THE EFFECTS OF ACUTE AEROBIC EXERCISE ON MOTOR SKILL LEARNING AND NEUROPHYSIOLOGY IN HEALTHY OLDER ADULTS

by

Beatrice Alexandra Francisco

B.Kin., The University of British Columbia, 2016

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

in

THE FACULTY OF GRADUATE AND POSTDOCTORAL STUDIES

(Rehabilitation Sciences)

THE UNIVERSITY OF BRITISH COLUMBIA

(Vancouver)

May 2018

© Beatrice Alexandra Francisco, 2018
The following individuals certify that they have read, and recommend to the Faculty of Graduate and Postdoctoral Studies for acceptance, a thesis entitled:

The effects of acute aerobic exercise on motor skill learning and neurophysiology in healthy older adults

submitted by Beatrice Francisco in partial fulfillment of the requirements

for the degree of Master of Science

in Rehabilitation Sciences

Examinining Committee:

Dr. Lara Boyd

Supervisor

Dr. Kristin Campbell

Supervisory Committee Member

Dr. Nicola Hodges

Supervisory Committee Member

Dr. Marc Roig

Additional Examiner
Abstract

Aging is associated with a reduced ability to perform motor tasks, as well as a decreased capacity for neuroplastic change and motor skill learning. Evidence in young healthy adults suggests a single session of aerobic exercise can facilitate motor learning and changes in movement-related neurophysiological circuits. However, we do not know whether these effects extend to healthy older adults. The objectives of the present thesis were to examine whether a single session of moderate-intensity aerobic exercise 1) facilitates motor skill acquisition and consolidation, and 2) modulates motor cortical and intracortical circuitry, in healthy older adults.

Twenty-two participants (55-75 years old) completed a maximal exercise stress test at least two days prior to all other sessions. Next, participants practiced a motor sequence task after 20-minutes of moderate-intensity cycling or 20-minutes of seated rest, on separate occasions. To assess motor learning, participants performed the motor task 24 hours later at a no-exercise retention test. On a separate day, neurophysiological measures using transcranial magnetic stimulation were obtained at two time-points prior to and two time-points following an acute bout of moderate-intensity cycling.

Performing aerobic exercise immediately prior to task practice did not yield any statistically significant differences in measures of motor skill acquisition or consolidation. However, we observed a non-significant trend towards improvements in motor memory consolidation, such that under the exercise condition there were greater improvements in repeated sequences compared to rest. Additionally, we found that after exercise there was an increase in long-interval intracortical inhibition (LICI), which returned to near baseline levels.
within 30 minutes post-exercise. Overall, these findings suggest that a single bout of
moderate-intensity aerobic exercise transiently modulates GABA-B mediated intracortical
inhibition in healthy older adults, however, these exercise-induced neurophysiological effects
may not necessarily translate to changes in motor behaviour.

This work is the first to investigate the efficacy of an acute bout of aerobic exercise in
facilitating motor performance and learning, as well as modulating motor cortical and
intracortical circuits, in healthy older adults. Further understanding of how exercise influences
motor learning and neurophysiology in the aging brain will be critical for the development of
potential rehabilitation strategies.
The ability to perform motor skills is important for completing daily activities, participating in sport, or responding to rehabilitation. Age-related changes in movement-related neural circuits are associated with a reduced ability to perform and learn motor tasks. Evidence in young healthy adults suggests performing aerobic exercise immediately before practicing a motor skill improves learning of the motor skill. Further, a single session of aerobic exercise influences movement-related neural circuits in young healthy adults. However, whether these effects extend to healthy older adults remains unknown. In this thesis, we examine whether performing aerobic exercise immediately prior to motor practice can improve motor learning in healthy older adults, compared to practicing without prior exercise. We also examine how aerobic exercise influences movement-related neural circuits in healthy older adults. The results from this project can be used to inform the development of rehabilitation strategies in aging and clinical populations.
Preface

This thesis contains the work of research studies conducted by the candidate, Beatrice A. Francisco, under the supervision of Dr. Lara Boyd, with guidance from Drs. Nicola Hodges and Kristin Campbell. Experimental design and conception, data acquisition and analysis, data interpretation, and documentation were primarily the work of the candidate. Drs. Lara Boyd, Nicola Hodges, and Kristin Campbell provided direction, support, and critical feedback on the design of the study. Drs. Jason Neva and Katlyn Brown assisted with data collection, analyses and editing the manuscript. The authors would also like to thank Asha Toner and Kelcey Bland for their tremendous assistance with recruitment and data collection.

Sections of this thesis will be submitted for publication as a manuscript in a peer-reviewed journal.

All research described in this thesis was approved by the University of British Columbia’s Clinical Research Ethics Board (certificate # H17-01273) and the Vancouver Coastal Health Research Institute (certificate # H17-01273).
Table of Contents

Abstract ........................................................................................................................................ iii
Lay Summary .................................................................................................................................. v
Preface .......................................................................................................................................... vi
Table of Contents ............................................................................................................................ vii
List of Tables ................................................................................................................................... xi
List of Figures ................................................................................................................................... xii
List of Abbreviations ........................................................................................................................ xiii
Acknowledgements ............................................................................................................................ xiv
Dedication ........................................................................................................................................ xv

Chapter 1: Introduction ...................................................................................................................... 1

1.1 Preamble ......................................................................................................................................... 1

1.2 Motor learning concepts ................................................................................................................. 3

1.2.1 Phases of motor learning ........................................................................................................... 4

1.2.2 Age-related declines in motor performance and learning ......................................................... 5

1.3 Neuroplasticity underpins the process of motor learning ............................................................. 6

1.3.1 Potential neural mechanisms underlying age-related motor deficits ...................................... 10

1.4 Exercise as a potential strategy to facilitate motor learning ......................................................... 11

1.4.1 Possible mechanisms through which exercise facilitates motor learning ................................. 17

1.4.1.1 Acute aerobic exercise modulates movement-related neural circuits ........................................ 17

1.4.1.2 Exercise increases peripheral concentrations of neurochemicals ......................................... 19

1.5 Specific aims and hypotheses ........................................................................................................ 20
1.6 Rationale .......................................................................................................................... 20

Chapter 2: Methods .............................................................................................................. 22

2.1 Participants ..................................................................................................................... 22

2.2 Study design .................................................................................................................. 22

2.3 Exercise procedures ..................................................................................................... 23

2.3.1 Maximal stress testing ............................................................................................... 23

2.3.2 Standardized exercise bout ....................................................................................... 25

2.4 Study 1: Effects of acute aerobic exercise on motor performance and learning .......... 25

2.4.1 Serial targeting task procedures .............................................................................. 27

2.4.2 STT Analysis ............................................................................................................ 29

2.4.3 Statistical Analyses .................................................................................................. 29

2.4.3.1 Baseline performance ......................................................................................... 30

2.4.3.2 Motor skill acquisition and consolidation ......................................................... 30

2.5 Study 2: Acute aerobic exercise on motor cortical and intracortical circuitry ............ 30

2.5.1 Electromyography (EMG) ....................................................................................... 31

2.5.2 Transcranial Magnetic Stimulation (TMS) ............................................................. 32

2.5.3 TMS Analysis ......................................................................................................... 33

2.5.4 Statistical Analysis .................................................................................................. 33

2.5.4.1 Neurophysiological measures .......................................................................... 34

Chapter 3: Results ............................................................................................................... 35

3.1 Participants .................................................................................................................... 35

3.2 Study 1: Effects of acute aerobic exercise on motor performance and learning .......... 36

3.2.1 Baseline performance .............................................................................................. 36
3.2.2 Motor skill acquisition .......................................................... 36
3.2.3 Motor memory consolidation .................................................. 36
3.2.4 Recognition .......................................................................... 37
3.2.5 Effect size calculations .......................................................... 37
3.3 Study 2: Effects of acute aerobic exercise on neurophysiology ........... 39
  3.3.1 Baseline measures ................................................................. 39
  3.3.2 Neurophysiological measures ............................................... 39

Chapter 4: Discussion ........................................................................ 41
4.1 Study 1: Effects of acute exercise on motor performance and learning ........ 41
  4.1.1 Motor skill acquisition ............................................................ 41
  4.1.2 Motor memory consolidation ................................................... 44
  4.1.3 Limitations ........................................................................... 47
4.2 Study 2: Effects of acute exercise on motor cortical and intracortical networks.... 47
  4.2.1 Acute exercise modulated GABA-B, but not GABA-A mediated intracortical
      inhibition .............................................................................. 47
  4.2.2 Acute exercise did not impact intracortical facilitation or corticospinal excitability 49
  4.2.3 Limitations ........................................................................... 50
4.3 Conclusion and Future Directions .................................................. 51

References ....................................................................................... 53

Appendices ......................................................................................... 61
  Appendix A: Edinburgh Handedness Inventory .................................... 61
  Appendix B: Montreal Cognitive Assessment ....................................... 62
  Appendix C: TMS Screening Form ...................................................... 63
Appendix D: Exercise Stress Test Protocol .......................................................... 64
Appendix E: Borg Rating of Perceived Exertion (RPE) Scale ................................. 65
Appendix F: Individual exercise data .................................................................... 66
Appendix G: Calculation of ventilatory thresholds ............................................... 67
Appendix H: Sleep Quality Questionnaire ............................................................ 68
Appendix I: Godin Leisure Time Questionnaire .................................................... 69
List of Tables

Table 1.1 Neuropsychological measures using TMS ................................................................. 10
Table 1.2 Overview of studies on acute aerobic exercise and motor behavior ...................... 15
Table 2.1 Schedule for experimental studies ........................................................................... 23
Table 3.1 Participant demographics and exercise metrics ....................................................... 35
Table 3.2 Effect sizes for acquisition and consolidation .......................................................... 37
Table F.1 Individual exercise data ............................................................................................ 66
List of Figures

Figure 1.1 Schematic of transcranial magnetic stimulation (TMS) ............................................. 9
Figure 2.1 Overview of study design for Study 1 ................................................................. 26
Figure 2.2 Experimental set-up of the KINARM End-Point Robot™ ........................................ 28
Figure 2.3 Overview of study design for Study 2 ................................................................. 31
Figure 3.1 Study 1 Results ........................................................................................................ 38
Figure 3.2 Study 2 Results ........................................................................................................ 40
Figure G.1 Ventilatory thresholds ............................................................................................. 67
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>ARUC</td>
<td>area under the recruitment curve</td>
</tr>
<tr>
<td>BDNF</td>
<td>brain-derived neurotrophic factor</td>
</tr>
<tr>
<td>CO₂</td>
<td>carbon dioxide</td>
</tr>
<tr>
<td>CS</td>
<td>conditioning stimulus</td>
</tr>
<tr>
<td>ECG</td>
<td>electrocardiogram</td>
</tr>
<tr>
<td>ECR</td>
<td>extensor carpi radialis</td>
</tr>
<tr>
<td>EMG</td>
<td>electromyography</td>
</tr>
<tr>
<td>GABA</td>
<td>gamma-aminobutyric acid</td>
</tr>
<tr>
<td>HR</td>
<td>heart rate</td>
</tr>
<tr>
<td>HR&lt;sub&gt;max&lt;/sub&gt;</td>
<td>maximal heart rate</td>
</tr>
<tr>
<td>ICF</td>
<td>intracortical facilitation</td>
</tr>
<tr>
<td>IGF</td>
<td>insulin-like growth factor</td>
</tr>
<tr>
<td>ISI</td>
<td>interstimulus interval</td>
</tr>
<tr>
<td>LICI</td>
<td>long-interval intracortical inhibition</td>
</tr>
<tr>
<td>LTP</td>
<td>long-term potentiation</td>
</tr>
<tr>
<td>M1</td>
<td>primary motor cortex</td>
</tr>
<tr>
<td>MEP</td>
<td>motor evoked potential</td>
</tr>
<tr>
<td>MoCA</td>
<td>Montreal Cognitive Assessment</td>
</tr>
<tr>
<td>MSO</td>
<td>maximum stimulator output</td>
</tr>
<tr>
<td>NMDA</td>
<td>N-methyl-D-aspartate</td>
</tr>
<tr>
<td>O₂</td>
<td>oxygen</td>
</tr>
<tr>
<td>PAR-Q</td>
<td>physical activity readiness questionnaire</td>
</tr>
<tr>
<td>PO</td>
<td>power output</td>
</tr>
<tr>
<td>RER</td>
<td>respiratory exchange ratio</td>
</tr>
<tr>
<td>rmANOVA</td>
<td>repeated measures analysis of variance</td>
</tr>
<tr>
<td>RMSE</td>
<td>root mean square error</td>
</tr>
<tr>
<td>RMT</td>
<td>resting motor threshold</td>
</tr>
<tr>
<td>RPE</td>
<td>rating of perceived exertion</td>
</tr>
<tr>
<td>RC</td>
<td>recruitment curve</td>
</tr>
<tr>
<td>RPM</td>
<td>revolutions per minute</td>
</tr>
<tr>
<td>SD</td>
<td>standard deviation</td>
</tr>
<tr>
<td>SEM</td>
<td>standard error of the mean</td>
</tr>
<tr>
<td>SICI</td>
<td>short-interval intracortical inhibition</td>
</tr>
<tr>
<td>STT</td>
<td>serial targeting task</td>
</tr>
<tr>
<td>TMS</td>
<td>transcranial magnetic stimulation</td>
</tr>
<tr>
<td>TS</td>
<td>test stimulus</td>
</tr>
<tr>
<td>VEGF</td>
<td>vascular endothelial growth factor</td>
</tr>
<tr>
<td>VO₂</td>
<td>volume of oxygen consumption</td>
</tr>
<tr>
<td>VO₂&lt;sub&gt;peak&lt;/sub&gt;</td>
<td>peak oxygen consumption</td>
</tr>
<tr>
<td>VCO₂</td>
<td>volume of carbon dioxide</td>
</tr>
<tr>
<td>VT</td>
<td>ventilatory threshold</td>
</tr>
</tbody>
</table>
Acknowledgements

Firstly, I would like to acknowledge and thank my primary supervisor, Dr. Lara Boyd, for giving me the opportunity to pursue this research project, and for her mentorship and continued support. I am incredibly grateful to have had so many opportunities to learn and develop professionally throughout my MSc, with Dr. Boyd’s guidance. I would also like to sincerely thank my committee members, Drs. Kristin Campbell and Nicola Hodges, for their feedback and expertise. With the help of my entire supervisory committee, I was able to merge my interests in neuroscience, exercise physiology, and motor learning into one cohesive project.

To all Brain Behaviour Lab members, both past and present – I consider myself very lucky to have had the opportunity to learn from and work with all of you. Thank you for being so generous with your help and advice, for believing in me, and for making my experience working in a lab with no windows quite enjoyable. Special thanks to Drs. Jason Neva and Kate Brown, for always being there to answer my endless questions and for showing me the ropes throughout my MSc. To my fellow Rehabilitation Sciences graduate students, thank you for your friendships, and for being such a fun and supportive community. To JF, thank you for the kind words of encouragement, and for being such a positive influence in my life.

I’d also like to thank my family and friends for their constant love and support. Mom and Pop, I can’t thank you enough for everything you’ve done for me. To my siblings, Margo and Raffy, thank you for your words of wisdom and/or comic relief. And lastly, I’d like to thank to my grandmother, Araceli B. Avellana, for being such a constant source of inspiration.
Dedication

To my parents, my #1 supporters.
Chapter 1: Introduction

1.1 Preamble

Worldwide, the population is aging at a rapid pace. Globally, there were 900 million people aged 60 or over in 2015, and this number is projected to reach 2.1 billion by 2050 (1). In Canada alone, the number of individuals aged 65 and older is projected to reach 10.9 million people by 2036, which will correspond to approximately 25% of the population (2). Due to increases in life expectancy Canadians are living longer, however not necessarily healthier lives (3). Advancing age is associated with higher levels of chronic disease (3), as well as declines in cognitive and physical functioning (4). Thus, identifying strategies to maintain functional independence in this population is a public health priority (5).

The ability to execute skilled movements is critical for performing activities of daily living and maintaining independence (6). Whether it is tying a shoelace, driving a car or recovering arm function after a stroke, the acquisition or reacquisition of motor skills is pertinent across the lifespan. Aging is associated with a reduced ability to perform motor tasks, as well as a decreased capacity for motor skill learning (7,8). Although motor deficits in older adults may arise from dysfunctions in the neuromuscular system as well as the central and peripheral nervous systems, age-related changes in brain anatomy and physiology likely play a major role (7,9). The ability of the brain to adapt based on experience, termed neuroplasticity, is the biological mechanism underlying the process of motor learning (10,11). However, in aging individuals this capacity for neuroplastic change is reduced (9,12).

Distinct neural circuits within the motor cortex can be measured non-invasively in vivo using transcranial magnetic stimulation (TMS) (13). A meta-analysis examining the effects of aging on neurophysiology using TMS measures revealed a reduction in motor cortical
excitability and plasticity, compared to younger individuals (9). Moreover, a large-scale study with the aim of establishing normative data of TMS measures across the lifespan demonstrated an age-dependent reduction in inhibitory mechanisms within the motor cortex (14). Importantly, cumulative evidence suggests there is a link between the decreased ability to modulate excitatory and inhibitory networks within the motor cortex and age-related motor deficits (9,15).

In theory, specifically targeting the modulation of cortical excitatory and/or inhibitory networks closely in time with motor practice may facilitate motor learning by creating an optimal neural environment for plasticity to occur (16–18). The idea of using adjunct therapies with motor practice to strategically target neurobiological mechanisms and facilitate the motor learning process is not novel; techniques such as repetitive brain stimulation (19) and pharmaceutical drugs (20) have previously been shown to optimize learning when paired with motor practice. Recently, aerobic exercise has been proposed as an endogenous neuromodulation strategy to optimize motor learning and brain plasticity (21,22). Animal and human research over the past decade have demonstrated the effects of exercise on brain structure and function at systemic, cellular, and molecular levels (22). Thus, there is biological plausibility that aerobic exercise may serve as a rehabilitation strategy to optimize motor recovery and function. Indeed, multiple studies conducted in young healthy adults have demonstrated that a single session of aerobic exercise can facilitate motor performance and learning (for review, see 12), as well as modulate motor cortical circuitry (18,23–29). However, whether these effects extend to healthy older adults warrants investigation. While a recent systematic review highlighted the positive effects of chronic physical activity on motor performance and learning in healthy older adults, the authors “found no appropriate study examining the influence of an acute bout of exercise on motor performance or motor learning processes in older adults” (30). Further, to our knowledge,
no study has evaluated the influence of acute aerobic exercise on motor cortical circuitry in healthy older adults.

To increase the utility of aerobic exercise as a strategy to promote healthy aging, further understanding of how acute aerobic exercise impacts brain physiology and function across the lifespan is warranted. Thus, the present thesis contains two proof-of-concept studies designed to assess the efficacy of a single session of moderate-intensity aerobic exercise in 1) facilitating motor performance and learning, and 2) modulating motor cortical and intracortical networks, in healthy older adults.

1.2 Motor learning concepts

Motor learning is a form of procedural learning, and refers to a relatively permanent change in the capability to perform skilled movement as a result of practice or experience (6). In the literature, there is an important distinction between motor performance and motor learning (6,31). The term motor *performance* reflects a transient status of behavior, for example what is assessed during a practice session (31). Improvements during a motor practice session, also referred to as the skill acquisition period, are thought to represent online gains (11). In contrast, motor *learning* refers to a relatively permanent change in the capability for movement, as a result of practice or experience (6). Because it cannot be directly observed, we typically infer motor learning by measuring changes in performance (6). This can be assessed through the use of delayed retention tests, following the completion of practice (6). Improvements that occur after a period of time without additional practice are termed offline gains (11), and are thought to reflect the consolidation of motor memories (32).

In a laboratory setting, two main types of motor learning are commonly evaluated: motor sequence learning and motor adaptation learning. Motor sequence learning refers to the process
by which a specific series of individual movement elements come to be performed effortlessly through repeated practice (32,33). Examples of sequential actions in everyday activities include typing on a keyboard or tying one’s shoelace. In contrast, motor adaptation refers to the capacity to compensate for environmental changes, such as visual or kinematic perturbations (32,33). For instance, walking on an icy surface or reaching in a force-field are examples of adaptation tasks. Motor sequence and adaptation learning are thought to share similar, yet distinct underlying mechanisms. In the present thesis, there will be a specific emphasis on motor sequence learning.

1.2.1 Phases of motor learning

In general, motor learning is thought to occur in distinct phases. An initial, “fast learning” phase is characterized by rapid improvements in performance, often within a single practice session (11,32,34,35). It has been proposed that during this early phase encoding is initiated, where information related to the motor task is processed and a motor memory trace begins to form (31). Encoding is thought to rely on cognitive processes required for stimulus identification and response selection, and execution (31). At this stage, the memory trace is thought to be in a labile state and highly susceptible to interference (36,37). Subsequent to this, a “slow learning” phase occurs over longer time spans and leads to relatively more permanent changes in behavior (11,32,34,35). Through the process of consolidation, offline gains occur and the motor memory transitions into a more robust form, which is less susceptible to interference (36). Slow learning is thought to eventually lead to automatization, where performance of the motor skill requires minimal attentional and cognitive resources (11).

It has been proposed that during motor sequence learning, this transition from a more cognitively demanding phase towards the automatization of motor execution involves a memory strategy termed chunking (38,39). Chunking refers to the storage of individual
memory units that eventually through practice, become linked together to form larger movement sequences, or “chunks” (38). Storing separate clusters of movements into one unified memory trace as a motor chunk improves efficiency and results in faster execution of the motor skill (40). It is also thought that explicit learning strategies are engaged during the early phase of sequence learning, when conscious problem-solving processes are needed to learn the goals of the task (41). When individuals unknowingly improve motor performance on repeated sequences relative to random sequences, it is thought to reflect implicit sequence learning (33,41).

1.2.2 Age-related declines in motor performance and learning

Age-related motor performance deficits include difficulties in coordination, increased spatial and temporal variability of movement, slowing of movement, and compromised balance and gait (7). Upper extremity motor performance, such as grasping or reaching for an object, also declines with increasing age (30). For instance, older adults demonstrate deficits in coordination of bimanual and multi-joint movements, as well as unimanual tasks (42,43). These alterations in motor behavior can have a significant impact on activities of daily living, and the ability to maintain functional independence. Many factors contribute to motor performance impairments with aging, such as changes to peripheral structures (ie. sensory receptors, sarcopenia) as well as central nervous system changes (7). In the present thesis, there will be a specific focus on age-related changes in motor cortical circuitry.

Although behavioural studies evaluating the effects of aging on motor skill learning have varying results depending on the type of motor task, it is generally agreed that there are age-related differences in motor performance and learning capabilities (8). Evidence suggests older adults may have deficits in the ability to encode motor memories during skill acquisition, and
might be more susceptible to motor memory interference (44–46). Regarding motor sequence learning, skill acquisition processes appear to be comparable among young and older adults during the learning of simpler tasks, however older adults demonstrate more difficulties acquiring high-complex motor tasks (8,33,47). For instance, older adults exhibited impairments in learning the serial response time task, where repeated sequences alternate with random trials, compared to younger individuals (47). However, regarding the consolidation of motor sequences, age-related deficits have been consistently reported (33). Age-related deficits in the consolidation of motor sequences may be attributed to degradations in brain structure and/or functioning(33), which will be further discussed below.

1.3 Neuroplasticity underpins the process of motor learning

Motor learning is underpinned by the ability of the central nervous system to adapt structurally or functionally at systemic, cellular, and molecular levels (11,17). At the systems level, multiple brain networks are involved, and the interaction of these neural networks has been shown to depend on the type of motor task and stage of learning (32,48,49). The initial fast learning phase is associated with (but not limited to) widespread activation of the prefrontal and motor cortices, basal ganglia, cerebellum, and hippocampus (33). Moreover, during fast learning for both sequence and adaptation tasks, the corticostriatal and corticocerebellar networks are thought to operate in parallel (32,48,49); whereas during consolidation and slow learning, sequence and adaptation tasks are thought to rely more on corticostriatal and corticocerebellar systems, respectively (32,33,48). Regardless of the type of motor task, large-scale shifts in brain activity occur from associative/premotor areas, regions known for higher order cognitive functioning, to sensorimotor networks; this supports the hypothesis that motor learning involves a transition from a more cognitively demanding phase to automaticity over time (50).
At the cellular level, motor learning involves the strengthening of neural connections (or formation of new ones) resulting from repetitious activation of synaptic networks, a process known as long-term potentiation (LTP) (51,52). Repeated stimulation of synaptic connections can occur through behavioural motor task practice, and LTP-like plasticity is strongly implicated in the consolidation of motor memories (24,53). It is thought that by recruiting the same functional circuitry repetitively during motor skill practice, the efficiency of that circuitry increases, making it easier to reproduce the motor skill. Within the motor cortex, excitatory and inhibitory interneurons likely facilitate the induction of LTP by directly modulating corticospinal output (54–56). Specifically, increases in motor cortical excitability and decreases in intracortical inhibition have been associated with LTP-like processes (57,58).

In humans, changes in excitatory and inhibitory networks within the motor cortex can be measured using non-invasive brain stimulation protocols, such as TMS (54). TMS sends a transient magnetic field over the motor cortical representation of a muscle, and through the principles of electromagnetic induction, activates interneurons within the primary motor cortex, which sends the signal down pyramidal corticospinal output neurons, synapses with motor neurons, and produces motor output (19). The motor output, termed motor evoked potential (MEP), is measured using electromyography (EMG) and reflects levels of corticospinal excitability (19). MEPs are typically quantified by the peak-to-peak MEP amplitude (19). By applying single-pulse TMS over a wide range of intensities, it is possible to measure the increase in excitability (ie. increase in peak-to-peak MEP amplitudes) within the corticospinal tract in response to increased stimulus intensity (19). This TMS protocol is also known as a recruitment curve and is thought to recruit neurons with different excitability thresholds, thus allowing for a
more extensive examination of corticospinal excitability compared to observing MEP amplitudes at a single intensity (19,59).

Inhibitory and facilitatory interneurons within the motor cortex influence the activation of pyramidal corticospinal output neurons, and are typically measured using paired-pulse TMS protocols (19). To measure interneuronal circuitry, two TMS pulses are delivered in close succession at various intensities and interstimulus intervals (ISI) (19). For instance, short-interval intracortical inhibition (SICI) can be measured by delivering a conditioning pulse at subthreshold intensity followed by a suprathreshold test stimulus separated by an ISI between 1-6 ms (19). The result is an MEP with decreased amplitude compared to that elicited from a single test stimulus alone, and reflects inhibitory interneuronal activity within the motor cortex that is mediated by gamma-Aminobutryic acid (GABA)-A receptors (54). To assess facilitatory interneuronal circuits within the motor cortex, intracortical facilitation (ICF) can be delivered using the same protocol except with an ISI of 10-15 ms (19). This results in an MEP with increased amplitude compared to that from a single test pulse alone, and likely reflects circuitry mediated by glutamate, with possible contributions from N-methyl-D-aspartate (NMDA) and GABA (54,60). Another similar, yet distinct measure of intracortical inhibition is long-interval intracortical inhibition (LICI). LICI is produced from two identical suprathreshold pulses separated by an ISI of 50-200ms, and is likely mediated by GABA-B receptors (19,23). Using single- and paired-pulse TMS protocols can help elucidate changes in distinct neural circuits within the motor cortex, and how these relate to functional improvements. A schematic depiction of TMS can be found in Figure 1.1. An overview of single- and paired-pulse TMS protocols can be found in Table 1.1.
Figure 1.1 Schematic of transcranial magnetic stimulation (TMS). A) The orientation of the TMS coil is typically 45° to the mid-sagittal plane over the motor cortex, with the handle pointing laterally and posteriorly. B) The principles of electromagnetic induction generate electrical current in the underlying brain tissue to activate intracortical interneurons which connect to descending corticospinal tracts, synapse in spinal gray matter and produce motor output from the target muscles. The motor output, or motor evoked potential (MEP), is a measure of cortical excitability and can be recorded using surface EMG electrodes. C) Representative traces of motor output: single-pulse MEP, SICI (decreased MEP amplitude following a subthreshold + suprathreshold paired-pulse), ICF (increased MEP amplitude following a subthreshold + suprathreshold paired-pulse), LICI (decreased MEP amplitude following two paired suprathreshold pulses). Figure adapted with permission from © Brown et al. 2014. Degener. Neurol. Neuromus. Dis. 4:133-151.
Table 1.1 Neurophysiological measures using TMS.

<table>
<thead>
<tr>
<th>Measure</th>
<th>Motor Evoked Potential</th>
<th>Short-interval intracortical inhibition</th>
<th>Intracortical facilitation</th>
<th>Long-interval intracortical inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proposed mechanism</td>
<td>Activation of pyramidal corticospinal neurons</td>
<td>GABA-A receptor mediated inhibitory interneurons</td>
<td>Glutamate mediated facilitatory interneurons</td>
<td>GABA-B receptor mediated inhibitory interneurons</td>
</tr>
<tr>
<td>Protocol</td>
<td>Single-pulse TMS</td>
<td>Subthreshold conditioning pulse followed by suprathreshold test pulse, with 1-6 ms ISI</td>
<td>Subthreshold conditioning pulse followed by suprathreshold test pulse, with 10-15 ms ISI</td>
<td>Two suprathreshold pulses separated by 50-200 ms ISI</td>
</tr>
</tbody>
</table>

1.3.1 Potential neural mechanisms underlying age-related motor deficits

Normal aging is characterized by structural and functional changes to the brain, which at a behavioral level, can present as declines in motor functioning. Age-related structural changes such as gray and white matter atrophy in prefrontal, motor cortical, and sensorimotor regions have been linked with declines in motor performance (7). In addition to these structural changes, age-related differences in brain neurochemistry, such as reduced norepinephrine, dopamine, and GABAergic transmission levels have also been linked to motor deficits (7,15). Other functional changes associated with aging include increased reliance on prefrontal regions (33), and compensatory recruitment patterns such as increased bilateral recruitment of the motor cortex for unimanual tasks, compared to younger individuals (61). As both the frontal cortex and striatum are heavily implicated in motor sequence learning, age-related degradations to cortico-striatal networks are thought to particularly contribute to deficits in motor sequence learning (33).

Along with these large-scale shifts in brain activity that likely contribute to age-related motor deficits, specific neural circuits within the motor cortex also appear to change with age. Aging is associated with significant reductions in corticospinal excitability (9,14), intracortical...
inhibition (14), and LTP-like cortical plasticity (9,62). Corticospinal excitability and intracortical inhibition are implicated in the induction and maintenance of neuroplasticity (9,58), and the consolidation of motor memories (24,63). Reductions in cortical excitability and inhibition are thought to contribute to the loss of modulatory capacity (14,64). Further, the inability to modulate cortical excitability and inhibition is associated with age-related motor impairments (9,15). Specifically, deficits in the capacity to modulate GABA-mediated inhibition have been linked with slower reaction times (64), decreased coordination (65), and overall declines in motor performance with age (7,15). There has been interest in exploring the potential of aerobic exercise to ameliorate age-related declines in inhibitory control (66), however, the efficacy of acute aerobic exercise in modulating cortical and intracortical circuitry in healthy older adults has yet to be demonstrated.

1.4 Exercise as a potential strategy to facilitate motor learning

The benefits of exercise on cognitive and memory processes across the lifespan are becoming widely known (67–72). Although the literature on exercise as a potential strategy to facilitate motor behavior is still in its infancy, numerous studies have been conducted to date. A recent systematic review demonstrated that in healthy older adults, chronic physical activity shows positive effects on measures of motor performance and the early phase of motor learning, with mixed results on consolidation (30). However, the effects of acute exercise on motor behavior in healthy older adults remain to be elucidated (30). To inform the development of future rehabilitation strategies using exercise to optimize motor memory consolidation, understanding the effects of acute exercise on motor behavior in older adults is a critical first step.
In young healthy adults, numerous studies evaluating the effects of acute aerobic exercise on motor skill performance and learning yield promising results, despite differences in study designs, exercise protocols and types of motor tasks that were utilized. Depending on the timing of the bout of exercise, it is thought that distinct phases of motor skill learning can be differentially impacted (22, 71, 73, 74). For instance, exercising immediately before task practice is thought to preferentially influence encoding processes and the skill acquisition phase (73), likely by “priming” the brain for neuroplasticity (21), or by simply increasing levels of arousal (22). However, because the effects of exercise persist after it has been performed, it is also possible to impact consolidation processes when the exercise bout is performed prior to task practice (73). In contrast, performing an exercise bout following task practice can solely impact the consolidation phase, likely by facilitating offline gains in learning, and the transition of the motor memory into a more stable form (73).

One of the first studies to evaluate the impact of acute exercise on skill acquisition and consolidation was conducted by Roig et al. in 2012 (75). The authors examined the effects of 20 minutes of high-intensity interval cycling performed either before or after practicing a visuomotor accuracy tracking task, compared to a resting control group (75). Although there were no exercise effects on skill acquisition, performing acute aerobic exercise facilitated performance improvements at 24-hour and 7-day retention tests regardless of whether it was performed before or after motor practice. Further, the group that exercised after task practice demonstrated the greatest offline gains in performance. Mang et al. (2014) expanded on these findings, noting high-intensity exercise immediately prior to motor task practice decreased time lag during skill acquisition, and these improvements were maintained at a 24-hour retention test (27). Because acute exercise specifically improved the temporal component of a
Continuous tracking task thought to rely on cerebellar functioning (27), the authors were interested in whether acute exercise could also enhance learning of a discrete sequence task. Continuous and discrete tasks are thought to rely on different memory processes, with cerebellar functioning playing a more significant role in continuous learning, and discrete sequence learning likely relying on greater contributions from basal ganglia circuits (6,40,76).

In this study, a single session of high-intensity exercise performed prior to task practice facilitated the consolidation of a discrete motor sequence task in young healthy adults (77). To further understand how the temporal proximity of exercise and motor task practice mediates exercise-induced changes in consolidation, Thomas and colleagues tested the effects of performing high-intensity exercise 20 minutes, 1 hour, or 2 hours following task practice (74). In this study, improvements in accuracy were greatest both 24 hours and 7 days later in the group that exercised 20 minutes following practice, and the effects decreased as the time between exercise and motor practice increased (74). Taken together, these studies provide evidence that acute high-intensity exercise performed closely in time with task practice can enhance skill acquisition and motor memory consolidation, across different types of motor tasks.

Although the previously mentioned studies utilized exercise bouts of high-intensity, the efficacy of moderate-intensity exercise in facilitating motor learning and performance has also been demonstrated. Notably, moderate-intensity exercise performed prior to task practice has been shown to improve (78) and maintain (79) spatial accuracy, and increase acceleration (80) during skill acquisition. To better understand the role of exercise intensity in mediating changes in motor behaviour, Thomas and colleagues (2016) directly compared the effects of moderate- to high-intensity exercise performed after task practice (81). In this study, both
exercise intensities increased motor memory consolidation compared to rest, however changes were greater after high-intensity (81). Cumulatively, these studies demonstrate the efficacy of moderate-intensity, as well as high-intensity, exercise performed closely in time with task practice, in facilitating skill acquisition and consolidation. Moderate-intensity exercise may be more feasible in certain populations; thus, understanding how moderate-intensity exercise impacts motor behavior in healthy older adults is an important step towards the potential use of exercise as a neuromodulation strategy in a clinical setting. A summary of the studies on acute aerobic exercise and motor behavior in young healthy adults can be found in Table 1.2.
Table 1.2 Overview of studies on acute aerobic exercise and motor behavior in young healthy adults

<table>
<thead>
<tr>
<th>Author, Year</th>
<th>Research Question</th>
<th>Participants</th>
<th>Exercise Prescription</th>
<th>Motor Task</th>
<th>Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Roig et al., 2012</td>
<td>Does high-intensity exercise immediately before or after motor task practice improve skill acquisition and retention, compared to a resting condition?</td>
<td>Between-subjects. Ex (pre) vs. Ex (post) vs. Control (seated rest): n=16/group, f=0, avg=24 yrs</td>
<td>Intensity: High (~200-315W, based on individual VO\textsubscript{2peak}); Type: Cycling Time: 3x3 min high intensity interspersed with 3x2 min low-intensity (50W); immediately prior / post motor practice</td>
<td>Task: Visuomotor accuracy-tracking (wrist flexion and extension) Acquisition: 3x5 min blocks Retention: 24h and 7d (3x5 min blocks)</td>
<td>Ex (pre) and Ex (post) had significantly better retention of motor skill (↓ root mean square error of distance) both 24h &amp; 7d after practice compared to rest. Ex (post) group improved the most at 7d retention.</td>
</tr>
<tr>
<td>Mang et al., 2014</td>
<td>Does high-intensity exercise immediately prior to motor task practice improve acquisition and retention of a continuous tracking task?</td>
<td>Within-subjects. Ex vs. seated rest: n=16, f=8, avg=24 yrs</td>
<td>Intensity: High (90% PO\textsubscript{max}); Type: Cycling; Time: 3x3 min high-intensity / 3x2 minute low-intensity (50W); immediately prior to motor practice</td>
<td>Task: Continuous tracking track (thumb tracking with joystick) Acquisition: 2 blocks of 10 trials Retention: 24h (1 block of 10 trials)</td>
<td>Ex improved temporal aspects of implicit sequence-specific learning (↓ time lag) during acquisition, which was maintained at 24h retention compared to rest. Ex did not impact the acquisition/retention of spatial aspects.</td>
</tr>
<tr>
<td>Statton et al., 2015</td>
<td>Does the timing of moderate-intensity exercise (immediately vs. 1 hour) prior to motor task practice influence motor skill acquisition?</td>
<td>Between-subjects. Ex immediately pre vs. Ex 1h pre vs. Control (slow walk immediately pre): n=8/group, f=16, avg=22 yrs</td>
<td>Intensity: Moderate (65-85% age-predicted HR\textsubscript{max}); Type: Running Time: 30 min; immediately vs. 1h prior to task practice. Control: slow walking (1m/s) for 35 min at low-intensity</td>
<td>Task: Sequential Visual Isometric Pinch Task (force matching with fingers) Acquisition: 4 blocks of 30 trials Retention: N/A</td>
<td>Running immediately before task practice improved skill acquisition (↑ accuracy) compared to low-intensity walking before practice. No improvements were observed when running was followed by 1h rest prior to practice.</td>
</tr>
<tr>
<td>Mang et al., 2016</td>
<td>Does high-intensity exercise immediately prior to practicing a discrete motor sequence task improve motor skill acquisition and consolidation?</td>
<td>Within-subjects. Ex vs. seated rest: n=16, f=10, avg=26 yrs</td>
<td>Intensity: High (90% PO\textsubscript{max}) Type: Cycling Time: 3x3 min high-intensity interspersed with 3x2 minute low-intensity (50W); immediately prior to motor practice</td>
<td>Task: Serial targeting task (hand-arm movements, manipulating a computer mouse) Acquisition: 3 blocks of 110 trials Retention: 24h (1 block of 110 trials)</td>
<td>Ex ↑ rate of implicit sequence-specific motor memory consolidation at 24h retention compared to rest. Ex did not impact rate of skill acquisition or overall change in performance during acquisition or consolidation.</td>
</tr>
<tr>
<td>Author, Year</td>
<td>Research Question</td>
<td>Participants</td>
<td>Exercise Prescription</td>
<td>Motor Task</td>
<td>Findings</td>
</tr>
<tr>
<td>--------------</td>
<td>-------------------</td>
<td>--------------</td>
<td>-----------------------</td>
<td>------------</td>
<td>----------</td>
</tr>
<tr>
<td>Thomas et al., 2016a</td>
<td>What is the role of exercise intensity in mediating the effects of acute aerobic exercise on motor skill learning?</td>
<td>Between-subjects. Moderate-intensity vs. High-intensity vs. Control (seated rest): n=12/group, f=0, avg=24 yrs</td>
<td>Intensity: Moderate (45% $\text{PO}<em>{\text{max}}$) vs. High (90% $\text{PO}</em>{\text{max}}$); Type: Cycling; Time: 3x3 min of work (45% or 90% $\text{PO}<em>{\text{max}}$) / 3x2 mi of active rest (25% or 60% $\text{PO}</em>{\text{max}}$); immediately after practice</td>
<td>Task: Visuomotor accuracy-tracking (wrist flexion and extension) Acquisition: 5 blocks of 20 trials Retention: 24h and 7d (1 block of 20 trials)</td>
<td>Acquisition was similar among groups. High-intensity ex ↑ retention (24h &amp; 7d) compared to moderate-intensity and rest. Moderate-intensity ↑ consolidation (at 7d) compared to control, but to a lesser extent than high-intensity.</td>
</tr>
<tr>
<td>Thomas et al., 2016b</td>
<td>What is the role of exercise timing in mediating the effects of acute aerobic exercise on motor skill learning?</td>
<td>Between-subjects. Ex 20 min vs 1h vs 2h post vs Control (seated rest): n=12/group, f=0, avg=24 yrs</td>
<td>Intensity: High (90% $\text{PO}<em>{\text{max}}$); Type: Cycling; Time: 3x3 min of work (90% $\text{PO}</em>{\text{max}}$) interspersed with 3x2 min of active rest (60% $\text{PO}_{\text{max}}$); 20 min, 1h, or 2h post motor practice</td>
<td>Task: Visuomotor accuracy-tracking (wrist flexion and extension) Acquisition: 5 blocks of 20 trials Retention: 24h and 7d (1 block of 20 trials)</td>
<td>Acquisition was similar among groups. At 24h, Ex20m ↑ accuracy than rest and Ex+2h. At 7d, all exercise groups ↑ accuracy; improvements were greatest for Ex20m, and decreased as time between exercise and motor practice decreased.</td>
</tr>
<tr>
<td>Perini et al., 2016</td>
<td>Does a single session of moderate-intensity exercise influence learning within visual and motor domains?</td>
<td>Between-subjects. Ex (n=18) vs Control (low-intensity cycling at 20W) (n=19); f=0, avg=22 yrs</td>
<td>Intensity: Moderate (70% $\text{HR}_{\text{max}}$); Type: Cycling; Time: 30 minutes; immediately prior to motor practice</td>
<td>Task: Thumb abduction motor task Acquisition: 6 blocks of 65 movements Retention: N/A</td>
<td>Ex ↑ acceleration in all blocks during task practice compared to rest</td>
</tr>
<tr>
<td>Snow et al., 2016</td>
<td>Does moderate-intensity exercise prior to motor task practice improve acquisition and retention of a continuous tracking task?</td>
<td>Within-subjects. Ex vs seated rest; n=16, f=7, avg=26 yrs</td>
<td>Intensity: Moderate (PO corresponding to 60% $\text{VO}_{2\text{peak}}$); Type: Cycling; Time: 30 minutes, immediately prior to practice</td>
<td>Task: Continuous tracking track (thumb tracking with joystick) Acquisition: 2 blocks of 10 trials Retention: 24h (1 block of 10 trials)</td>
<td>Ex maintained spatial accuracy (root mean square error) whereas spatial accuracy ↓ after rest; Ex facilitated the maintenance of motor skill performance.</td>
</tr>
</tbody>
</table>
1.4.1 Possible mechanisms through which exercise facilitates motor learning

Physical activity and exercise are thought to promote neuroplasticity at systemic, cellular, and molecular levels across the lifespan (22,82–84). Although the benefits of exercise on the brain and cognition are widely known (69,70,85,86), exercise-induced brain changes are also thought to positively influence motor learning (for review, see (22)). In this section, we will outline potential mechanisms through which exercise is thought to promote neuroplasticity, in specific relation to motor learning.

1.4.1.1 Acute aerobic exercise modulates movement-related neural circuits

Literature has begun to establish the effects of acute aerobic exercise on neurophysiological outcomes as measured using TMS. Single bouts of lower-limb moderate- and high-intensity aerobic exercise have been shown to increase the neurophysiological response in upper limb muscles to paired associative stimulation, a TMS paradigm shown to evoke LTP-like mechanisms (27,87). Similarly, acute aerobic exercise has been shown to promote the neuroplastic response to a repetitive brain stimulation protocol used to suppress cortical excitability (28). Together, these findings demonstrate acute exercise can facilitate neuroplastic mechanisms thought to reflect synaptic efficacy (88).

Although the body of literature is still relatively small, multiple studies in young adults demonstrate acute aerobic exercise modulates specific excitatory and inhibitory networks within the motor cortex. In young healthy adults, an acute bout of lower-limb moderate-intensity aerobic exercise modulates intracortical inhibition in upper limb muscles (18,25,26). Cumulatively, it appears that reduced SICI is the most consistently reported after an acute bout of exercise (18,24,26,29), suggesting that GABA-A receptor mediated circuits may be the most sensitive to exercise-induced neuroplastic change. This effect has been
shown across participants with different levels of physical activity (26), and correlates with exercise-induced improvements in motor memory consolidation (24). Together, these findings support the hypothesis that a release of GABA-A mediated inhibition is important for LTP-like plasticity and motor skill learning (24,34,57).

It is interesting to note that Stavrinos and Coxon (24) demonstrated a decrease in SICI with an ISI of 2ms after exercise, whereas SICI with an ISI of 1ms did not change. Varying the ISI between paired pulses is thought to recruit different neuronal populations (23), suggesting that exercise may differentially influence distinct circuitry within the motor cortex. This might also explain the inconsistencies regarding acute exercise effects on ICF. The literature is mixed, with studies showing an increase (18) and decrease (26) after acute moderate-intensity exercise; however, Singh et al. (2014) used an ISI of 12ms (18), and Lulic et al. (2017) used an ISI of 10ms (26). The notion that acute exercise might differentially modulate distinct motor cortical circuitry is further supported by the observation that GABA-B receptor mediated LICI does not seem to be modulated after exercise as consistently as SICI. For instance, Mooney and colleagues (25) noted reduced LICI at 10 and 20 minutes post-exercise, Stavrinos and Coxon (24) found no change in LICI after exercise, and Singh and colleagues (18) found no change in LICI immediately after exercise and a non-significant trend towards reduced LICI after 30 minutes. In contrast, a reduction in SICI is often reported after exercise (18,24–26,29). Although both are measures of GABAergic intracortical inhibition, SICI and LICI reflect distinct neuronal pools and underlying mechanisms (89).

Although the majority of studies did not report a statistically significant effect of lower-limb exercise on upper limb corticospinal excitability (18,25,27,29), Lulic et al. (2017) noted exercise-induced changes in corticospinal excitability are influenced by physical
activity levels (26). Specifically, corticospinal excitability was increased in the high physical activity group, whereas it did not change in the low physical activity group (26). In this study, the authors speculated that individuals who have higher levels of physical activity may have increased efficiency in utilizing exercise-induced neurochemicals that are implicated in cortical excitability and LTP-like processes (26). Further work is warranted to better understand how exercise-induced changes are mediated by changes in physical activity and fitness levels.

Overall, these findings indicate that acute exercise modulates corticospinal excitability and intracortical networks in young healthy adults, with numerous studies specifically reporting decreases in GABAergic inhibition (18,24–26,29). Further, this may have implications for LTP-like plasticity and motor skill learning (24,57). However, whether these acute exercise effects extend to healthy older adults remains to be elucidated.

1.4.1.2 Exercise increases peripheral concentrations of neurochemicals

Although investigations on a molecular level are outside of the scope of the present thesis, it is important to acknowledge that increased concentrations of neurochemicals as a result of exercise likely underlie exercise-related changes in LTP and motor learning (21,22,71,75). It has been proposed that exercising closely in time with motor practice will increase the availability of these neurochemicals for their uptake and utilization in LTP-like processes during encoding and consolidation processes, and therefore facilitate motor learning (21,73). These neurochemicals include neurotrophic growth factors such as brain-derived neurotropic factor (BDNF), lactate, and neurotransmitters such as dopamine, epinephrine, norepinephrine, and GABA (22,90,91). Exercise-induced increases in these neurochemicals are correlated with improvements in declarative learning and motor memory (90,92).
1.5 Specific aims and hypotheses

The primary motivation of this thesis was to determine whether acute aerobic exercise facilitates motor behavior and movement-related neural circuits in a sample of healthy older adults. The proof-of-concept studies contained in this thesis were designed to build upon findings in young healthy adults, with the intention of informing future research in healthy aging and clinical populations. There were two main aims:

Aim 1: To examine the effects of 20 minutes of moderate-intensity cycling performed immediately prior to motor practice of the serial targeting task (STT) on motor skill performance and learning in healthy older adults, compared to a resting condition. We hypothesized that a single session of moderate-intensity aerobic exercise performed immediately prior to motor practice of the serial targeting task would significantly improve both motor performance and learning of the STT. Study 1 was designed to evaluate this aim.

Aim 2: To investigate whether 20 minutes of moderate-intensity cycling exercise modulates corticospinal excitability, intracortical facilitation, and intracortical inhibition in healthy older adults. We hypothesized that a single session of moderate-intensity aerobic exercise would significantly modulate corticospinal excitability, intracortical inhibition, and intracortical facilitation. Study 2 was designed to evaluate this aim.

1.6 Rationale

The ability to exploit the neuroplastic benefits of exercise to optimize motor memory consolidation has large implications for rehabilitation. Based on literature in young healthy adults, aerobic exercise has potential to serve as an alternative neuromodulation strategy. However, there is little evidence demonstrating the effects of exercise on motor behavior and
neurophysiological circuits across the lifespan. To inform future work in aging and clinical populations, it is necessary to understand the efficacy of acute exercise in modulating motor learning and movement-related neural circuits in healthy older adults. Moderate-intensity exercise may be more feasible for certain populations, such as sedentary individuals or individuals with chronic disease, thus understanding the priming effects of moderate-intensity exercise is a critical first step. As the average age of the world’s population continues to rise, investigation into possible adjuvant therapies to target neurobiological mechanisms and facilitate motor memory consolidation is warranted.
Chapter 2: Methods

2.1 Participants

Twenty-two individuals between the ages of 55-75 years old were recruited from UBC and the surrounding community using a sample of convenience. We included right-handed volunteers as determined using the Edinburgh Handedness Inventory (93) (Appendix A). Participants were excluded if they showed any signs of cognitive impairment (score <24 on the Montreal Cognitive Assessment) (Appendix B), or had a history of head trauma, any neurological or psychiatric diagnoses, neurodegenerative disorder, or substance abuse. Participants were screened for contraindications to TMS and exercise procedures using a TMS screening form (Appendix C) and an exercise-based stress test (Appendix D), respectively. All participants independently provided written and verbal informed consent, in accordance with the UBC’s Clinical Research Ethics Board and the Declaration of Helsinki.

2.2 Study design

Each participant first completed an initial, pre-experimental session at least two days prior to all other sessions to complete a maximal exercise stress test. Next, participants completed five sessions in a within-subjects experimental design to assess the effects of a 20-minute period of seated rest and a 20-minute bout of moderate-intensity lower limb aerobic exercise on: 1) motor skill learning of an upper limb task (Study 1), and 2) corticospinal excitability and intracortical circuitry of an upper limb muscle representation within the primary motor cortex (Study 2). On all testing days, participants were asked to refrain from any exercise besides that involved in the experimental sessions. Experimental design is depicted below in Table 2.1.
<table>
<thead>
<tr>
<th>Day</th>
<th>Test/Training</th>
<th>Dependent measures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-experimental session</td>
<td>1</td>
<td>Screening and cardiac stress testing</td>
</tr>
<tr>
<td>Study 1: Motor Learning</td>
<td>2</td>
<td>Exercise or Rest before motor task practice</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>No-exercise 24-hour retention test</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>Exercise or Rest before motor task Practice</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>No-exercise 24-hour retention test</td>
</tr>
<tr>
<td>Study 2: Neurophysiology</td>
<td>6</td>
<td>Neurophysiological measures before and after rest and exercise</td>
</tr>
</tbody>
</table>

Table 2.1 Schedule for experimental studies. VO₂peak: Peak oxygen consumption, STT: Serial tracking task.

2.3 Exercise procedures

2.3.1 Maximal stress testing

All participants completed a maximal stress test on a recumbent bicycle (Scifit ISO 7000R, Tulsa, OK, USA) in the UBC Hospital Cardiovascular Centre of Excellence during the initial pre-experimental session. The purpose of completing a maximal exercise stress test with electrocardiogram (ECG) monitoring was two-fold: 1) to screen for adverse cardiac responses during exercise and 2) to inform subsequent prescription of a standardized bout of moderate-intensity cycling (aerobic exercise; 50% of maximal power output (PO) in watts). Upon arrival for testing, the participant was asked to lay down comfortably in a hospital bed for 5 minutes while a member of the study team explained the stress test protocol. After 5 minutes of lying down, resting heart rate (HR) and blood pressure were recorded. Each participant was required to have a resting HR < 100 beats per minute (bpm), systolic blood pressure < 160 mm Hg, and diastolic blood pressure < 100 mm Hg before proceeding to
exercise testing. Participants were asked to inform the study team member if s/he was using beta-blocker medications.

Prior to the maximal stress test, a registered cardiology technician attached 12-lead ECG electrodes to the participant’s torso to monitor electrical activity of the heart throughout the test. After a 2-minute warm-up at a baseline workload of 10W, the workload increased every minute by 10W increments. If needed, these increments were adjusted by ± 5W on an individual basis, based on current exercise testing recommendations of 8-12 minutes in duration (94). Throughout the exercise test, participants were asked to maintain a pedaling cadence of 50 rotations per minute (rpm). Additionally, air flow and expired O₂ and CO₂ concentrations were continuously monitored via metabolic cart (ParvoMedics TrueOne 2400, Sandy, UT, USA) and HR was continuously monitored via ECG. Rating of perceived exertion (RPE) using the Borg’s 1-10 scale (Appendix E) was recorded after every minute. Additionally, the cardiology technician used a manual blood pressure cuff to record blood pressure every second minute. The stress test was terminated at volitional exhaustion, inability to maintain 50 rpm cadence, or if the participant requested to stop. Upon termination of the test, participants were asked to perform a cool down cycle at 10W for 2-5 minutes to gradually decrease HR. Participants were then asked to sit relaxed in a chair for a period of 6 minutes, after which resting HR and blood pressure were recorded. Participation in subsequent experimental sessions required the receipt of medical clearance from a cardiologist.

\[ \text{VO}_{2peak} \] was determined post hoc and confirmed by at least one of the following conditions: peak HR > age-predicted HR_{max}, RPE > 7, or peak respiratory exchange ratio (RER) ≥ 1.10 (95). From the stress tests, peak values of VO₂, PO, HR, and RER were
extracted (Appendix F). To ensure that participants were prescribed aerobic (as opposed to anaerobic) exercise, ventilatory thresholds were determined post hoc by the V-slope method, and further confirmed by the ventilatory equivalent method and RER ≥ 1.10 (Appendix G) (95,96).

2.3.2 Standardized exercise bout

The acute bout of moderate-intensity aerobic exercise performed under the exercise condition consisted of a 2-minute warm-up at 10W and self-selected cadence, 20 minutes of continuous cycling at a workload corresponding to 50% maximal power output (W) from the final stage of the maximal exercise test, and a 2-minute cool-down at 10W and self-selected cadence. This exercise prescription was based on previous work indicating that acute moderate-intensity lower-limb exercise can facilitate the acquisition (78,79) and consolidation (81) of an upper-limb motor task, and modulate upper-limb primary motor cortical and intracortical circuitry (18,26,97). Additionally, moderate-intensity exercise may be more feasible in rehabilitation settings for aging or clinical populations (74). To ensure that the exercise bout was perceived as moderately intense to participants, PO was adjusted online to try and maintain a RPE value ≤ 5. Throughout the exercise session, HR and RPE were measured and recorded every 5 minutes, and at the cessation of warm-up and cool-down periods. Participants performed the aerobic exercise bout on two occasions: once immediately before practicing a motor task (Study 1) and once before and after neurophysiological assessments (Study 2).

2.4 Study 1: Effects of acute aerobic exercise on motor performance and learning

To examine the effects of a single bout of moderate-intensity aerobic exercise on the acquisition and consolidation of a discrete motor sequence task, participants practiced the
serial targeting task (STT) immediately following two conditions (exercise and rest) in a crossover design with repeated measures. Following the stress test, participants were randomly allocated to complete either the exercise or rest condition first, prior to cross over. Motor task practice under the different experimental conditions was followed by a no-exercise retention test 24 ± 2 hours later. To prevent any order effect on subsequent practice, motor task practice under the two conditions were separated by at least a two-week washout period. Additionally, the order of participation under each condition was counter-balanced across the study sample. During the rest condition, participants were asked to watch a video clip for 20 minutes. The experimental procedures are depicted in Figure 2.1.

---

**Figure 2.2 A) Overview of study design for Study 1.** Participants completed screening questionnaires, provided informed consent, and underwent a maximal exercise stress test before being pseudo-randomized to complete the Rest or Exercise condition first. STT practice sessions were followed by 24 ±2 hour retention tests. Participation in the different conditions was separated by a minimum washout period of 2 weeks. **B) Overview of practice session and retention days.** 5 consecutive blocks of 111 trials of the STT was practiced immediately after either moderate-intensity aerobic exercise or seated rest. Each 24-hour retention test consisted of 1 block of 111 trials.
2.4.1 Serial targeting task procedures

The STT involved the manipulation of the KINARM End-Point Robot™ (BKIN Technologies Ltd, Ontario, Canada). Participants were asked to grasp the handle of the robotic arm with their dominant hand, which was represented by a cursor (a white dot) on the visual display. The objective of the task was to move the cursor between a series of targets (red dots) as quickly and accurately as possible. The targets all consisted of the same size and could appear in one of nine locations: one central target surrounded by eight additional targets arranged in an equidistant circular array (77). To initiate the appearance of the next target, the participant had to place the cursor in the current target for 500 ms. The next target appeared in a different location after an inter-target interval of 500 ms. The target movement response time was defined as the time in seconds from target appearance to the presentation of the next target (corrected for the 500 ms stationary period and the 500 ms inter-target interval) and was extracted using a custom MATLAB script (The Mathworks, Inc., Natick, MA, USA). Cursor position and all stimuli was presented at 200 Hz.

To differentiate between implicit sequence-specific learning and improvements in motor control, the presentation of a repeated 6-target sequence was alternated with 7-target random sequences. The same repeated 6-target sequence was practiced during all blocks. Each random sequence consisted of different target configurations for each trial within a block. The difficulty of random and repeated sequences were controlled for and equated using Fitt’s Law (77,98). Target movements were reversed between conditions (rest and exercise) to ensure equivalent difficulty yet different target movements (77). The order of presentation of conditions (rest and aerobic exercise) and sequences (regular and reversed) was randomly
assigned to each participant and counterbalanced across the sample. Participants were not informed of the embedded repeated sequence. This method allows differentiation between implicit sequence-specific learning (i.e. repeated sequences) and improvements in motor control (i.e. random sequences) (99,100).

To assess whether participants detected the repeated sequence, they completed an explicit recognition test following the retention test on the last experimental session (Day 6) (77). The explicit recognition test consisted of 10 target sequences and participants were asked to verbally answer ‘yes’ or ‘no’ after the presentation of each sequence to indicate whether it was explicitly recognized. Seven of the presented sequences were random and three were the repeated sequences from both (exercise and rest) conditions. We expected that if explicit learning of the sequence occurred, the average number of correct responses would be higher than that associated with chance (ie. 2/3 repeated sequences and 4/7 random sequences) (77,101,102). A depiction of the STT task performed on the KINARM robot can be seen in Figure 2.2.

**Figure 2.2** Experimental set-up of the KINARM End-Point Robot™. A) Participants were seated comfortably in a chair in front of a visual display, with hands grasping the handle of the robotic arm. B) Animated image of a participant reaching to a target during the STT. C) Schematic of a participant’s movement trajectory throughout the task (image adapted from Mang et al. (2016) (77).
Each practice day began with either seated rest or a single session of aerobic exercise, then STT practice (5 consecutive blocks total). Each block was comprised of 8x6-target repeated sequences and 9x7-target random sequence trials, for a total of 111 movements. At each no-exercise retention test, participants completed 1 block of 111 movements (8x6-target repeated sequences and 9x7-target random sequence trials). On retention test days, participants were asked to complete a visual analogue scale of sleep quality (Appendix H) as well as the Godin Leisure-Time Exercise Questionnaire (Appendix I). (103) to obtain measures of general sleep quality and physical activity levels, respectively.

2.4.2 STT Analysis

All STT data were processed using a custom MATLAB script (Version R2016b, The Mathworks, Inc., Natick, MA, USA). Motor performance was evaluated using target response time, the sum of the reaction and movement times to reach to each individual target, and was evaluated separately for repeated and random sequences. To evaluate changes in skill acquisition during practice, a change score was calculated from the mean target response time of the last practice block and the first practice block. To evaluate motor memory consolidation, a change score was calculated from the mean target response time of the single block of the delayed retention test and the last practice block.

2.4.3 Statistical Analyses

Statistical tests were performed using SPSS statistics 22.0 (IBM Corporation, USA) and Statistica (Version 13, TIBCO Software Inc., USA) using a significance threshold of α = .05. Data were checked for normality, with the Shapiro-Wilks significance level set at p < 0.001 (104) and visual inspection of histogram plots. Data were log-transformed for statistical analysis when not normally distributed. Omnibus statistical tests were conducted using
repeated-measures analyses of variance (RM-ANOVAs). Assumptions of normality, heterogeneity of variances, and independence of error were evaluated prior to performing parametric statistical tests. For all RM-ANOVAS, *post-hoc* analyses (Fisher’s LSD) were conducted when appropriate.

### 2.4.3.1 Baseline performance

To test for differences in baseline STT performance between conditions, we conducted a two-way Condition (Ex, Rest) by Sequence (Repeated, Random) RM-ANOVA with target response time during the first sequence of each practice day as the dependent variable.

### 2.4.3.2 Motor skill acquisition and consolidation

To test whether a single session of acute aerobic exercise performed immediately prior to STT practice enhanced motor skill acquisition compared to motor practice preceded by rest, a 2-way Condition (Ex, Rest) by Sequence (Repeated, Random) RM-ANOVA was conducted on the change score value of target response time from the last practice block to the first practice block.

To test whether a single session of acute aerobic exercise performed immediately prior to STT practice enhanced motor memory consolidation compared to motor practice preceded by rest, a 2-way Condition (Ex, Rest) by Sequence (Repeated, Random) RM-ANOVA was conducted on the change score value of target response time from the delayed retention block to last block of practice.

### 2.5 Study 2: Acute aerobic exercise on motor cortical and intracortical circuitry

To determine whether an acute bout of lower limb aerobic exercise modulates corticospinal and intracortical excitability in healthy older adults, single- and paired-pulse transcranial magnetic stimulation (TMS) was performed at two baseline time-points (separated by a 20-minute period of seated rest) prior to an exercise bout, and at two time-points
(immediately, 30 minutes) post-exercise. The purpose of the two baseline measures was to assess the stability of the neurophysiological measures after a 20 minute period of seated rest. The procedure for Study 2 is depicted below in Figure 2.3.

Figure 2.3  Overview of study design for Study 2. Participants underwent neurophysiological assessments before and after a period of seated rest, and at two time-points following an acute bout of aerobic exercise.

2.5.1 Electromyography (EMG)

Surface EMG was used to record motor evoked potentials (MEPs) elicited with transcranial magnetic stimulation. Specifically, 1cm by 1cm Ag/AgCl square surface recording electrodes (Covidien, USA) were placed over the right extensor carpi radialis (ECR) muscle belly for the duration of the experiment. Ground electrodes were placed over the dorsal surface of each hand. EMG signals were triggered by TMS stimuli and collected using LabChart software (LabChart 8.0, AD Instruments Inc., Colorado Springs, CO, USA). Data were pre-amplified (1000 times), band-pass filtered at 10-1000 Hz with PowerLab amplification and EMG systems (AD instruments, USA), sampled at 2000 Hz, and recorded in a 300 ms time window relative to TMS stimuli (100 ms pre- to 200 ms post-stimulus). All EMG data were stored on a personal computer for offline analysis.
2.5.2 Transcranial Magnetic Stimulation (TMS)

During TMS, participants were seated comfortably in a chair. Single- and paired-pulse TMS stimuli were delivered over the left (dominant) hemisphere using the Magstim BiStim\textsuperscript{2} and 200\textsuperscript{2} magnetic stimulators, via a 70mm P/N 9790 figure-of-eight coil (Magstim Co Ltd, Whitland, Carmarthenshire, UK). Coil location for the ECR motor cortical representation was monitored using Brainsight\textsuperscript{TM} neuronavigation system and a template MRI (Rogue Research Inc., Montreal, QC, Canada). Calibration of TMS coil and participant localization in space occurred at the beginning of each TMS session. The TMS coil was held tangentially to the participant’s skull, with the handle pointing laterally and posteriorly at 45° to the mid-sagittal plane, to induce a posterior-to-anterior electrical current over the ECR motor cortical hotspot. RMT was defined as the lowest stimulation intensity required to produce MEPs ≥ 50 μV in the relaxed ECR, in at least five out of 10 consecutive trials. To evaluate corticospinal excitability, recruitment curves (RCs) were collected at 3 different intensities (100% RMT, 130% RMT, 150% RMT). Ten pulses were delivered per intensity for a total of 30 pulses at each time-point, and the order of stimulation intensity was randomized. To evaluate measures of intracortical inhibition and facilitation, paired-pulse TMS (conditioning stimulus (CS) + test stimulus (TS)) was delivered using different intensities and inter-stimulus intervals. First, ten single pulses were delivered using a suprathreshold TS of ~0.3-0.5 mV. Short-interval intracortical inhibition (SICI) was then recorded using a subthreshold CS at 80% RMT, followed by a suprathreshold TS at ~0.3-0.5, separated by an interstimulus interval (ISI) of 2 ms. Intracortical facilitation (ICF) was delivered using the same procedure, except with an ISI of 12ms. Long-interval intracortical inhibition (LICI) was recorded by delivering ten paired-pulses of a CS and TS both at ~0.3-0.5mV, separated by an ISI of 50ms.
2.5.3 TMS Analysis

All raw MEP data were first pre-processed and visually inspected using a custom MATLAB script (Version R2016a, The Mathworks, Inc., Natick, MA, USA). An MEP was excluded from further analysis if there was visible pre-stimulus (100ms) EMG activity. To evaluate corticospinal excitability, the area under the recruitment curve (AURC) was calculated and analyzed taking an average of the 10 stimuli at each stimulus intensity (100%, 130% and 150% RMT), for all participants at each time point. For measure of intracortical circuitry, single- (TS) and paired-pulse (CS + TS) peak-to-peak MEP amplitudes were averaged across the ten trials of each condition. Inhibition and facilitation were then analysed as a ratio of the paired stimuli (CS+TS) over single pulse stimuli (TS) MEP amplitude (mV), and expressed as a percentage of inhibition and facilitation, where greater values (ie. above 100%) represent enhanced facilitation, and lower values (ie. below 100%) represent increased inhibition.

2.5.4 Statistical Analysis

Statistical tests were performed using SPSS statistics 22.0 (IBM Corporation, USA) and Statistica (Version 13, TIBCO Software Inc., USA) using a significance threshold of $\alpha = 0.05$. Data were checked for normality, with the Shapiro-Wilks significance level set at $p < 0.001$ (104) and visual inspection of histogram plots. Data were log-transformed for statistical analysis when not normally distributed. Omnibus statistical tests were conducted using repeated-measures analyses of variance (RM-ANOVAs). Assumptions of normality, heterogeneity of variances, and independence of error were evaluated prior to performing parametric statistical tests. For all RM-ANOVAS, post-hoc analyses (Fisher’s LSD) were conducted when appropriate.
2.5.4.1 Neurophysiological measures

To confirm there were no differences in neurophysiological measures between the two baseline time-points (ie. $T_0$ and $T_1$), separate paired samples $t$-tests were conducted for AURCs, SICI, ICF, and LICI. To evaluate whether a single session of acute aerobic exercise modulated corticospinal excitability, intracortical inhibition, and intracortical facilitation, one-way Time ($T_1$, $T_2$, $T_3$) RM-ANOVAs were conducted separately for each dependent measure. Post-hoc analysis was applied using Fisher’s LSD where appropriate.
Chapter 3: Results

3.1 Participants

Participant demographic and exercise metrics are described below in Table 3.1. Out of the 22 individuals who participated in this study, 14 were female and 8 were male. Due to logistical reasons, we were unable to record airflow and expired gases from one female participant during the exercise stress test (S02). All other participants achieved at least one criterion for VO$_{2\text{peak}}$ during the stress test. With the exception of S02, each participant’s ventilatory threshold was calculated post hoc, and each participant’s prescribed standardized bout of moderate-intensity exercise was confirmed to be below anaerobic threshold. For individual aerobic exercise data, see Appendix F.

<table>
<thead>
<tr>
<th>Demographics</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>64.5</td>
<td>6.3</td>
<td>[55, 74]</td>
</tr>
<tr>
<td>BMI (kg/m$^2$)</td>
<td>23.9</td>
<td>3.1</td>
<td>[10.2, 33.2]</td>
</tr>
<tr>
<td>Handedness</td>
<td>89.5</td>
<td>13.3</td>
<td>[60, 100]</td>
</tr>
<tr>
<td>MoCA</td>
<td>28.0</td>
<td>1.6</td>
<td>[24, 30]</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Exercise Stress Test</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>VO$_{2\text{peak}}$ (ml/kg/min)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males</td>
<td>32.5</td>
<td>12.8</td>
<td>[16.6, 54.1]</td>
</tr>
<tr>
<td>Females</td>
<td>24.7</td>
<td>4.4</td>
<td>[19.1, 32.8]</td>
</tr>
<tr>
<td>PO$_{\text{peak}}$ (watts)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males</td>
<td>131.9</td>
<td>49.0</td>
<td>[80, 210]</td>
</tr>
<tr>
<td>Females</td>
<td>88.3</td>
<td>18.3</td>
<td>[60, 120]</td>
</tr>
</tbody>
</table>

Table 3.1 Participant demographics and exercise metrics: age, body mass index (BMI), handedness (Edinburgh Handedness Inventory: Right $\geq$ 60), Montreal Cognitive Assessment (MoCA): unimpaired $\geq$ 24, peak level of oxygen consumption during stress test (VO$_{2\text{peak}}$), peak power output during exercise stress test (PO$_{\text{peak}}$).
3.2 Study 1: Effects of acute aerobic exercise on motor performance and learning

All STT data were found to be normally distributed at all time-points.

3.2.1 Baseline performance

The RM-ANOVA examining initial performance demonstrated no main effect of Condition ($F_{(1, 21)}=0.002, p=0.964$) or Sequence ($F_{(1, 21)}=0.580, p=0.455$), and no Condition by Sequence interaction effect ($F_{(1, 21)}=1.501, p=0.234$). Therefore, initial STT performance was not significantly different between conditions for either sequence.

3.2.2 Motor skill acquisition

Two-way ANOVA results revealed a main effect of Sequence ($F_{(1, 21)}=12.931, p=.002$, $\eta^2_{\text{partial}}=.381$), with greater improvements for the repeated sequences relative to the random sequences, regardless of condition. There was no main effect of Condition ($F_{(1, 21)}=0.064, p=0.803$, $\eta^2_{\text{partial}}=.003$), and no Condition by Sequence interaction ($F_{(1, 21)}=0.419, p=0.524$, $\eta^2_{\text{partial}}=.020$). These results can be seen in Figure 3.1A.

3.2.3 Motor memory consolidation

Two-way ANOVA results demonstrated a trend towards significance for the Sequence by Condition interaction effect ($F_{(1, 21)}=3.999, p=.059$, $\eta^2_{\text{partial}}=.160$), such that under the exercise condition there were greater improvements in both sequences compared to rest, however the difference was not statistically significant. There was no main effect of Sequence ($F_{(1, 21)}=3.230, p=.087$, $\eta^2_{\text{partial}}=.133$), and no main effect of Condition ($F_{(1, 21)}=1.401, p=.250$, $\eta^2_{\text{partial}}=.063$). These results can be seen in Figure 3.1B.
3.2.4 Recognition

Across the group, participants did not demonstrate explicit knowledge of a repeated sequence during the recognition testing. The repeated sequence was correctly identified at a level less than chance (0.95 ± 1.05/3 or 31.81 ± 34.85 % correct).

3.2.5 Effect size calculations

To quantify the effect of acute exercise on motor skill acquisition, the effect size (Cohen’s $d$) was calculated from the difference between performance (last practice block – first practice block) divided by the pooled SD. Similarly, the effect of acute exercise on motor memory consolidation was calculated from the difference between performance (retention block – last practice block) divided by the pooled SD. Effect sizes were calculated separately for condition (exercise and rest) as well as sequence (repeated and random) (Table 3.2).

<table>
<thead>
<tr>
<th>Effect size (Cohen’s $d$)</th>
<th>Skill acquisition</th>
<th>Consolidation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Exercise</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Repeated sequences</td>
<td>0.35</td>
<td>0.18</td>
</tr>
<tr>
<td>Random sequences</td>
<td>0.19</td>
<td>0.17</td>
</tr>
<tr>
<td>Rest</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Repeated sequences</td>
<td>0.45</td>
<td>0.02</td>
</tr>
<tr>
<td>Random sequences</td>
<td>0.28</td>
<td>0.11</td>
</tr>
</tbody>
</table>

Table 3.2. Effect sizes (Cohen’s $d$) calculated for acquisition and consolidation. Effect sizes were calculated separately for each condition (exercise, rest) as well as sequence (repeated, random).
Figure 3.1  
Response time per target changes scores for motor skill acquisition (A), and motor memory consolidation (B). Average target response times per block are represented for the skill acquisition period (C) and delayed retention test, compared to the last practice block (D). Error bars represent standard error of the mean (SEM).
3.3 Study 2: Effects of acute aerobic exercise on neurophysiology

After visual inspection of the data, one participant (S05) was removed from the ARUC data set due to excessive background noise in EMG trials. Thus, 21 participants were included in the final ARUC analysis, while 22 participants were included in the SICI, ICF, and LICI analyses. LICI was found to be non-normal at one time-point, thus all LICI data were log-transformed prior to statistical analysis. However, for data visualization, all plots display raw data. All other measures were normally distributed across all time-points.

3.3.1 Baseline measures

The paired-samples t-tests revealed no differences between T₀ and T₁ for ARUC (t(20) = -1.516, p = .134), SICI (t(21) = -.598, p = 0.556), ICF (t(21) = -1.116, p = 0.277), or LICI (t(21) = 1.675, p = .109), indicating that a 20-minute period of seated rest did not impact any of the neurophysiological measures.

3.3.2 Neurophysiological measures

One-way ANOVA results demonstrated a main effect of time (F(2, 42)=4.629, p=.015, \( \eta^2_{\text{partial}}=0.181 \)) for LICI. Post-hoc analysis revealed a significant increase in inhibition from pre-exercise (T1) to immediately post-exercise (T2) (p=.030), and this effect returned to pre-exercise levels at 30-minutes post-exercise (T3) (p=.006). There were no significant effects of time on ARUC (F(2, 40)=2.211, p=0.123, \( \eta^2_{\text{partial}}=0.100 \)), SICI (F(2, 42)=0.049, p=0.952, \( \eta^2_{\text{partial}}=0.002 \)), or ICF (F(2, 42)=0.707, p=0.499, \( \eta^2_{\text{partial}}=0.033 \)). Results for Study 2 can be seen in Figure 3.2.
Figure 3.2 Influence of a single session of moderate-intensity aerobic exercise on neurophysiological measures. A) Long-interval intracortical inhibition (LICI), B) area under the recruitment curve (AURC), C) short-interval intracortical-inhibition (SICI), D) intracortical facilitation (ICF). Error bars depict standard error of the mean (SEM).
Chapter 4: Discussion

These experimental studies are the first to investigate the effects of a single session of aerobic exercise on motor performance and learning, as well as motor cortical and intracortical networks, in healthy older adults. Based on previous findings in young healthy adults, we hypothesized that exercising at a moderate intensity before practicing the STT would lead to significantly improved motor skill acquisition and consolidation, as well as modulations in intracortical inhibition and facilitation, compared to a period of seated rest. In Study 1, performing aerobic exercise immediately prior to motor task practice did not facilitate differences in measures of motor skill acquisition or consolidation. However, we observed a non-significant trend towards improvements in offline motor memory consolidation, such that under the exercise condition there was greater change in repeated sequences only compared to rest. In Study 2, we found that after exercise there was an increase in LICI, which returned to near baseline levels by 30 minutes post-exercise. In contrast, we did not observe any significant changes in motor cortical excitability, SICI, or ICF after exercise. This may imply that acute aerobic exercise has differential effects on distinct intracortical circuits within the motor cortex. Taken together, these findings suggest that a single bout of moderate-intensity aerobic exercise transiently modulates GABA-B receptor mediated intracortical inhibition in healthy older adults, however, no significant exercise-induced changes in motor behavior were observed.

4.1 Study 1: Effects of acute exercise on motor performance and learning

4.1.1 Motor skill acquisition

In the present study, acute aerobic exercise performed prior to practicing the STT did not impact motor skill acquisition. In previous studies with young healthy adults, an acute bout of moderate-intensity aerobic exercise performed prior to motor practice has been shown to
maintain (79) and improve (78) spatial accuracy, and increase acceleration (80) of various types of motor tasks during skill acquisition, which may reflect enhanced encoding processes. Further, a meta-analysis on the effects of acute, moderate-intensity exercise revealed a strong beneficial effect for response time on working memory type tasks (72). Thus, we hypothesized that cycling at moderate-intensity prior to motor practice might also lead to significantly faster response times during STT skill acquisition in our sample of healthy older adults. However, in the present study participants’ target response times improved similarly during motor skill acquisition, under both the exercise and rest conditions.

Cognitive studies suggest that moderate-intensity exercise has the most beneficial effects on information processing speeds and levels of arousal, compared to low- or high-intensity exercise (68,105). However, the progressive slowing of reaction times associated with advancing age may be due to changes in response generation, rather than information processing and sensorimotor stages (106). Thus, although acute exercise has been thought to enhance encoding processes during skill acquisition in young healthy adults, these exercise-induced effects may not impact motor behavior in healthy older adults. Considering that performance effects are highly transient and susceptible to numerous variables such as motivation and arousal (31), it is also possible that exercising prior to motor practice has detrimental effects on skill acquisition by inducing fatiguing effects (75), or increasing neural noise (72). However, given there were no statistically significant differences between conditions during skill acquisition, and similar levels of variability in performance were demonstrated under the exercise and rest conditions, these explanations remain speculative. Additionally, we did not control for attention during performance of the STT, which is a limitation of this study. Older adults often experience poorer
attention (107), so understanding how levels of attention impacted the results in the present study may have helped with the interpretation of findings.

It is possible that the null impact of exercise on skill acquisition in the present study was attributed to the prescription of moderate-intensity exercise. High-intensity exercise is also known to enhance information processing speed and levels of arousal (86). In addition, high-intensity exercise has been associated to a greater extent than moderate-intensity exercise with the alteration of neurotransmitters and growth factors that are known to facilitate memory formation (92). For instance, higher levels of lactate after exercise are associated with better skill acquisition (90), so our null finding might be due to the prescription of moderate-intensity exercise confirmed to be below each individual’s anaerobic threshold. However, our rationale for moderate-intensity was based on multiple studies in young healthy adults demonstrating positive effects on skill acquisition and consolidation at moderate-intensities of exercise (78–81). Because age-related degradations in dopaminergic neurotransmission are related to motor deficits (7), it is possible that higher intensities of exercise may be necessary to affect skill acquisition in older adults. However, interactions between exercise-induced neurochemicals and memory processes are highly complex, and larger concentrations of these substrates as a result of higher exercise intensities may not necessarily translate to better behavioural outcomes (73). To our knowledge, the present study is the first to investigate the effects of acute aerobic exercise on skill acquisition in healthy older adults, thus future work to determine the role of exercise intensity is warranted.

In the present study, greater change in motor behavior during practice was noted for repeated sequences as compared to random sequences, after both exercise and rest. Moreover, participants did not demonstrate explicit recognition of the repeated sequence. Taken together,
these results suggest that implicit sequence-specific improvements in skill acquisition occurred during STT practice, similarly under both exercise and rest conditions. This is in line with previous findings from our laboratory demonstrating enhanced performance on repeated relative to the random sequences, after both high-intensity exercise and rest, during practice of a similar discrete motor task in young healthy adults (77). Mang et al. (2016) suggested these results could be attributed to similar sequence-specific encoding processes occurring during exercise and rest conditions (77). Alternately, Roig and colleagues (2012) suggested the possibility of exercise-induced improvements in encoding processes being cancelled out due to fatigue-related decreases in performance (75). However, other work by Mang and colleagues demonstrated that high-intensity exercise improved temporal accuracy of repeated, relative to random, sequences during skill acquisition of a continuous tracking task, and these improvements were maintained at retention (27). Collectively with previous work, the null finding in the present study provides further evidence that the impact of exercise on implicit sequence-specific skill acquisition may also depend on the type of motor task, and/or exercise intensity (22,73).

4.1.2 Motor memory consolidation

Based on previous findings in young healthy adults (74,75,77,81) and individuals with chronic stroke (108) demonstrating that both moderate- and high-intensity acute exercise can facilitate offline gains in motor learning, we anticipated that a single session of moderate-intensity exercise performed prior to STT practice might improve motor memory consolidation in healthy older adults. Acute exercise performed closely in time with task practice is thought to enhance offline gains and consolidation processes by stabilizing the motor memory trace into a more robust form (22). Possible mechanisms through which this might occur are by facilitating synaptic plasticity (ie. LTP processes) and promoting resistance against interfering stimuli, or by
increasing the availability of neurotrophins and catecholamines that play an important role in memory formation (73). In the present study there appeared to be greater improvements in motor memory consolidation for both sequences after exercise, whereas under the rest condition, response times seemed to improve to a lesser degree for the random sequences and remain at similar levels for the repeated sequences. However, the interaction effect did not reach statistical significance ($p=.059$).

This null finding might be explained by several reasons. First, the temporal placement of the acute exercise bout prior to motor practice may have diminished any possible effects on the consolidation phase. In young healthy adults, acute exercise performed after motor practice demonstrates largest effects on motor memory consolidation, and these effects have been shown to decrease as the time between exercise and motor practice is increased (74,75). Given that older individuals consistently report deficits consolidating learned motor sequences (33), exercise may need to be performed following motor practice to maximize the effects of exercise on consolidation processes in this population. Although acute exercise prior to task practice improves offline gains in young healthy adults (27,75,77), the use of higher intensities may have helped the exercise effects persist long enough after its termination to impact the consolidation phase (73). For instance, in a direct comparison of acute moderate- and high-intensity exercise performed after task practice in young healthy adults, both exercise intensities enhanced consolidation, high-intensity a greater extent (81). Additionally, evidence suggests there is a decline in LTP-like plasticity with age (9), so it is possible that a single session of aerobic exercise is not enough to facilitate changes in motor behavior in healthy older adults. It has been speculated that multiple bouts of exercise paired with motor practice might facilitate cumulative gains (108); this may positively benefit consolidation processes in healthy older adults.
When interpreting the effects of acute exercise on motor memory, it is also important to consider participant characteristics such as age and fitness levels (73). Although several studies in young healthy adults report positive effects of acute exercise on motor memory consolidation, the majority of these studies were conducted in participants with very high fitness levels, with VO$_{2\text{peak}}$ scores ranging from 43-53 ml/kg/min (see Table 1.1). The inclusion of a wide range of participants in terms of age (55-75 years old) and fitness levels (VO$_{2\text{peak}}$: 16.6-54.1 ml/kg/min) within the current study likely relates to the observed high variability in motor performance during both practice and retention, and is a potential limitation. For instance, Etnier and colleagues (2001) demonstrated that 51% of the variance in retention of a motor skill can be attributed to age and fitness, whereby there is an inverse relationship with age and a positive relationship with aerobic fitness (109). As the current study was proof-of-concept and designed to evaluate whether positive effects observed in young healthy individuals could be replicated in healthy older adults, the wide range in participant inclusion criteria was intentional to increase the external validity of results to an aging population with varying characteristics. However, this may have affected internal validity and decreased statistical power. The power calculation of a sample size of 22 participants for the present study was based on a similar study conducted in our laboratory examining the effects of acute high-intensity exercise on STT acquisition and consolidation in young healthy adults (average age = 26 years), using a beta criterion level set at 0.8 (77). Considering the average age of participants in the current study is 65 years old, and the observed power for the Sequence by Condition interaction is 0.479, low statistical power could be one possible reason for not detecting a statistically significant interaction effect. While the results from the current study should be interpreted with caution due to the lack of statistically
significant differences, the data points towards the potential role of exercise in enhancing motor memory consolidation in healthy older adults.

4.1.3 Limitations

As previously mentioned, low statistical power and high variability may have limited our capacity to detect exercise-induced changes in motor memory consolidation. Another limitation is that we did not directly measure sleep quality. Sleep may be a potential confounding variable as age-related declines in sleep quality have been shown to impact motor memory consolidation (110). In the present study, we asked participants to complete a screening form to determine levels of risk for obstructive sleep apnea (STOP-Bang Questionnaire). Although risk for sleep apnea did not correlate with any of our behavioural measures in the present study, the STOP-Bang Questionnaire does not objectively measure sleep quality, which may have been helpful data in the interpretation of these results. Future research should account for the effect of age-related declines in sleep quality on motor memory consolidation using objective measures.

4.2 Study 2: Effects of acute exercise on motor cortical and intracortical networks

4.2.1 Acute exercise modulated GABA-B, but not GABA-A mediated intracortical inhibition

In Study 2, we noted that immediately after exercise, there was an increase in LICI, and this returned to near baseline levels within a 30-minute period. LICI reflects inhibition mediated by GABA-B receptors at the cortical level, directly modulates corticospinal output (54–56) and is thought to facilitate the induction of LTP (111). In healthy older adults, a decreased ability to modulate GABA-mediated intracortical inhibition has been linked with declines in motor performance (15). Although preliminary, our finding that an acute bout of moderate-intensity
exercise transiently modulates GABA-B mediated intracortical inhibition in healthy older adults may reflect a precursor to cortical plasticity (111), and is an interesting direction for future studies.

Our finding differs from previous studies in young healthy adults that showed either a reduction or no change in LICI after an acute bout of exercise (18,24,25). There is evidence to suggest that intracortical inhibition changes in opposite directions for young and older adults during motor skill acquisition (112), so this might explain why the observed effect of exercise on LICI in the present study is towards an increase in inhibition, whereas studies in younger adults demonstrate a decrease (18,25). Another reason contributing to our finding could be that on average (±standard error), our participants did not show baseline levels of inhibition (103.1±17.2% of test stimulus amplitude; levels below 100% reflect inhibition, and levels above 100% reflect facilitation). Given that there were reduced baseline levels of LICI in the present study, a potential ceiling effect may explain why the direction of change was towards an increase in inhibition, and differs from what was measured in young healthy adults (18,25). The reduction in LICI at baseline in the present study is actually in support of other work demonstrating a reduction in intracortical inhibition with increasing age (9,14,64), and reduced resting-state inhibition is associated with the loss of modulatory capacity (64). Considering that the age-related inability to modulate intracortical inhibition has been linked with motor declines (7,15,64), the modulation of LICI after exercise in the current study, regardless of direction, may still have positive implications for neurorehabilitation. Further work is needed to determine normative values of exercise-induced changes in adults across the lifespan.

The null effect of acute exercise on SICI in the present study contrasts with studies in young healthy adults showing a reduction in SICI after acute exercise (18,24,26,29). The present
study adds to the literature by demonstrating that an acute bout of exercise did not modulate SICI in healthy older adults. Evidence suggests there are age-related differences in resting values of SICI; a large-scale study obtaining TMS measures across multiple age groups noted a significant reduction in SICI with increasing age (14). Further, multiple studies demonstrate an age-related inability to modulate GABA-A mediated inhibition, which has been linked to motor deficits (9,14,15,64). Thus, the lack of change in SICI after exercise in the present study may be due to age-related changes to GABAergic transmission within the motor cortex.

4.2.2 Acute exercise did not impact intracortical facilitation or corticospinal excitability

The null impact of acute exercise on intracortical facilitation (ICF) in the present study is not surprising, considering the inconsistent results on this measure after acute exercise in young healthy adults. These inconsistencies may be attributed to differences in TMS or exercise parameters (26), such as varying ISIs, which are thought to probe different neural populations (23). However, even in young adults, cortical mechanisms underlying ICF at rest are not fully understood (18), so it is difficult to interpret the effects of exercise on neural mechanisms underlying ICF. No robust age-related changes in ICF have been documented (9), so it is unclear whether age-related changes to the mechanisms underlying ICF contributed to the present findings. This study furthers our knowledge by being the first to evaluate changes in ICF after an acute bout of exercise in healthy older adults. However, future research is needed to better understand the impact of exercise on this neuronal circuit, and how any exercise-induced changes might relate to LTP-like processes.

Lastly, the present study also demonstrated that corticospinal excitability (as assessed using the area under the recruitment curve, AURC) did not change significantly after exercise. This is in line with most studies in young healthy adults assessing the impact of lower-limb
exercise on the cortical excitability of an upper limb muscle representation, using single-pulse TMS (18,24,25,29). A potential reason exercise did not impact corticospinal excitability in the present study is due to the wide range of fitness levels in our sample. Physical activity levels are known to mediate exercise-induced changes in corticospinal excitability (26), thus fitness likely plays a role. Further, the age-related decrease in corticospinal excitability is well documented, and has been linked with age-related structural changes like cortical atrophy (9,14). Thus, we cannot rule out the possibility that age-related brain changes impacted the null effect of exercise on modulating cortical excitability in this study. Our results should be interpreted with caution as we did not account for possible exercise-induced changes in muscle contractility that may have impacted MEP amplitudes, such as fatigue (73). Future work should consider the impact of fitness, as well as muscle fatigue, when evaluating changes in corticospinal excitability after exercise across the lifespan.

### 4.2.3 Limitations

A potential limitation of this study is that changes in cortical excitability after exercise were assessed in a single arm muscle, and is it possible that exercise influenced cortical excitability in other non-exercised muscles. Further, older adults tend to rely on both hemispheres as a compensatory mechanism due to age-related structural and functional brain changes (15). Thus, examining a single muscle in one hemisphere might have limited our ability to fully capture exercise-induced changes in cortical excitability of the upper limb. Future work should consider evaluating changes in excitability of other muscle representations within the motor cortex, and other cortical regions. A second limitation of this study is that we did not re-test RMT after exercise. RMT was collected at the initial timepoint to determine testing intensities, which needs to remain consistent throughout the testing session to make accurate
comparisons (113). However, acute exercise has not been shown to change RMT in young healthy adults (26). Further, it is unlikely that this influenced paired-pulse measures due to the normalization of paired-pulse MEPs to the 0.3-0.5mV test stimulus, which was determined at each time-point.

4.3 Conclusion and Future Directions

The benefits of exercise on the brain are becoming widely known. However, we are just beginning to understand the potential of aerobic exercise to optimize the consolidation of motor memories across the lifespan. To our knowledge, the current studies are the first to investigate the effects of aerobic exercise on motor performance and learning, as well as motor cortical and intracortical circuits, in healthy older adults. Acute moderate-intensity exercise did not demonstrate significant changes in skill acquisition or consolidation in healthy older adults, however continued efforts should address factors such as exercise intensity and timing, participant age and fitness levels, and type of motor task. Although preliminary, our finding that acute exercise modulated GABA-B receptor mediated intracortical inhibition supports the hypothesis that aerobic exercise has potential to act as an endogenous neuromodulation strategy. However, changes in neurophysiology may not necessarily translate to improved behavioural outcomes. Further work is needed to understand the effects of exercise on motor cortical excitability in different muscles, as well as brain regions other than the motor cortex. Additionally, the relationship between exercise-induced changes in neurophysiological circuits and exercise-induced changes in motor behavior is ripe for exploration. The identification of biomarkers to predict an individual’s capacity to respond to exercise as a neuromodulation strategy is also an interesting future direction to consider.
As the world’s population continues to rise steadily, investigating strategic and innovative ways to promote functional independence and healthy aging is important and timely. Although the benefits of physical activity and exercise for health are widely understood, research is critical to maximize its effectiveness and to inform evidence-based recommendations. In this thesis, we were interested in the potential use of exercise as an adjunct neuromodulation strategy to facilitate motor learning in healthy older adults, and further explored potential underlying mechanisms through which exercise exerts its influence on the brain. Understanding the effects of aerobic exercise on neural mechanisms as well as behaviour has large implications for neurorehabilitation in clinical populations.
References


111. Mott, David D. and Lewis DV. Facilitation of the Induction of Long-Term Potentiation by


Appendices

Appendix A: Edinburgh Handedness Inventory

Participant Code: ______________

Please indicate with a check (□) your preference in using your left or right hand in the following tasks.

Where the preference is so strong you would never use the other hand, unless absolutely forced to, put two checks (□□).

If you are indifferent, put one check in each column ( □ | □).

Some of the activities require both hands. In these cases, the part of the task or object for which hand preference is wanted is indicated in parentheses.

<table>
<thead>
<tr>
<th>Task / Object</th>
<th>Left Hand</th>
<th>Right Hand</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Writing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Drawing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3. Throwing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4. Scissors</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5. Toothbrush</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6. Knife (without fork)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7. Spoon</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8. Broom (upper hand)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>9. Striking a Match (match)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10. Opening a Box (lid)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Total checks:  
CT = \( \text{LH} + \text{RH} \)

Difference  
D = \( \text{RH} - \text{LH} \)

Result  
\( R = \left( \frac{D}{CT} \right) \times 100 \)

Interpretation:  
(Left Handed: \( R < -40 \))  
(Ambidextrous: \(-40 \leq R \leq +40 \))  
(Right Handed: \( R > +40 \))
Appendix B: Montreal Cognitive Assessment

Montreal Cognitive Assessment (MOCA)

**VISUOSPATIAL / EXECUTIVE**

- Copy cube
- Draw clock (ten past eleven)

**NAMING**

- Rhinoceros
- Camel

**MEMORY**

- Read list of words, subject must repeat them. Do 2 trials. Do a recall after 5 minutes.

**ATTENTION**

- Read list of digits (1 digit/sec.). Subject has to repeat them in the forward order.
- Read list of letters. The subject must tap with his hand at each letter. A no points if ≥ 2 errors.

**LANGUAGE**

- Repeat: I only know that John is the one to help today.
- Fluency: Name maximum number of words in one minute that begin with the letter F.

**ABSTRACTION**

- Similarity between e.g. banana - orange = fruit
- Train - bicycle

**DELAYED RECALL**

- Has to recall words with no cue

**ORIENTATION**

- Date
- Month
- Year
- Day
- Place
- City

Subject #  
Date:  

Total: __/30

Add 1 point if ≤ 12 yr ed.
Appendix C: TMS Screening Form

BRAIN BEHAVIOR LAB
TRANSCRANIAL MAGNETIC STIMULATION (TMS) SCREENING FORM

Below is a questionnaire used to exclude participants considered not suitable for transcranial magnetic stimulation (TMS). This information, as well as your identity, will be kept confidential.
Plekase complete form below:

Participant Code: ________________________________

Please circle one:

Neurological or Psychiatric Disorder
YES NO Multiple Sclerosis
YES NO

Head Trauma
YES NO Depression
YES NO

Stroke
YES NO Clinical Depression
YES NO

Brain surgery
YES NO Treatment with amitriptyline and haloperidol
YES NO

Metal in cranium
YES NO Implanted medication pump
YES NO

Brain Lesion
YES NO Intracranial Pathology
YES NO

Pacemaker
YES NO Albinoism
YES NO

History of seizure
YES NO Intractable anxiety
YES NO

Family history of epilepsy
YES NO Prognostic
YES NO

History of epilepsy
YES NO Headache or Hearing problems
YES NO

Intracorporal electronic devices
YES NO Family History of Hearing Loss
YES NO

Intracardiac lines
YES NO Other medical conditions
YES NO

If you answered “yes” to any of the above questions, please provide details below.

________________________________________________________________________________________
Appendix D: Exercise Stress Test Protocol (adapted from Klassen et al. (2017))

Participant code:  Evaluator Initials:  Date:
DOB:  Age:  Height:  Weight:

Medical Diagnosis/Date of injury:

Past Medical History:

Current Medications:

**Resting HR:** _____(*Must be < 100 bpm)  **Resting BP:** _____(*Must be <160/110 mm Hg)

Is the participant on a beta-blocker medication? □ Yes  □ No

*Ensure that all members of the testing team are aware if the participant is on a beta-blocker*

**Maximum HR for exercise test:**
Age-predicted max: 206.9 – (0.67 x age) = _____  OR Age-predicted max (on a beta-blocker):
164 – (0.7 x age) = ____

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Increase (W)</th>
<th>Watts</th>
<th>HR (at end of minute)</th>
<th>RPE (at end of minute)</th>
<th>BP (every second trial)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-2</td>
<td>Warm Up</td>
<td>10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>5/10/15</td>
<td>20</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>5/10/15</td>
<td>30</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>5/10/15</td>
<td>40</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>5/10/15</td>
<td>50</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>5/10/15</td>
<td>60</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>5/10/15</td>
<td>70</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>5/10/15</td>
<td>80</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>5/10/15</td>
<td>90</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>5/10/15</td>
<td>100</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>5/10/15</td>
<td>110</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>5/10/15</td>
<td>120</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>5/10/15</td>
<td>130</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>5/10/15</td>
<td>140</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>16</td>
<td>5/10/15</td>
<td>150</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>5/10/15</td>
<td>160</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18</td>
<td>5/10/15</td>
<td>170</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19</td>
<td>5/10/15</td>
<td>180</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>5/10/15</td>
<td>190</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-5 min</td>
<td>Cool Down</td>
<td>10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-5 min</td>
<td>Cool Down</td>
<td>10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5 min</td>
<td>Rest /Sitting</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Bike Seat Position:
Therapist Assistance During Test: Y
Appendix E: Borg Rating of Perceived Exertion (RPE) Scale

<table>
<thead>
<tr>
<th></th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Nothing at all</td>
</tr>
<tr>
<td>1</td>
<td>Very Light</td>
</tr>
<tr>
<td>2</td>
<td>Light</td>
</tr>
<tr>
<td>3</td>
<td>Moderate</td>
</tr>
<tr>
<td>4</td>
<td>Somewhat Hard</td>
</tr>
<tr>
<td>5</td>
<td>Hard</td>
</tr>
<tr>
<td>6</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>Very Hard</td>
</tr>
<tr>
<td>8</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Very, Very Hard (Absolute Maximum)</td>
</tr>
</tbody>
</table>
## Appendix F: Individual exercise data

### Table F.1 Individual exercise data

<table>
<thead>
<tr>
<th>Participant</th>
<th>Age</th>
<th>Sex</th>
<th>VO&lt;sub&gt;2peak&lt;/sub&gt;</th>
<th>Aerobic Exercise Test (Final Stage)</th>
<th>Exercise Bout (Study 1)</th>
<th>Exercise Bout (Study 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>VO&lt;sub&gt;2peak&lt;/sub&gt;</td>
<td>PO</td>
<td>HR</td>
</tr>
<tr>
<td>M01</td>
<td>63</td>
<td>M</td>
<td>26.7</td>
<td>26.7</td>
<td>100</td>
<td>149</td>
</tr>
<tr>
<td>M02</td>
<td>72</td>
<td>F</td>
<td>28.1</td>
<td>28.1</td>
<td>130</td>
<td>163</td>
</tr>
<tr>
<td>M03</td>
<td>62</td>
<td>M</td>
<td>23.2</td>
<td>23.2</td>
<td>60</td>
<td>148</td>
</tr>
<tr>
<td>M04</td>
<td>71</td>
<td>F</td>
<td>27.7</td>
<td>27.7</td>
<td>95</td>
<td>121</td>
</tr>
<tr>
<td>M05</td>
<td>73</td>
<td>M</td>
<td>27.5</td>
<td>27.5</td>
<td>110</td>
<td>157</td>
</tr>
<tr>
<td>M06</td>
<td>58</td>
<td>F</td>
<td>29.3</td>
<td>29.3</td>
<td>100</td>
<td>182</td>
</tr>
<tr>
<td>M07</td>
<td>71</td>
<td>F</td>
<td>19.1</td>
<td>19.1</td>
<td>80</td>
<td>159</td>
</tr>
<tr>
<td>M08</td>
<td>65</td>
<td>F</td>
<td>23</td>
<td>23</td>
<td>80</td>
<td>177</td>
</tr>
<tr>
<td>M09</td>
<td>57</td>
<td>F</td>
<td>23.2</td>
<td>23.2</td>
<td>80</td>
<td>137</td>
</tr>
<tr>
<td>M11</td>
<td>57</td>
<td>M</td>
<td>29</td>
<td>29</td>
<td>100</td>
<td>173</td>
</tr>
<tr>
<td>M12</td>
<td>56</td>
<td>F</td>
<td>32.8</td>
<td>32.8</td>
<td>110</td>
<td>161</td>
</tr>
<tr>
<td>M13</td>
<td>55</td>
<td>M</td>
<td>54.1</td>
<td>54.1</td>
<td>210</td>
<td>156</td>
</tr>
<tr>
<td>M14</td>
<td>62</td>
<td>F</td>
<td>21.9</td>
<td>21.9</td>
<td>80</td>
<td>146</td>
</tr>
<tr>
<td>M15</td>
<td>59</td>
<td>F</td>
<td>31.3</td>
<td>31.3</td>
<td>120</td>
<td>172</td>
</tr>
<tr>
<td>M16</td>
<td>69</td>
<td>F</td>
<td>26.6</td>
<td>26.6</td>
<td>100</td>
<td>164</td>
</tr>
<tr>
<td>M17</td>
<td>65</td>
<td>F</td>
<td>19.6</td>
<td>19.6</td>
<td>60</td>
<td>145</td>
</tr>
<tr>
<td>M18</td>
<td>57</td>
<td>M</td>
<td>50.1</td>
<td>50.1</td>
<td>205</td>
<td>161</td>
</tr>
<tr>
<td>M19</td>
<td>70</td>
<td>M</td>
<td>16.6</td>
<td>16.6</td>
<td>80</td>
<td>113</td>
</tr>
<tr>
<td>M20</td>
<td>65</td>
<td>M</td>
<td>27.9</td>
<td>27.9</td>
<td>120</td>
<td>161</td>
</tr>
<tr>
<td>M21</td>
<td>65</td>
<td>F</td>
<td>21.9</td>
<td>21.9</td>
<td>70</td>
<td>170</td>
</tr>
<tr>
<td>M22</td>
<td>72</td>
<td>F</td>
<td>21.8</td>
<td>21.8</td>
<td>80</td>
<td>163</td>
</tr>
<tr>
<td>Mean</td>
<td>64.5</td>
<td>27.7</td>
<td>103.2</td>
<td>156.1</td>
<td>1.2</td>
<td>9.6</td>
</tr>
<tr>
<td>SD</td>
<td>6.3</td>
<td>9.1</td>
<td>9.1</td>
<td>16.7</td>
<td>0.15</td>
<td>0.9</td>
</tr>
</tbody>
</table>
Appendix G: Calculation of ventilatory thresholds

Figure G.7. Ventilatory thresholds were calculated post-hoc by the V-Slope method (A), further confirmed by ventilatory equivalent method (B) and RER values $\geq 1.10$ (C).
Appendix H: Sleep Quality Questionnaire (for retention test days)

On a scale of 0-10, with “0” being worst quality sleep and “10” being best quality sleep, please rate how your overall sleep quality was last night by placing a single vertical line through the scale provided below.

Is this typical?

_____________________________________________________________________

Worst quality sleep

Best quality sleep
Appendix I: Godin Leisure Time Questionnaire

<table>
<thead>
<tr>
<th>Exercise Type</th>
<th>Times Per Week</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) STRENUOUS EXERCISE</td>
<td></td>
</tr>
<tr>
<td>(e.g., running, jogging, hockey, football, soccer, squash, basketball, cross country skiing, judo, roller skating, vigorous swimming, vigorous long distance bicycling)</td>
<td></td>
</tr>
<tr>
<td>b) MODERATE EXERCISE</td>
<td></td>
</tr>
<tr>
<td>(e.g., fast walking, baseball, tennis, easy bicycling, volleyball, badminton, easy swimming, alpine skiing, popular and folk dancing)</td>
<td></td>
</tr>
<tr>
<td>c) MILD EXERCISE</td>
<td></td>
</tr>
<tr>
<td>(e.g., yoga, archery, fishing from river bank, bowling, horseshoes, golf, snow-mobiling, easy walking)</td>
<td></td>
</tr>
</tbody>
</table>

2. During a typical 7-Day period (a week), in your leisure time, how often do you engage in any regular activity long enough to work up a sweat (heart beats rapidly)?

<table>
<thead>
<tr>
<th>Frequency</th>
<th>1.</th>
<th>2.</th>
<th>3.</th>
</tr>
</thead>
<tbody>
<tr>
<td>OFTEN</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SOMETIMES</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NEVER/RARELY</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>