adv Neurol*, *73*, 277–296.

Sanes, J. N., & Donoghue, J. P. (2000). Plasticity and primary motor cortex. *Annual Review of Neuroscience*, *23*, 393–415.

Sanes, J. N., Wang, J., & Donoghue, J. P. (1992). Immediate and delayed changes of rat motor cortical output representation with new forelimb configurations. *Cerebral Cortex*, *2*(2), 141–152.

Schaal, S., Sternad, D., Osu, R., & Kawato, M. (2004). Rhythmic arm movement is not discrete. *Nature Neuroscience*, *7*(10), 1136–1143.

Scripture, E. W., Smith, T. L., & Brown, E. M. (1894). On the education of muscular control and power. *Yale Psychol Studies*, *2*, 114–119.

Sekiguchi, H., Kimura, T., Yamanaka, K., & Nakazawa, K. (2001). Lower excitability of the corticospinal tract to transcranial magnetic stimulation during lengthening contractions in human elbow flexors. *Neuroscience Letters*, *312*(2), 83–86.

Sekiguchi, H., Nakazawa, K., & Suzuki, S. (2003). Differences in recruitment properties of the corticospinal pathway between lengthening and shortening contractions in human soleus muscle. *Brain Research*, *977*(2), 169–179.

Shinoda, Y., Zarzecki, P., & Asanuma, H. (1979). Spinal branching of pyramidal tract neurons in the monkey. *Experimental Brain Research*, *34*(1), 59–72.

Siggelkow, S., Kossev, A., Schubert, M., Kappels, H. H., Wolf, W., & Dengler, R. (1999). Modulation of motor evoked potentials by muscle vibration: the role of vibration frequency. *Muscle & Nerve*, *22*(11), 1544–8.

Sohn, Y. H., & Hallett, M. (2004). Surround inhibition in human motor system. *Experimental Brain Research*, *158*(4), 397–404.

Sohn, Y. H., Jung, H. Y., Kaelin-Lang, A., & Hallett, M. (2003). Excitability of the ipsilateral motor cortex during phasic voluntary hand movement. *Experimental Brain research*, *148*(2), 176–185.

Sohn, Y. H., Kang, S. Y., & Hallett, M. (2005). Corticospinal disinhibition during dual action. *Experimental Brain Research*, *162*(1), 95–9.

Staines, W. R., McIlroy, W. E., Graham, S. J., & Black, S. E. (2001). Bilateral movement

enhances ipsilesional cortical activity in acute stroke: a pilot functional MRI study. *Neurology*, *56*(3), 401–404.

Stedman, A., Davey, N. J., & Ellaway, P. H. (1998). Facilitation of human first dorsal interosseous muscle responses to transcranial magnetic stimulation during voluntary contraction of the contralateral homonymous muscle. *Muscle & Nerve*, *21*(8), 1033–1039.

Stefan, K., Kunesch, E., Cohen, L. G., Benecke, R., & Classen, J. (2000). Induction of plasticity in the human motor cortex by paired associative stimulation. *Brain*, *123*(Pt 3), 572–584.

Steyvers, M., Levin, O., Van Baelen, M., & Swinnen, S. P. (2003). Corticospinal excitability changes following prolonged muscle tendon vibration. *Neuroreport*, *14*(15), 1901–5.

Stinear, C. M., Barber, P. A., Coxon, J. P., Fleming, M. K., & Byblow, W. D. (2008). Priming the motor system enhances the effects of upper limb therapy in chronic stroke. *Brain: A Journal of Neurology*, *131*(Pt 5), 1381–90.

Stinear, C. M., Walker, K. S., & Byblow, W. D. (2001). Symmetric facilitation between motor cortices during contraction of ipsilateral hand muscles. *Experimental Brain research*, *139*(1), 101–105.

Stinear, J. W., & Byblow, W. D. (2002). Disinhibition in the human motor cortex is enhanced by synchronous upper limb movements. *The Journal of Physiology*, *543*(Pt 1), 307–316.

Summers, J. J., Kagerer, F. A., Garry, M. I., Hiraga, C. Y., Loftus, A., & Cauraugh, J. H. (2007). Bilateral and unilateral movement training on upper limb function in

chronic stroke patients: A TMS study. *Journal of the Neurological Sciences*, *252*(1), 76–82.

Swayne, O., Rothwell, J., & Rosenkranz, K. (2006). Transcallosal sensorimotor integration: effects of sensory input on cortical projections to the contralateral hand. *Clinical Neurophysiology*, *117*(4), 855–863.

Swinnen, S. P. (2002). Intermanual coordination: from behavioural principles to neural-network interactions. *Nature Reviews. Neuroscience*, *3*(5), 348–59.

Swinnen, S. P., Dounskaia, N., Verschueren, S., Serrien, D. J., & Daelman, A. (1995). Relative phase destabilization during interlimb coordination: the disruptive role of kinesthetic afferences induced by passive movement. *Experimental Brain Research*, *105*(3), 439–454.

Takahashi, K., Maruyama, A., Hirakoba, K., Maeda, M., Etoh, S., Kawahira, K., & Rothwell, J. C. (2011). Fatiguing intermittent lower limb exercise influences corticospinal and corticocortical excitability in the nonexercised upper limb. *Brain Stimulation*, *4*(2), 90–6.

Takahashi, K., Maruyama, A., Maeda, M., Etoh, S., Hirakoba, K., Kawahira, K., & Rothwell, J. C. (2009). Unilateral grip fatigue reduces short interval intracortical inhibition in ipsilateral primary motor cortex. *Clinical Neurophysiology*, *120*(1), 198–203.

Tanji, J., Okano, K., & Sato, K. C. (1988). Neuronal activity in cortical motor areas related to ipsilateral, contralateral, and bilateral digit movements of the monkey. *Journal of Neurophysiology*, *60*(1), 325–343.

Tazoe, T., Endoh, T., Nakajima, T., Sakamoto, M., & Komiyama, T. (2007).

Disinhibition of upper limb motor area by voluntary contraction of the lower limb muscle. *Experimental Brain Research*, *177*(3), 419–30.

Tazoe, T., Sakamoto, M., Nakajima, T., Endoh, T., Shiozawa, S., & Komiyama, T. (2009). Remote facilitation of supraspinal motor excitability depends on the level of effort. *European Journal of Neuroscience*, *30*(7), 1297–1305.

Terao, Y., & Ugawa, Y. (2002). Basic mechanisms of TMS. *Journal of Clinical Neurophysiology*, *19*(4), 322–43.

Thickbroom, G. W., Phillips, B. A., Morris, I., Byrnes, M. L., & Mastaglia, F. L. (1998). Isometric force-related activity in sensorimotor cortex measured with functional MRI. *Experimental Brain research*, *121*(1), 59–64.

Tinazzi, M., & Zanette, G. (1998). Modulation of ipsilateral motor cortex in man during unimanual finger movements of different complexities. *Neuroscience Letters*, *244*(3), 121–124.

Topka, H., Cohen, L. G., Cole, R. a, & Hallett, M. (1991). Reorganization of corticospinal pathways following spinal cord injury. *Neurology*, *41*(8), 1276–1283.

Tsuboi, F., Nishimura, Y., Yoshino-Saito, K., & Isa, T. (2010). Neuronal mechanism of mirror movements caused by dysfunction of the motor cortex. *The European Journal of Neuroscience*, *32*(8), 1397–406.

Uehara, K., Morishita, T., & Funase, K. (2011). Excitability changes in the ipsilateral primary motor cortex during rhythmic contraction of finger muscles. *Neuroscience Letters*, *488*(1).

Uehara, K., Morishita, T., Kubota, S., & Funase, K. (2013). Neural mechanisms

underlying the changes in ipsilateral primary motor cortex excitability during unilateral rhythmic muscle contraction. *Behavioural Brain Research*, *240*, 33–45.

Uematsu, A., Obata, H., Endoh, T., Kitamura, T., Hortobágyi, T., Nakazawa, K., & Suzuki, S. (2010). Asymmetrical modulation of corticospinal excitability in the contracting and resting contralateral wrist flexors during unilateral shortening, lengthening and isometric contractions. *Experimental Brain Research*, *206*(1), 59–69.

van Duinen, H., Renken, R., Maurits, N. M., & Zijdewind, I. (2008). Relation between muscle and brain activity during isometric contractions of the first dorsal interosseus muscle. *Human Brain Mapping*, *29*(3), 281–299.

Wassermann, E. M., Pascual-Leone, A., & Hallett, M. (1994). Cortical motor representation of the ipsilateral hand and arm. *Experimental Brain research*, *100*(1), 121–132.

Werner, W., Bauswein, E., & Fromm, C. (1991). Static firing rates of premotor and primary motor cortical neurons associated with torque and joint position. *Experimental Brain Research*, *86*(2), 293–302.

White, E. L., & Keller, A. (1989). *Cortical circuits : synaptic organization of the cerebral cortex--structure, function, and theory*. Birkhäuser.

Wiesendanger, M., & Miles, T. S. (1982). Ascending pathway of low-threshold muscle afferents to the cerebral cortex and its possible role in motor control. *Physiological Reviews*, *62*(4 Pt 1), 1234–70.

Witt, S. T., Laird, A. R., & Meyerand, M. E. (2008). Functional neuroimaging correlates

of finger-tapping task variations: An ALE meta-analysis. *NeuroImage*, *42*(1), 343–356.

Yahagi, S., Ni, Z., Takahashi, M., Takeda, Y., Tsuji, T., & Kasai, T. (2003). Excitability changes of motor evoked potentials dependent on muscle properties and contraction modes. *Motor Control*, *7*(4), 328–45.

Zhou, S. (2000). Chronic neural adaptations to unilateral exercise: mechanisms of cross education. *Exercise and Sport Sciences Reviews*, *28*(4), 177–84.

Ziemann, U., Rothwell, J. C., & Ridding, M. C. (1996). Interaction between intracortical inhibition and facilitation in human motor cortex. *The Journal of Physiology*, *496*(Pt 3), 873–81.

Zwarts, M. J. (1992). Central motor conduction in relation to contra- and ipsilateral activation. *Electroencephalography and Clinical Neurophysiology*, *85*(6), 425–428.