The Examination of Potential Mechanisms Underlying the Cross Education Phenomenon

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Abstract

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Cross education is the strength or skill gain that is found in the contralateral, homologous limb following a unilateral training program or practice. This phenomenon provides a beneficial rehabilitation model for unilateral injuries or neurologic disorders, such as stroke. Although the cross-body transfer of strength and skill are each widely studied, they are rarely examined concurrently, despite each contributing to the goal of functional movement rehabilitation. Therefore, the overall purpose of this thesis was to examine the neuromuscular adaptations of unilateral resistive exercise training contributing to the transfer of strength and skill, while employing the necessary methodological controls that have been under-examined and under-used.

The assessment of neuromuscular mechanisms requires both voluntary and evoked contractions to be performed simultaneously. Therefore Manuscript 1 examined a novel electrode configuration, consisting of one electrode on the electrically identified motor point and the second electrode directly adjacent in a bipolar configuration. Both voluntary surface electromyography measures and evoked potentials were found to be reliable (ICCs > 0.75) and effective across multiple test sessions.

Manuscript 2 was a comprehensive review of 90 unilateral training studies in young and older able-bodied participants and in patient populations. The cross education strength gain was estimated at 18% in young, and 17% in older able-bodied participants. The cross education strength gain was 29% in patient populations consisting of post-stroke, multiple...
sclerosis, osteoarthritis, and neuromuscular disorder patients. The meta-analysis identified the efficacy of electromyostimulation (EMS) training over voluntary training modalities. The magnitude of strength transfer was similar between upper and lower and between males and females.

Lastly, manuscript 3 consisted of a 6-week unilateral training program resulting in contralateral strength gains of 11% in the wrist flexors and 15% in the dorsiflexors. A continued increase in contralateral strength at retention demonstrated the persistence of cross education following 6-weeks of detraining. Skill transfer in the contralateral limbs was evident in the force variability measures calculated during contractions without concurrent feedback (noKR). Agonist RMS amplitude, V-wave amplitude, and central activation ratio indicated neuromuscular adaptations; however, there was no change in motor unit firing rates at 60% of maximal force.
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# Table of Contents

Abstract........................................................................................................................................... ii

Acknowledgments................................................................................................................................ iv

Table of Contents.............................................................................................................................. v

List of Abbreviations ......................................................................................................................... viii

List of Tables...................................................................................................................................... ix

List of Figures................................................................................................................................... x

List of Appendices .......................................................................................................................... xi

1 INTRODUCTION................................................................................................................................. 1

1.1 Background.................................................................................................................................. 1

1.2 The Contribution of Neuromuscular Adaptations to Cross Education................................. 2

1.3 The Contribution of Motor Learning to Cross Education....................................................... 4

1.4 Significance .............................................................................................................................. 6

2 REVIEW OF LITERATURE.................................................................................................................. 8

2.1 Anatomy of the Ankle Muscles............................................................................................... 8

2.2 Anatomy of the Wrist Muscles ............................................................................................ 9

2.3 Cross Education Background............................................................................................... 9

2.4 Potential Mechanisms ........................................................................................................... 14

2.4.1 Cortical (Supraspinal) Mechanisms.................................................................................. 18

2.4.2 Spinal Mechanisms........................................................................................................... 22

2.4.3 Neural Adaptations.............................................................................................................. 26

2.4.4 Muscle Coordination.......................................................................................................... 31

2.4.5 Muscular Adaptations........................................................................................................ 34
2.5 Methodology to Examine Cross Education .......................................................... 39
  2.5.1 Unilateral Training for the Cross Education of Strength .............................. 39
  2.5.2 Unilateral Practice for the Cross Education of Skill ................................. 40
  2.5.3 Methodological Controls .............................................................................. 42
  2.5.4 Surface Electromyography ........................................................................... 43
  2.5.5 Surface Decomposition ................................................................................ 45

2.6 Conclusion ............................................................................................................. 47
References ..................................................................................................................... 48

3 PURPOSE .................................................................................................................. 66

3.1 Manuscript 1 ......................................................................................................... 66
3.2 Manuscript 2 ......................................................................................................... 67
3.3 Manuscript 3 ......................................................................................................... 67

4 MANUSCRIPT 1 ....................................................................................................... 68
  Abstract ..................................................................................................................... 69
  Introduction ............................................................................................................... 70
  Methods .................................................................................................................... 71
  Results ...................................................................................................................... 75
  Discussion ............................................................................................................... 80
  References ............................................................................................................... 86

5 MANUSCRIPT 2 ....................................................................................................... 91
  Abstract ..................................................................................................................... 92
  Introduction ............................................................................................................... 93
  Methods .................................................................................................................... 95
  Results ...................................................................................................................... 99
  Discussion ............................................................................................................... 104
  Conclusion ............................................................................................................... 107
  References ............................................................................................................... 108
6 MANUSCRIPT 3 ................................................................. 116

Abstract .................................................................................. 117
Introduction ............................................................................... 118
Methods .................................................................................. 121
Results ..................................................................................... 132
Discussion ............................................................................... 142
Conclusion ............................................................................... 148
References ............................................................................... 151

7 GENERAL DISCUSSION .......................................................... 157

7.1 Summary of Findings ......................................................... 158
7.2 Limitations ......................................................................... 165
7.3 Future Directions .............................................................. 167

APPENDIX A ............................................................................ 172

Pilot Study for Manuscript 3 ...................................................... 172

APPENDIX B ............................................................................ 178

Ethics Approval for Manuscript 1 .............................................. 178
Ethics Approval for Manuscript 3 .............................................. 179

APPENDIX C ............................................................................ 180

Sample Traces (Manuscript 3) .................................................... 180

APPENDIX D ............................................................................ 183

Training Forms and Instructions (Manuscript 3) ......................... 183
### List of Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>A/D</td>
<td>Analogue-to-digital</td>
</tr>
<tr>
<td>ANCOVA</td>
<td>Analysis of Covariance</td>
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<tr>
<td>ANOVA</td>
<td>Analysis of Variance</td>
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<tr>
<td>CAR</td>
<td>Central activation ratio</td>
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<tr>
<td>CMAP</td>
<td>Compound muscle action potential (synonymous with M-wave)</td>
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<tr>
<td>CNS</td>
<td>Central nervous system</td>
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<tr>
<td>CV</td>
<td>Coefficient of variation</td>
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<tr>
<td>dEMG</td>
<td>Decomposition EMG (Delsys®)</td>
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<tr>
<td>ECR</td>
<td>Extensor carpi radialis</td>
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<tr>
<td>EMG</td>
<td>Electromyography</td>
</tr>
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<td>EMS</td>
<td>Electrical muscle stimulation (electromyostimulation)</td>
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<tr>
<td>FCR</td>
<td>Flexor carpi radialis</td>
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<tr>
<td>GTO</td>
<td>Golgi tendon organ</td>
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<tr>
<td>H-reflex</td>
<td>Hoffmann’s reflex</td>
</tr>
<tr>
<td>ICC</td>
<td>Intraclass correlation coefficient</td>
</tr>
<tr>
<td>KR</td>
<td>Knowledge of results</td>
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<tr>
<td>M-wave</td>
<td>Compound muscle action potential (synonymous with CMAP)</td>
</tr>
<tr>
<td>M1</td>
<td>Primary motor cortex</td>
</tr>
<tr>
<td>MEP</td>
<td>Motor evoked potential</td>
</tr>
<tr>
<td>Mmax</td>
<td>Maximal compound muscle action potential</td>
</tr>
<tr>
<td>MPF</td>
<td>Mean power frequency</td>
</tr>
<tr>
<td>MRI</td>
<td>Magnetic resonance imaging</td>
</tr>
<tr>
<td>MUFR</td>
<td>Motor unit firing rate</td>
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<tr>
<td>MVC</td>
<td>Maximal voluntary contraction</td>
</tr>
<tr>
<td>noKR</td>
<td>Knowledge of results removed</td>
</tr>
<tr>
<td>PMC</td>
<td>Premotor cortex</td>
</tr>
<tr>
<td>PNF</td>
<td>Proprioceptive neuromuscular facilitation</td>
</tr>
<tr>
<td>RFD</td>
<td>Rate of force development</td>
</tr>
<tr>
<td>RMS</td>
<td>Root-mean-square</td>
</tr>
<tr>
<td>RMSE</td>
<td>Root-mean-square error</td>
</tr>
<tr>
<td>SEM</td>
<td>Standard error of the mean</td>
</tr>
<tr>
<td>sEMG</td>
<td>Surface electromyography</td>
</tr>
<tr>
<td>SMA</td>
<td>Supplementary motor cortex</td>
</tr>
<tr>
<td>TA</td>
<td>Tibialis anterior</td>
</tr>
<tr>
<td>TMS</td>
<td>Transcranial magnetic stimulation</td>
</tr>
<tr>
<td>V-wave</td>
<td>Volitional wave</td>
</tr>
<tr>
<td>V/M Ratio</td>
<td>V-wave / M-wave Ratio</td>
</tr>
</tbody>
</table>
List of Tables

Chapter 4 Manuscript 1
Table 4-1 Means and standard deviations across the 4 testing days

Chapter 5 Manuscript 2
Table 5-1 Median and range of training characteristics
Table 5-2 Effect size (standardized mean difference), percent gain, and controlled percent gain for the untrained (contralateral) limb
Table 5-3 Effect size (standardized mean difference), percent gain, and controlled percent gain for the trained (ipsilateral) limb
Table 5-4 The number of units that fall within each category: sex of the unit, the usage of familiarization, the limb involved, and the presence of a control group from the able-bodied participants (N = 125 units).

Chapter 6 Manuscript 3
Table 6-1 Participant demographics, baseline strength, and training compliance
Table 6-2 Cross education of strength and skill, and neuromuscular adaptations at post-training and retention testing in the upper limb.
Table 6-3 Cross education of strength and skill, and neuromuscular adaptations at post-training and retention testing in the lower limb.
List of Figures

<table>
<thead>
<tr>
<th>Chapter 2</th>
<th>Review of Literature</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 2-1</td>
<td>Model of cerebral plasticity</td>
</tr>
<tr>
<td>Figure 2-2</td>
<td>Schematic representation of cross education hypotheses</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 4</th>
<th>Manuscript 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 4-1</td>
<td>Bipolar electrode placement and experimental setup</td>
</tr>
<tr>
<td>Figure 4-2</td>
<td>Monopolar and bipolar sEMG recordings with cross-correlation</td>
</tr>
<tr>
<td>Figure 4-3</td>
<td>Monopolar and bipolar CMAPs</td>
</tr>
<tr>
<td>Figure 4-4</td>
<td>Intraclass correlation coefficient, standard error of measurement, and intrasubject coefficient of variation</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 5</th>
<th>Manuscript 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 5-1</td>
<td>Forest plot for the untrained (cross education) limb of the <em>young</em> group</td>
</tr>
<tr>
<td>Figure 5-2</td>
<td>Forest plot for the untrained (cross education) limb of <em>older</em> and <em>patient</em></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 6</th>
<th>Manuscript 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 6-1</td>
<td>Statistical comparisons for the training effects on the trained and untrained limbs</td>
</tr>
<tr>
<td>Figure 6-2</td>
<td>Testing protocol</td>
</tr>
<tr>
<td>Figure 6-3</td>
<td>Maximal voluntary contraction force at baseline, post-training, and retention</td>
</tr>
<tr>
<td>Figure 6-4</td>
<td>Correlation between trained and untrained strength gains</td>
</tr>
<tr>
<td>Figure 6-5</td>
<td>Agonist root-mean-square activity at baseline, post-training, and retention</td>
</tr>
<tr>
<td>Figure 6-6</td>
<td>V-wave to M-wave (V/M) ratio at baseline, post-training, and retention</td>
</tr>
<tr>
<td>Figure 6-7</td>
<td>Central activation ratio calculated from the interpolated twitch during maximal voluntary contractions at baseline, post-training, and retention</td>
</tr>
<tr>
<td>Figure 6-8</td>
<td>Force variability (variance ratio and root-mean-square-error) during ‘no feedback’ ramp contractions</td>
</tr>
</tbody>
</table>
List of Appendices

Appendix A  Reliability Study of Neuromuscular Mechanisms – Manuscript 3
Appendix B  Ethics Approval – Manuscripts 1 & 3
Appendix C  Sample Traces of V-waves and Central Activation Ratio – Manuscript 3
Appendix D  Training Information – Manuscript 3
1 INTRODUCTION

1.1 Background

Cross education is the strength gain or skill improvement that is found in the contralateral limb following unilateral training or practice on the homologous limb. Cross education was first discovered in 1984 by Scripture, Smith and Brown [1] who determined that task steadiness and muscular strength could be improved in the contralateral limb following unilateral training. Many proposed mechanisms for cross education have been examined at the cortical, spinal, neural, and muscular levels. However, the exact mechanisms for cross education and the amount that they contribute to the contralateral strength gain remains unknown. This phenomenon is of great importance for clinical applications and rehabilitation and requires further mechanistic investigation. Cross education provides a beneficial rehabilitation model for unilateral injuries or disorders, including acute injuries or immobilization (casting) of a single limb, and neurologic disorders, such as stroke, affecting the body unilaterally.

Meta-analyses have determined that the average contralateral strength gain from cross education is approximately 7-12% [2–5]. This amount corresponds to approximately 35-60% of the strength increase that is found in the ipsilateral (trained) limb [2,4,6]. A more recent review of the literature identified 90 studies that reported their methods and results with sufficient detail for analysis. The review determined that the average amount of cross education in young, able-bodied participants was an 18% increase from initial strength, corresponding to a 29% strength increase in the ipsilateral limb. The mechanisms behind cross education have been investigated using a variety of tools such as fMRI, TMS, sEMG, and EMS training, to name a few. This research has helped to identify and support
the two main hypotheses behind cross education: ‘bilateral access’ and ‘cross-activation’[7,8]. The ‘bilateral access’ hypothesis suggests that the homologous untrained muscle can access the adaptations of training from the unilateral associated motor areas. Alternatively, the ‘cross-activation’ hypothesis proposes that unilateral activity excites both ipsilateral and contralateral cortical motor areas [7,8].

1.2 The Contribution of Neuromuscular Adaptations to Cross Education

The neuromuscular adaptations that accompany contralateral force increases include some inconsistencies, likely due to wide variations in training programs and measurement methodology. Neural adaptations in the contralateral limb can be assessed using surface or needle EMG. A review of literature identified 26 studies that reported surface EMG from the contralateral agonist muscle. The average increase in contralateral agonist sEMG amplitude is approximately 32% (range from 3 to 62%) [9–25], with 9 studies finding no significant change in agonist sEMG [26–34]. Additionally, the two studies examining motor unit firing rates (MUFRs) in the contralateral agonist muscle found no significant increase following unilateral training [22,35].

Adaptations in central drive to the contralateral limb have been examined in cross education literature but not confirmed. Fimland and colleagues [10] found a 29% increase in contralateral V-wave amplitude, however, it was not a significant change likely due to a high amount of variability. Recently, Tøien and colleagues [20] found a 73% increase in contralateral V-wave amplitude in older adults following unilateral training. Alternatively, Colomer-Poveda and colleagues [30] showed no change in V-wave in either the trained or contralateral soleus following low-load training with and without blood flow resistance. Another measure of central and peripheral drive, voluntary activation, has been examined
in a few studies using twitch interpolation. Following unilateral training, Shima and colleagues [15] reported a significant increase of 3.6% in contralateral voluntary activation, Tøien and colleagues [20] reported an increase of approximately 5%, and Lee and colleagues [36] found a 2.9% increase using TMS. Alternatively, Tillin and colleagues [17] found no significant change following training. Magnus and colleagues [37] also found no significant change following unilateral limb immobilization. However, the lack of change may have demonstrated the capabilities of unilateral training to preserve an expected loss of voluntary activation during immobilization.

Previous resistance training literature has demonstrated that the increased muscle strength as well as the associated central and peripheral adaptations, are susceptible to a period of detraining [14,38–40]. It is not surprising then, that the effects of cross education have also been shown to diminish with detraining. A review of the cross education literature identified 7 studies that specifically examined the effects of cross education after a period of detraining. Although detraining reduced the training adaptations, 5 of the 7 studies found that significant amounts of ipsilateral and contralateral strength were retained following 4-12 weeks of detraining [14,15,40–44]. The five studies had a median training time of 8 weeks (range 6-10) and a median detraining time of 8 weeks (range 6-12). Following detraining, there was an average retention of 81% of the ipsilateral strength gain and 71% of the contralateral strength gain [15,40–43]. Shima and colleagues [15] was the only detraining study to record agonist sEMG and found that the gains in agonist activity were retained, but to a much greater extent in the ipsilateral limb (63% retained) than the contralateral limb (10% retained). Additionally, the 3.6% increase in voluntary activation gained in the contralateral limb completely dissipated after detraining, while the trained
limb maintained its improvement [15]. This indicates that the retention of strength in the contralateral limb is not due solely to the retention of neural adaptations, but rather suggests the presence of motor learning.

1.3 The Contribution of Motor Learning to Cross Education

A motor skill is the ability to plan and execute a movement with a certain goal [45]. Therefore, motor learning is the development, retention, and transfer of the neural connections required to complete the task [46]. Specifically, the cross education of skill (often referred to as interlateral skill transfer) is the cross-body transfer of motor learning evident in the contralateral limb following unilateral practice. Skill transfer literature primarily focuses on motor tasks (i.e., goal-oriented to a target or timed), which are practiced and tested within a single session. While the cross education of strength is defined by an increase in contralateral strength, skill transfer is defined by contralateral task improvement. This can be quantified by improvements in accuracy (lower number of errors), number of successful attempts, variability, and time to completion, to name a few. Furthermore, motor learning is measured by the ability to retain the skill after a period of detraining, and transfer the skill to another joint or a slightly different task [46,47].

As previously demonstrated for unilateral strength training, all resistive exercise includes a skill component [47–49], and therefore the potential for motor learning. To date, the presence of motor learning in the cross education of strength has been limited to the study of coactivation in the contralateral limb following unilateral training. While some studies have found no change in the antagonist amplitude in the contralateral limb following unilateral training, studies that calculated a coactivation ratio have uncovered significant improvements suggesting that motor learning may be contributing to cross
McGuire et al. [47] and Green et al. [48] also showed that motor learning of maximal effort contractions can also alter the variability of the force- and EMG-time profiles. However, in existing cross education literature the use of variability measures has not been employed to assess the motor learning of a resistive exercise task.

The study of motor learning examines the acquisition, retention, and transfer of a movement skill using various indices of learning. In motor learning literature, acquisition consists of massed practice within a short time frame (1-3 sessions) to initially learn the skill. Retention of the skill is then evaluated within a few hours to a few days and represents the ‘permanency’ of motor learning. Transfer of the skill is evaluated immediately after acquisition or coupled with retention, and consists of a slightly altered version of the practiced skill to represent the ‘generalizability’ of motor learning [51,52]. Although the retention and transfer of motor learning is typically examined within a few days of skill acquisition, cross education literature examining the effect of detraining over time has found no significant change within 4 weeks [40,43]. Therefore, the longer period of acquisition (training) may demand a longer period of detraining before the permanency of motor learning can be assessed.

The findings of Shima and colleagues [15], can provide insight into the permanency of motor learning. Participants engaged in 6 weeks of unilateral plantar flexion training, which resulted in significant increases in force, agonist EMG, and voluntary activation in the trained and untrained limbs. Following 6 weeks of detraining, the trained limb retained 60% of the gained force, 63% of the gained agonist sEMG amplitude, and the gained voluntary activation. Similarly, the untrained limb retained 40% of the gained force and most (80%) of the gained voluntary activation, but only 10% of the gained agonist sEMG
amplitude. The dissipation of agonist activity coupled with the retention of increases in force indicates a relative permanency of motor learning. The results of the untrained limb suggest that the internal model was transferred, demonstrating that motor learning of the trained limb can be generalized to the contralateral limb. What is missing from the study by Shima and colleagues, and the cross education literature in general, is the exploration of motor learning adaptations in the untrained limb.

1.4 Significance

Although primarily studied in healthy, able-bodied individuals, the cross education phenomenon has the greatest implications for clinical populations. Recently, cross education has been examined as a rehabilitation technique in clinical populations with both acute [37,53–56] and chronic [5,23,24,57–59] unilateral disorders. For orthopaedic injury resulting in immobilization or disuse (fractures, casting, etc.) cross education can provide prevention from atrophy and strength loss, and allow for pre-recovery rehabilitation [53,60]. For chronic disorders such as stroke, cross education can contribute to the restoration of bilateral functional capacity [61] and help to foster post-stroke independence. The application of cross education to clinical populations requires more in-depth knowledge of the mechanisms contributing to this phenomenon and the influence of motor learning.

The effects of motor learning on rehabilitation, especially for hemiparesis, has been mainly examined using a robotic paradigm for the arm [45,51]. Although a major component in rehabilitation is the prevention of atrophy or return of strength, this is often ignored in motor learning literature. The combined benefits of skill and strength transfer are crucial for rehabilitation involving losses of function and strength. The present research
will examine skill components of resistive exercise and its contribution to the cross education phenomenon. The cross education literature primarily focuses on the use of high intensity contractions to evoke contralateral strength gains. If, however, the main adaptation is through neural processes associated with motor learning, then such high intensity contractions may be unnecessary. Unilateral coordination training may prove equally beneficial, making rehabilitation programs more accessible for clinical populations by removing the constraints of resistive exercise training.

The present thesis is critical to furthering the work of cross education as it will employ the methodological controls necessary to distinguish between the contributions of neuromuscular mechanisms and motor learning. It is easy to recognise that inconsistencies exist in the literature since a current review has identified 90 cross education studies, but less than 31 studies have been included in the previous meta-analyses [2,4,5,62]. The lack of adequate familiarization and control groups has been detailed by Carroll and colleagues [2], which also highlighted the inconsistencies in reporting of training group and control group results. The use of surface EMG to discern neuromuscular measures has often resulted in high variability leading to no statistical significance when changes are apparent. The use of methodological controls should be imperative when comparing studies with a range of training methodologies (frequency, duration, contraction type, intensity, etc.), populations, neuromuscular measures, and research designs. However, this has not appeared to be a primary concern in the literature to date, and the field requires greater methodological controls for the mechanistic and clinical investigation of cross education.
2 REVIEW OF LITERATURE

The following section reviews each component of the three dissertation studies presented in chapters 4-6. To begin, the anatomy of the dorsiflexors and wrist flexors are detailed as the primary muscles used in manuscripts 1 and 3. Subsequently, the potential mechanisms contributing to the cross education of strength are identified, separated by the cortical, spinal, neural, coordination, and muscular adaptations. The phenomenon of skill transfer (or the cross education of skill) is then detailed, along with the potential contribution of motor learning to the cross education of strength. Lastly, this review will demonstrate the methodological inconsistencies which plague the field of cross education and initiated an updated meta-analysis (manuscript 2).

2.1 Anatomy of the Ankle Muscles

Dorsiflexion contractions were selected as the lower body activity for the present work due to the importance of the dorsiflexors in gait and posture and the common occurrence of post-stroke weakness [23,63,64]. The tibialis anterior (TA) is the primary agonist for dorsiflexion and inversion. It originates on the upper half of the lateral and anterior surface of the tibia and inserts on the medial surface of the base of the first metatarsal [65]. It is innervated by the deep peroneal nerve, accessible in the popliteal fossa and posterior to the head of the fibula. The TA is a parallel-fibre muscle composed mainly of type I slow twitch fibres [66]. The motor unit recruitment threshold of the tibialis anterior is up to 90% force [67–69]. The soleus muscle is the primary antagonist for dorsiflexion and the agonist for plantar flexion when the knee is bent at 90°. It originates on the upper portions of the posterior tibia and fibula and inserts on the into the calcaneus via the Achilles tendon [65].
2.2 Anatomy of the Wrist Muscles

Wrist flexion was selected as the focus for upper body activity because they are vital for activities of daily living, the upper limb equivalent to dorsiflexion contractions, and the joint most susceptible to post-stroke contractures [70]. The flexor carpi radialis (FCR) is the primary agonist for wrist flexion and abduction. It originates on the medial epicondyle of the humerus and inserts on the palmar base of the second and third metacarpals. It is innervated by the median nerve accessible at the cubital fossa [65]. The FCR is a bi-pennate oriented muscle composed mainly of type II fast twitch fibres which are primarily located in the superficial muscle, as identified in cats [71] and monkeys [72]. The motor unit recruitment threshold (recruitment range) of the flexor carpi radialis is up to approximately 50% force [73]. The extensor carpi radialis longus and brevis is the primary antagonist for wrist flexion and the agonist for wrist extension and abduction. It originates on the lower third and lateral epicondyle of the humerus and inserts on the dorsal base of the second and third metacarpals [65].

2.3 Cross Education Background

Cross education was first discovered in 1894 by Scripture, Smith, and Brown when examining the contralateral limb following exercises of muscular control and muscular power [1]. In 1900, Wissler and Richardson [74] first proposed the mechanism of cross education to be the diffusion of motor impulses “in the cells of the cortex or in the spinal cord.” Research in the following few decades primarily focused on the cross transfer of motor skills, rather than strength, using a motor learning paradigm [75,76]. In 1947, Hellebrandt revisited the transfer of strength and although cross education was present, it was speculated to be caused by force irradiation due to the strenuous exercises and need for
contralateral stabilization [77]. This research encouraged the methodological control of concurrent contralateral activity in following studies in order to eliminate the presence of force irradiation and further the identification of cross education [78–81].

Since these initial studies, cross-body transfer has primarily been divided into the cross education of strength (also known as cross-transfer, cross-over, or contralateral training) or the cross education of skill (also known as interlateral transfer of learning, bilateral transfer, or intermanual transfer). As these phenomena have intertwined mechanistic theories (detailed in the following section), it is the acquisition paradigm that separates them. The cross education of strength is acquired via unilateral resistive exercise over multiple sessions. Most meta-analyses have considered the cross education of strength only after a minimum 2-weeks of training [2,4,5]. Alternatively, the cross education of skill is acquired via unilateral practice of a motor task typically occurring within a single session. While this dissertation will primarily focus on the cross education of strength, it will also examine the potential for the cross education of skill using a strength paradigm.

Over the past century, cross education of strength has been demonstrated under a multitude of circumstances with a variety of training programmes (for reviews see Manca et al. [5], Hendy et al. [3], Carroll et al. [2], Munn et al. [4], and Zhou [6]). The amount of strength that is gained in the untrained, contralateral limb is highly variable due to inconsistent factors, such as methodologies, training programs, and subject populations. On average, the effect of cross education was previously estimated to be approximately 7-8%, which corresponds to 35-60% of the ipsilateral training effect [2,4,6]. A more recent meta-analysis by Manca and colleagues [5] identified 31 studies meeting their criteria for inclusion and found that the magnitude of the cross education strength gain was 12%. In a
review of 90 cross education studies (manuscript 2), the effect is estimated to be an 18% strength gain in young able-bodied participants, corresponding to approximately 67% of the gain in the trained limb. Although a greater magnitude of cross education has been identified in the upper limbs [2,4,5], a more comprehensive review of the literature (manuscript 2) identified no difference between the upper (17%) and lower (19%) limb strength gains.

Previous meta-analyses have reported that cross-education can be observed in a variety of muscles, in the upper and lower limbs, across different age-groups, and in both males and females. However, many of these factors have not been experimentally or statistically examined. To date, many studies have assumed an equality between sexes in the magnitude of cross education, often citing the review by Zhou [6], which does not compare sexes. In the literature, only two studies [82,83] included sex comparisons following unilateral training. Both studies found significant differences between sexes in the magnitude of the trained limb strength gain, but no difference in the magnitude of the untrained limb strength gain. This indicates that there is a difference in the amount of cross-body transfer (or ratio between trained and untrained limbs) between the sexes. However, Hubal and colleagues [82] found a significantly higher cross-body transfer ratio in females compared to males, while Tracy and colleagues [83] found a lower cross-body transfer ratio in females compared to males.

The magnitude of the cross education response has been linked to the ‘law of initial values’ [62,84], meaning that where there is greater ‘room for improvement’ there will be greater gains. For example, Farthing and colleagues [9,32,60] demonstrated that the magnitude of cross education was greater in the non-dominant limb following dominant
limb training than vice versa. Therefore, it is reasonable to expect varying magnitudes of strength gain due to participant factors such as age, initial training status, regular use of the movements involved in training, etc. Although cross education is primarily studied in young able-bodied participants, a selection of work has been conducted using older adults. Ehsani and colleagues [85] reported an increased transfer of strength in older adults compared to young, as may be expected since there are typically decreases in muscular strength associated with aging. These strength losses are also accompanied by decreased motor unit firing rates, agonist muscle activation, and decreased excitability of spinal and corticospinal pathways [20,86,87]. However, a review of the literature identified no difference on average in the magnitude of cross education between young (18%) and older (17%) able-bodied participants.

Cross education can also prevent the anticipated atrophy of limb immobilized via cross-body transfer. Therefore, the presence of cross education in the field of rehabilitation is widespread and of great functional importance [61]. The phenomenon of cross education lends itself to unilateral injuries or disorders where the more affected side can benefit from exercise in the less affected side. Therefore, the benefits of cross education are most abundant for clinical populations suffering from acute unilateral injuries, or chronic disorders such as stroke, multiple sclerosis, and various other neuromuscular disorders where there is typically a ‘more affected’ side. Although further work from a clinical perspective is needed, the clinical applications have influenced much of the cross education research. For example, Adamson and colleagues [88] examined the cross education of the rate of force development (RFD) following unilateral training in a healthy population. This is an important area of focus for clinical populations with unilateral gait impairment since
dorsiflexion RFD is highly linked to gait performance, which is a primary complaint in patient populations [89].

Additionally, a number of studies have examined the ‘preventative’ effects of cross education during acute injuries resulting in limb immobilization [37,54,55,90]. The effects of cross education have been shown to prevent the typical strength loss seen in an immobilized arm, and in some cases even improve the strength of the immobilized arm, when the ‘uninjured’ arm is unilaterally trained [37]. In the case of limb fractures or surgery requiring casting, cross education during the casting period has been shown to accelerate the post-cast recovery of strength when unilateral resistance training is added to a patient’s standard physiotherapy program [90,91].

The benefits of cross education have been demonstrated in clinical populations with hemiplegia [23,24,57], neuromuscular disorders [58], and osteoarthritis [59], with an average strength improvement of 29%. Dragert and Zehr [23] examined the functional impact of cross education on stroke patients. Following training of the less-affected dorsiflexors, strength gains of 31% were demonstrated in the more-affected side resulting in small improvements in clinical measures such as a 6% decrease in the ‘Timed Up-and-Go’ test and a 4% increase in walking speed. Although these functional changes were small and statistically non-significant, the authors considered them to be clinically relevant [23]. Urbin and colleagues [24] reported increased strength and range of motion following unilateral wrist extension exercises on the less affected limb of stroke patients, however, TMS measures did not demonstrate changes in corticospinal excitability or interhemispheric inhibition. Kim and colleagues [57] assessed the functional effects of cross education on acute (3-9 months) post-stroke patients using tilt table therapy.
Following single-leg standing or single-leg movements (e.g., kicking) with the less affected limb while secured to a tilt table, the patients demonstrated improvements in gait velocity, cadence, symmetry, double support periods, as well as lower limb contralateral muscle strength [57].

McCartney and colleagues [58] assessed cross education in muscular atrophy and dystrophy patients following unilateral arm exercises. Despite large variations in the 5 patients, the average strength increase in the contralateral arm was 16%, accompanying the 34% strength gain in the trained arm. Onigbinde and colleagues [59] implemented a 6-week unilateral knee extension training program on the unaffected leg of knee osteoarthritis patients. Both the affected (cross education) limb and the unaffected (trained) limb increased strength approximately 20%. The authors concluded that unilateral training in osteoarthritis patients “produced meaningful and significant increase in strength of the affected contralateral homologous group of muscles.” It is evident that cross education has the potential to improve current therapeutic interventions and greatly influence functional tasks.

2.4 Potential Mechanisms

The initial improvement in strength following a short period (<4-6 weeks) of training can be attributed to neural adaptations prior to hypertrophic gains at the muscular level [13,92]. Both Ikai and Fukunaga [92] and Moritani and DeVries [13] determined that neural adaptations were the primary contributor (>80%) to the contralateral strength gain following 6-14 weeks of unilateral training. The lack of any significant increase in cross-sectional area of the untrained limb points to cross education being a purely neural adaptation [13,40,92–94].
The continued practice of a movement can result in neural plasticity, which is the reorganization of the neural connections that control movement after a period of training or learning. The initiation of skeletal muscle activation begins in the premotor area where our behaviour is planned and a motor program is formed. This program is then transmitted to the primary motor cortex (M1). The pyramidal cells which exist in the M1 are called the upper motor neurons; these fibres descend to the pyramidal tract and form the corticospinal tract where 85-90% decussate to the contralateral side of the body. The upper motor neurons which decussate at the pyramidal tract synapse with lower motor neurons whose axons then innervate the skeletal muscles.

The 10-15% of pyramidal tract neurons that do not decussate at the pyramidal tract have been suggested to be a small contributor to cross education through the excitation of ipsilateral pathways and the diffusion of central drive [3,12,26]. However, these neurons primarily innervate the trunk and are absent below the cervical enlargement suggesting that any contribution to cross education would be present only in the upper limb [95]. Therefore, this is not considered to be one of the primary mechanisms of cross education.

The control of skeletal muscle is monitored and regulated by the basal nuclei and the cerebellum. The basal nuclei are responsible for muscle control via a feedback loop between the cerebral cortex and thalamus. While it monitors all movement, it is the primary control for automatic movements that have been ingrained, such as writing, typing, or tying a shoe [96]. Similarly, the cerebellum plays a crucial role in movement control and coordination through error detection and correction. For this reason, the cerebellum is important for motor learning and skill acquisition by coordinating the muscles and joints involved in the movement task [96].
Unilateral resistance exercise has been shown to involve bilateral activation in the primary motor cortex (M1), premotor cortex (PMC), and supplementary motor area (SMA) [9,97,98]. For motor learning tasks the model constructed by Doyon and Ungerleider [99] and subsequently revised [100–102] states that the regions of neural activation associated with motor learning depends on the type of task and the stage of learning. The two main classes of motor learning: motor sequence learning consisting of fine motor tasks, and motor adaptation consisting of environmental or gross motor tasks; are dependent on different areas of the brain. Additionally, the regions of neural activation are dependent on the stage of learning from acquisition to retention. In Figure 2-1 reprinted from Doyon and Benali [102] the progress of neural activation over the motor learning process is detailed for each type of motor learning.
2.4.1 **Cortical (Supraspinal) Mechanisms**

Previous reviews of cross education mechanisms have proposed that cross education can primarily be explained by two distinct, but not necessarily mutually exclusive, hypotheses (see Figure 2-2) [2,7,8]. The first hypothesis focuses on the adaptations occurring in the associated contralateral motor areas as a result of unilateral training. This has been termed ‘bilateral access’, or ‘callosal access’ in motor learning literature, and suggests that the homologous untrained muscle can access the unilateral adaptations of training occurring in the trained primary motor cortex and associated motor areas [7,8,103]. Or similarly, that the untrained limb can access the unilateral adaptations of practice (motor schema) occurring in the supplementary motor area [104,105]. This hypothesis is primarily associated with the cross education of skill [103,106]. The second hypothesis focuses on motor pathways and the bilateral activation of neural circuits. This hypothesis has been termed ‘cross-activation’ and proposes that unilateral activity excites both ipsilateral and contralateral cortical motor areas, which can lead to increased drive not only to the trained muscle, but also to the homologous untrained muscles [7,8,103]. This hypothesis is primarily associated with the cross education of strength [103,106].
Lee and colleagues [103] describe the site of neural adaptation in Figure 2-2A (left panel) as “cortical, or sub-cortical, motor areas that project bilaterally.” This refers to adaptations which occur unilaterally in the trained hemisphere but have bilateral projections. The most likely sites of this adaptation are the primary motor cortex (M1), supplementary motor area, and premotor cortex, which have “sparse and weak” ipsilateral projections both transcortical (cortical) and via the corticospinal tract (subcortical) [107]. A second potential site of adaptation with bilateral projection is the Rexed laminae of the spinal cord. The “intermediate gray matter” (lamina VII and VIII) coordinates ipsilateral
and contralateral motorneurons via interneurons projecting bilaterally [108,109]. The use of fMRI to examine the human cervical spinal cord during a unilateral hand-closing task with forearm contraction resulted in consistent ipsilateral activation, as expected, and noticeable but inconsistent (~50% occurrence) contralateral activation [109]. These results provide evidence to the bilateral projection component of the “bilateral access” hypothesis, albeit inconsistent and ‘weak’ in magnitude.

The interhemispheric component of the “bilateral access” hypothesis (i.e., callosal access) has been long examined in the cross education literature. The diffusion of motor impulses was first suggested in 1900 by Wissler and Richardson [74], and later reiterated by Hellebrandt and colleagues [77], opening the door to the examination of supraspinal adaptations in cross education. Recent investigations using fMRI and transcranial magnetic stimulation (TMS) suggest that there are multiple cortical adaptations contributing to cross education. The diffusion of motor impulses through interhemispheric paths has been further examined with the inference that a decrease in interhemispheric inhibition is present with unilateral training [7,25,110–112].

To directly examine the horizontal connections of pyramidal cells (i.e., interhemispheric communication), Rioult-Pedotti and colleagues [113] trained rats to reach for food using only one forelimb for a period of one hour over 1-5 days. Coronal brain slices were taken from the rats at 24-45 hours post-training. Stimulation of the ‘trained’ M1 was recorded in the ‘untrained’ M1 of the training group and a control group. An increased amplitude of contralateral recordings demonstrated the increased efficacy of the horizontal connections following unilateral motor practice [113]. This evidence of interhemispheric communication and reductions in interhemispheric inhibition following unilateral training
provides evidence to support the bilateral (callosal) access hypothesis. The diffusion of activation, or decrease in interhemispheric inhibition, has been shown to be linked to the intensity of the resistive exercise [114,115]. This finding highlights the high-intensity requirement of training in the pursuit of cross education. The decrease in interhemispheric inhibition with unilateral resistive exercise has been found acutely (during simultaneously performed contractions) [114–116], and chronically following unilateral strength training [25], and with varied results following unilateral strength training in a post-stroke population [24].

Lastly, the “cross-activation” hypothesis is supported by neuroimaging work observing bilateral activation of the sensorimotor cortex during unilateral activity of the upper [117] and [118] lower limbs. Recent work has demonstrated plasticity of the primary motor cortex (M1) associated with contralateral strength gains. Farthing and colleagues have used fMRI to examine cortical adaptations following strength training alone [9] and in conjunction with limb immobilization [53]. Cross education was present as an increase in strength [9] and as a prevention of strength loss [53] compared to control groups. Functional MRI revealed increased activation in the sensorimotor cortex (M1 and S1) of the ‘untrained’ hemisphere [9,53]. Similarly, Pearce and colleagues [55] used TMS stimulation before and after limb immobilization and found that unilateral training maintained motor evoked potential (MEP) amplitude in the untrained, immobilized limb, providing further evidence of an adaptation in the primary motor cortex. The findings presented in this section demonstrate the cortical and subcortical adaptations contributing to cross education and outline the simultaneous occurrence of the previously mentioned hypotheses.
2.4.2 Spinal Mechanisms

The main argument for the contribution of a spinal mechanism is demonstrated by the enhanced contralateral strength gains caused by electrical stimulation training [6,12,119,120] and blood flow resistance training [30]. Zhou and colleagues [18] found that voluntary isometric resistance training and electromyostimulation (EMS) over four weeks produced equal strength gains (21%) in the contralateral limb. Hortobágyi and colleagues [12] compared voluntary and EMS training in the quadriceps over 6 weeks. Both training groups completed voluntary and EMS pre- and post-testing to examine the specificity of training effects. The voluntary training group showed contralateral gains after 6 weeks for voluntary contractions (19% increase) and EMS contractions (27% increase). However, the EMS training group had much larger gains than the voluntary training group in both voluntary contractions (38% increase) and EMS contractions (85% increase). Similarly, Bezerra and colleagues [119] demonstrated enhanced cross education strength gains when training with electrical stimulation added to voluntary contractions (28% increase), as compared to voluntary contractions alone (9% increase).

Hortobágyi and colleagues [12] suggested that Group II afferents are activated during EMS and the stimulation causes a muscle stretch, which excites the homologous muscles through a medium latency reflex. This was also suggested by Bezerra and colleagues [119] who postulated that the afferent inputs from electrical stimulation are the source of CNS adaptations extending to the contralateral limb. Additionally, the completeness of muscle fibre activation with electrical stimulation as compared to voluntary contractions and the preferential recruitment of high threshold motor units, have both been suggested as mechanisms contributing to CE from electrical stimulation training [119–121]. Further
support for spinal adaptations is the increased contralateral strength gain caused by blood flow resistance during voluntary training [30]. Colomer-Poveda and colleagues [30] postulated that activation of Group III and IV afferents contributed to increased excitability at the spinal and supraspinal levels.

To further assess spinal mechanisms, the Hoffman Reflex (H-reflex) can be evaluated as a measure of spinal excitability. Specifically, the H-reflex gives a measure of $\alpha$-motorneuron excitability and the presynaptic inhibition of interneuronal circuits [122,123]. It is this inhibition that contributes to the “production of muscle synergy or movement pattern”, which may be augmented during training to better perform a task [123]. Therefore, the H-reflex can be used as a tool to assess the presence of spinal mechanisms contributing to cross education through the alteration of spinal inhibitory circuit in the ipsilateral and contralateral limbs. Previous work examining bilateral exercise has shown that strength training augments H-reflexes when they are elicited during a voluntary contraction [122,124,125]. Alternatively, H-reflexes elicited at rest are typically not affected by training [122,124,125]. Taube and colleagues [125] found that 6 weeks of strength training increased the H-reflex whereas balance training decreased the H-reflex. The authors suggested that strength training requires an enhanced motorneuron output and enhanced excitatory drive for explosive movements whereas balance or precision tasks would benefit from a diminished excitatory drive [125].

The examination of H-reflexes on the contralateral limb during a unilateral training program is limited. To date, six studies have examined the effect of cross education on H-reflexes with the majority finding no increase in the untrained limb [10,20,30,50,126,127]. Hortobágyi and colleagues [128] originally examined H-reflexes in the contralateral
forearm while the ipsilateral limb performed voluntary wrist flexion and extension contractions. Although not a training study, this work examined the real-time effect of ipsilateral stimulation on contralateral spinal excitability. There was a 22% and 50% reduction in the contralateral FCR H-reflex amplitude during ipsilateral wrist flexion at 50% and 75% MVC, respectively. Interestingly, wrist extension contractions at 75% MVC also diminished H-reflex amplitude, but not to the same extent (33% reduction). The authors suggested that the reduction in the H-reflex was caused by “a strong presynaptic inhibition of Ia afferents brought about by descending activity” [128]. The majority of cross education studies using the H-reflex found no change in the contralateral limb following a unilateral training program regardless of the H-reflex alteration in the ipsilateral limb [10,20,30,50,127]. This suggests that the contralateral spinal effects of ipsilateral activity may be a simultaneous (immediate) phenomenon rather than a persistent (trained) one.

All muscle activity initiated from the cortical motor areas travels to the spinal cord and is modulated by the excitability of α-motorneurons. Therefore, the alteration of spinal reflexive loops with strength training can alter the motor unit recruitment and discharge rate, as well as muscle coordination, via α-motorneurons. The spinal reflex loops present during resistive exercise includes golgi tendon organs of the agonist muscle, muscle spindles of the antagonist muscle, reciprocal inhibition via Ia inhibitory interneurons, recurrent inhibition via Renshaw cells, and presynaptic inhibition modulated by descending drive and afferent input. Furthermore, the presence of commissural interneurons indicates that alterations in spinal reflex loops on the ipsilateral (trained) side may be projected to contralateral α-motorneurons and inhibitory interneurons, thereby altering muscle
coordination in the contralateral (untrained) limb. Although spinal reflex loops are difficult to test and nearly impossible to distinguish, limited research has examined their contributions to cross education. Previous research has also examined the commissural interneurons in the brain stem and spinal cord in cross education. Delwaide and Pepin [129] found bilateral effects of unilateral stimulation in hemiplegic patients and speculated that commissural interneurons were responsible for cross-spinal excitatory potentials.

Reciprocal inhibition is the inhibitory effect that an antagonist muscle has on the agonist motorneuron [7]. This process consists of a web of inhibitory and facilitatory interneurons controlling movement, primarily mediated by Ia afferents and Renshaw cells [7]. Delwaide and colleagues [129,130] have examined the effects of reciprocal inhibition on the contralateral limb during ipsilateral limb movement and stimulation. The authors found that the contralateral limb H-reflex was diminished (28-33% inhibition) during passive movement, active movement, and nerve stimulation of the ipsilateral limb. The authors concluded that contralateral afferents have an ipsilateral influence via Ia interneurons crossing the midline [129].

Presynaptic inhibition is an inhibitory effect on Ia afferents that is modulated by corticospinal inputs. Although both spinal and supraspinal inputs can excite inhibitory interneurons to presynaptically affect Ia afferents, the use of conditioned H-reflexes or TMS evoked MEPs may be able to provide some differentiation [131,132]. Adaptations in presynaptic inhibition have been found as a result of motor learning [133]. As well, subcortical (spinal) modulation of presynaptic inhibition has been found to alter MEP facilitation cross-body with unilateral muscle activation [131]. Therefore, the potential for cross-body adaptations in presynaptic inhibition to be a contributor to cross education, as
suggested by [110,128], is conceivable. In fact, Lagerquist and colleagues [50] attributed an increase in H-reflex amplitude, taken on the ascending portion of a recruitment curve, to the reduction in presynaptic inhibition acting on low threshold motor units. Although more research is needed in this area, it is very likely that the cross-body adaptations of spinal reflexes and inhibitory loops are potential contributors to cross education.

2.4.3 Neural Adaptations

The neural adaptations to resistance training may be cortical or spinal, but that distinction cannot always be elucidated. Typical neural adaptations to training include increased descending (central) drive to the muscle causing increased motor unit recruitment and/or firing rates, decreased antagonist co-activation, and an increased incidence of doublets mainly affecting the rate of force development [13,122,124,134]. In cross education literature one of the primary adaptations in the contralateral, homologous muscle is an increase in central drive. This increase could be explained by either of the two cortical hypotheses (detailed in section 2.4.1), or by the adaptations in the spinal cord transferred to the contralateral limb via commissural interneurons (detailed in section 2.4.2). Most likely, no one explanation is mutually exclusive, but rather a combination of adaptations contributes to increased contralateral agonist muscle activation.

The amplitude of muscle activity is frequently used in cross education literature to reflect the amount of neural drive to the contralateral muscles. Changes in EMG amplitude following resistance training is equivocal with some studies reporting an increase while many other report no significant change (for a review see Griffin and Cafarelli [135]. Therefore, it’s not surprising that contralateral adaptations in muscle activity are also equivocal in the cross education literature. The mean or median power frequency of the
signal is also used in conjunction with the amplitude measures to provide global information about changes in motor unit activity patterns [136]. Even though specific control strategies (i.e., firing rate or recruitment) cannot be inferred from these measures, they can be used to support motor unit behaviour results. To date, it appears that no study has reported sEMG power spectrum analysis data for the contralateral limb following resistance training.

The reporting of muscle activity amplitude in the contralateral homologous muscle in cross education literature is abundant; however, methodological differences make it difficult to compare findings across studies. Additionally, results are often reported with use of graphs, rather than raw data, from which means and standard deviations must be estimated. Of the 90 studies reviewed, 25 measured and reported adaptations in muscle activity amplitude in the homologous, contralateral muscle. Nine studies found non-significant changes after training, while 16 studies reported an increase in agonist EMG amplitude averaging 28% (range 3–58%). It is unknown whether the increase in contralateral muscle activity is due to increased activation of cortical motor areas, increased agonist α-motorneuron excitability, or alterations in spinal reflex loops affecting the agonist α-motorneurons.

Following bilateral resistance training, motor unit firing rates have been found to increase [137–140] or remain consistent [141,142] in the trained limb. To date, two studies have investigated the ‘cross education of motor unit firing rates’ in the contralateral limb following unilateral training. Rich and Cafarelli [22] examined the effects of an 8-week unilateral knee extensor training program on strength and motor unit firing rates. Although strength gains of 36% occurred in the trained limb, strength gains in the untrained,
contralateral limb (~5% increase, figure estimation) were not significant. Motor unit firing rates from contractions at 50% MVC also showed no significant change in either limb following training despite an increase in twitch rate of force development [22]. Patten, Kamen, and Rowland [35] examined the effects of a 6-week unilateral fifth finger abduction on strength and motor unit firing rates in young and older adults. Cross education was not the focus of this study; however, the contralateral, untrained limb was used as a control and alterations were seen in both limbs. For the young adults, strength increased 25% in the trained hand and 19% in the untrained hand. In both the trained and untrained hands, maximal MUFR significantly increased within 48 hours (prior to training); however, the MUFR of the untrained limb returned to baseline and showed no significant change following training. The authors speculated that the initial increase, and subsequent decrease, in MUFR might have been caused by changes in MU recruitment or antagonist coactivation. The similar pattern of change in MUFR between the trained and untrained hand, led the authors to suggest that similar mechanisms were responsible for both hands [35].

To further elucidate the cortical versus spinal origins of central drive alterations, peripheral nerve or transcranial stimulations can be used. Measures such as the V-wave, voluntary activation via twitch interpolation, TMS evoked MEPs, and fMRI cortical activity investigations have attempted to clarify the origination of augmented contralateral descending drive. The use of supramaximal nerve stimulation during a MVC (twitch interpolation) can be used to assess the “completeness of skeletal muscle activation” [143–145]. This is accomplished by comparing the twitch interpolation force to either (1) a subsequent potentiated twitch (voluntary activation), or (2) the maximal voluntary force
(central activation ratio). Twitch interpolation via nerve stimulation and TMS has demonstrated significant increases (~3-5%) in the level of activation of the contralateral limb [15,20,36]. Alternatively, Tillin et al. [17] examined the effects of 4 weeks of knee extension training on maximal explosive force and voluntary activation. Despite increases in maximal explosive force for both the trained (+20%) and untrained (+8%) limbs, there was no change in voluntary activation for either limb [17]. However, Tillin et al. [17] used the relationship between the twitch ratio and voluntary force to extrapolate what they termed “theoretical maximum force”. This may have accounted for the lack of any change in voluntary activation since explosive maximal force and ‘theoretical maximal force’ changed proportionally following the training program [17]. Additionally, Magnus and colleagues [37] examined the effect of unilateral training during immobilization of the contralateral limb. The experiment included 4-weeks of unilateral elbow flexion training, while the contralateral untrained arm was unweighted using a shoulder sling. Unilateral training was able to prevent losses in strength and muscle thickness in the immobilized arm and maintained voluntary activation in the contralateral arm [37].

Supramaximal nerve stimulation to elicit the interpolated twitch also produces antidromic propagation of action potentials, which results in the appearance of the V-wave in the sEMG activity of the agonist muscle. The V-wave may be used as a measure of descending central drive from supraspinal centers to the spinal motoneuron pool [146]. As electrical stimulation is progressively increased from low levels, there is a concomitant increase in antidromic propagation of motor action potentials from the cathode site that block/collide with the action potentials descending from Ia recruitment. Maximal voluntary contractions allow the descending action potentials from Ia recruitment to
continue with the efferent flow. This is expressed as a V-wave, with the same latency as the H-reflex, during a voluntary contraction. Greater descending central drive results in more collision and a greater V-wave peak-to-peak amplitude [10,122,147].

Increases in the ipsilateral V-wave amplitude following a training program have been well documented, demonstrating an increase in the central drive to the muscles with training [122,148–150]. However, only three studies have examined the V-wave in the contralateral limb following a unilateral resistance training program. Fimland et al. (2009) examined the ipsilateral and contralateral legs following 4 weeks of plantar flexion strength training. Cross education was apparent with a 32% increase in the untrained leg strength, accompanied by a 44% increase in the trained leg. After normalizing the V-wave to the simultaneously elicited M-wave (both superimposed on MVCs) there was a significant 109% increase in the trained leg V/M ratio and a non-significant 29% increase in the untrained leg. Although the contralateral increase was non-significant compared to pre-training values, it was significantly greater than the control group’s slight decrease in V/M ratio. This discrepancy is most likely due to the high level of variability in this measure.

Alternatively, Colomer-Poveda [30] found no significant change in the V-wave amplitude of the trained or untrained legs following 4 weeks of plantar flexion training despite significant increases in contralateral force.

Additionally, Lapole and colleagues [127] examined contralateral V-wave amplitude following 14 days of daily unilateral Achilles vibration. The unilateral vibration did produce plantar flexion strength increases in both the vibrated (+9.1%) and non-vibrated (+10.2%) legs. This was accompanied by increases in the V-waves of 43.3% in the vibrated soleus, and 41.6% in the non-vibrated soleus [127]. Most recently, Tøien and colleagues
[20] found a significant 75% increase in V-wave amplitude in older adults following unilateral training, identifying a ‘neural drive enhancement’ in the contralateral limb. It is evident that more research is needed to determine the effect of unilateral training on the contralateral V-wave, but these initial studies, and the twitch interpolation evidence, strongly suggests that bilateral increases in efferent drive may be contributing to cross education.

Transcranial magnetic stimulation (TMS) is a measurement tool used in cross education research to assess corticospinal excitability. Unilateral training has been shown to increase in corticospinal excitability of the untrained M1, as demonstrated by increases in motor evoked potential (MEP) amplitude or decreases in stimulation threshold [25,26,103,111,151]. Additionally, decreases in intracortical inhibition have been demonstrated in the untrained hemisphere via decreases in short intracortical inhibition (SICI) or silent period duration [33,111,151]. These results primarily support the cross-activation hypothesis as a mechanism of cross education. However, since MEPs are evoked at the cortical M1 and traverse the corticospinal pathways before being recorded via sEMG at the muscle, the alterations in MEP amplitude could potentially be caused by cortical, spinal, or muscular adaptations.

2.4.4 Muscle Coordination

During any movement antagonist co-activation is present to stabilize and protect the joint. However, this means that it is also opposing the movement goal. The balance between force production and joint protection may be adaptable, but it is unknown which the CNS will prioritize. There are both supraspinal and spinal contributions to antagonist co-activation, as well as biomechanical contributors. It is likely that no one adaptation of
antagonist co-activation mechanisms is mutually exclusive. A decrease in antagonist muscle activity is a common adaptation with resistance training. This may be an alteration in the amount of descending drive to the antagonist muscle or an alteration in the spinal reflex loops creating antagonist activity in response to afferent inputs. A limited number of cross education studies have examined contralateral antagonist sEMG and most reported no significant changes in activation of the contralateral limb [9–11,23,32]. However, although these studies found no significant reduction in antagonist activity, the agonist activity was found to increase an average of 38% indicating that there was less relative opposition to force production from the antagonist muscle. Alternatively, Carolan and Cafarelli [31] found a 13% reduction in contralateral antagonist coactivation when calculated as a percent of the muscle’s maximum activation. This was accompanied by a cross education strength gain of 16%. Lagerquist and colleagues [50] also examined coactivation following unilateral resistance training. Antagonist activity was not measured during maximal force contractions but rather a coactivation ratio was calculated during a low-level (10% MVC) contraction. Following training, the ratio increased over 300% in both legs demonstrating increased agonist activity relative to antagonist activity [50].

The decrease in antagonist central drive seen with training may contribute to cross education strength gains via changes in interhemispheric inhibition supporting either the bilateral access or cross-activation hypotheses. Dimitrijevic and colleagues [152] examined the activity of the ipsilateral and contralateral TA, soleus, and quad muscles during a prolonged submaximal dorsiflexion contraction. They found that there was a consistent pattern of co-activation, in both legs, across subjects. The consistency of the amount of activity and timing (i.e., when co-activation began for each muscle) of the co-activation led
the authors to suggest that a cortical motor pattern generator was responsible for co-activation. Lévénez and colleagues (2005) found similar results during a submaximal fatiguing contraction. The agonist and antagonist activity changed linearly, but H-reflexes recorded intermittently during the contraction changed in a biphasic pattern. The authors concluded that co-activation was not due to spinal adaptations (since it did not match the H-reflex changes) and therefore the linear increase was a supraspinal adaptation.

This being said, there are numerous spinal reflex loops, which undoubtedly contribute to antagonist co-activation via α-motorneuron excitation or inhibition. Presynaptic inhibition is controlled by both descending drive and agonist Ia afferents via excitation of a inhibitory interneuron, which then inhibits the antagonist α-motorneuron. Reciprocal inhibition also minimizes antagonist activation via Ia inhibitory interneurons excited by the agonist Ia afferent. Alternatively, recurrent inhibition can increase antagonist activation. Activation of Renshaw Cells inhibits the Ia inhibitory interneuron thereby increasing antagonist α-motorneuron excitability, and also inhibits the agonist α-motorneuron directly. Biomechanically, golgi tendon organs (GTOs) and muscle spindles can also affect antagonist co-activation. Agonist muscle GTOs respond to an increasing load by inhibiting the agonist α-motorneuron (via inhibitory interneurons) and exciting the antagonist α-motorneuron in an effort to prevent overload of the agonist muscle. Similarly, muscle spindles of the antagonist muscle respond to a quick stretch or extreme end range of motion by exciting the antagonist α-motorneuron to contract, and inhibiting the agonist α-motorneuron in an effort to prevent overstretch of the antagonist muscle. The adaptation of these spinal reflex loops may affect the contralateral limb via commissural interneurons leading to an increase in cross education.
One of the confounding factors related to the measurement of muscle coordination is the variability in how the measures are calculated and reported. A common co-activation measure is the normalization of antagonist activity to the maximal activity that the muscle can produce [31]. For example, the soleus is the antagonist muscle during dorsiflexion contractions, therefore the activity that the soleus produces as an antagonist would then be normalized to the maximal amount of activity it can produce during plantar flexion. However, Aagaard and colleagues [154] caution that this is only appropriate if the two contractions types are equal. That is, during a concentric dorsiflexion contraction the soleus (antagonist) is contracting eccentrically. Therefore, it should be normalized to its maximal activity as an agonist during an eccentric plantar flexion contraction. The use of isometric contractions can help mitigate this issue. While this normalization is common, Billot and colleagues [155] demonstrated that it is an underestimation of force opposition and can only be a valid representation of force opposition if the muscle size is accounted for. Although this method may result in a force underestimation, the relative change in opposition would still be valid in a repeated testing design.

### 2.4.5 Muscular Adaptations

Strength gains, as a product of resistance training, are a function of both neural and muscular adaptations, since strength is a product of the muscle fibres available and the activation of those fibres [134]. Previous research has demonstrated that the early strength gains (<4 weeks) are due to neural factors with evidence of hypertrophy beginning around 6-8 weeks of training [13,156]. The muscular adaptations to resistive exercise can include muscle hypertrophy, increased contractile proteins, an increase in type IIA fibres, vascular adaptation, and possible changes in enzyme and hormone levels [134]. Zoeller and
colleagues [157] examined the effects of artery diameter and muscle cross-sectional area bilaterally. Following 12 weeks of unilateral training bilateral strength increases were present indicating cross education, however vascular and hypertrophic changes occurred only in the trained arm, which is in line with previous researching examining contralateral limb cross-sectional area.

To date, there is a lack of muscular adaptations seen in the contralateral limb following unilateral strength training. It is unlikely that muscular adaptations will contribute to a contralateral strength gain since they are highly localized. Therefore, muscular adaptations are not considered to be a contributor to cross education, with a large body of primary research and meta-analyses supporting this conclusion.

2.4.6 Motor Learning

The cross education of skill is the cross-body transfer of motor learning following practice of a motor task. There are a number of terms used to describe improvements in contralateral task variability following unilateral practice such as, interlateral transfer of learning, bilateral transfer, or intermanual transfer. In the following sections this phenomenon will be referred to as the cross education of skill, or skill transfer, to simplify the terminology. The mechanistic theory behind the cross education of skill is akin to that of the cross education strength, with the distinct difference being the parameters of ‘acquisition’. Cross education of strength employs unilateral training programs lasting a minimum of 2-weeks for the purpose of ‘acquiring’ strength bilaterally. Alternatively, cross education of skill typically looks at unilateral motor skill acquisition within 1-2 sessions consisting of hundreds of practice trials for the purpose of learning a motor task.
bilaterally. Although these two phenomena appear different from the surface, the mechanisms behind the transfer are very similar.

The two main hypotheses leading the phenomenon of skill transfer are the ‘callosal access’ and ‘cross-activation’ of motor learning engrams. The callosal access model suggests that when learning a task an engram is stored in the dominant hemisphere, which allows the dominant limb direct access and the non-dominant limb indirect access to the adaptations [158,159]. This theory may support the preferential transfer from non-dominant to dominant limb. The cross-activation model suggests that when learning a task an engram is stored in the contralateral (associated) hemisphere and a weaker engram is also stored in the ipsilateral hemisphere [160]. This theory supports the preferential transfer from dominant to non-dominant limb. These hypotheses were developed in an attempt to explain the asymmetry of transfer between dominant and non-dominant limbs. However, they were ‘updated’ by Sainburg and Wang [161] who proposed a modified access model which hypothesizes that an engram stored in the contralateral hemisphere with learning can be accessed by the ipsilateral hemisphere (contralateral limb) regardless of lateral dominancy. It is evident that the cross education of strength and skill are one in the same with differing terminology and training (practice) paradigms.

The dynamic dominance theory [162] suggests that each hemisphere has specific responsibilities for movement, which can help explain asymmetrical transfer. For example, the dominant limb has been shown to be better suited to control trajectory and movement direction, whereas the non-dominant limb has been proven better at controlling endpoint location and movement outcomes [161,163]. Additionally, it has been proposed that the contralateral transfer during complex tasks relies more on the callosal access model,
whereas *simple* tasks rely more on the cross-activation model, while still recognizing that these are not necessarily mutually exclusive [103]. Just like resistance training, unilateral skill training with the dominant versus non-dominant side for the optimization of skill transfer has shown equivocal results [164,165]. However, Imamizu and Shimojo [164] have suggested that the initial movement control deficit in the non-dominant arm, akin to the strength deficit in the non-dominant limb, can account for the inconsistency in the literature regarding direction of transfer.

Skill transfer has been identified in both simple and complex tasks, in the upper and lower limbs, and for spatial, temporal, or force accuracy. The evaluation of skill transfer and variability may differ depending on the task being practiced or tested. Common measures to assess adaptations of a *fine* motor control task often include time to completion, accuracy, number of errors, and calculation of extraneous movements. Alternatively, common measures to assess adaptations of a *gross* motor control task often include muscle coordination and variability or reproducibility of the criterion measures (e.g. force) as assessed by RMS error or a variance ratio.

In resistance training literature, the improvement of task variability exists in the trained limb following single-session and multi-session training [48,49,166]. Since the effects of motor learning are relatively immediate, most research examines skill transfer within a single session. Skill transfer has been examined in a variety of tasks including drawing or printing [76,158], braille recognition [167], pointing [164,165], fine force control [165,168], and ballistic finger abduction [103], to name a few. The change in variability, specifically, in the trained and untrained limbs following multi-session training
has been examined using task-specific training to complete an “occupationally embedded task” [169] and generalized training to improve task steadiness [170].

Nagel and Rice [169] had participants complete a toy maze while assessing the time to completion and force oscillations (or smoothness). Participants practiced the maze with one hand, every day for seven days. Post-testing on day 8 showed that the task-specific training resulted in decreases in time to completion and force oscillations for both the trained and untrained hands [169]. Alternatively, Keogh and colleagues [170] demonstrated that generalized training, that is not task-specific, can also lead to improvements in the variability of task performance. This was accomplished by having older adults (males aged 70-80) complete 4 pointing tasks during which the tremor of the index finger was assessed. A coactivation ratio was calculated using the flexor digitorum superficialis and extensor digitorum to determine the muscle coordination used to point the index finger and ‘resist’ tremor. The generalized training program consisted of 6-weeks of biceps curls, wrist flexion, and wrist extension contractions. Participants were separated into two groups, completing either strength training using low reps and high weight, or coordination training using a quasi-random joint angle trajectory.

Keogh and colleagues [170] reported that the effectiveness of the coordination training was evident as a decrease in RMS error from the beginning of training to the end of training for the three contraction types. Following the 6-week training protocol, there was an increase in strength for trained and untrained arms of both training groups in each movement tested, i.e., wrist flexion, wrist extension, and elbow flexion. The coactivation index significantly decreased for both groups in the trained limb only. Lastly, the coordination group had reduced finger tremor in both the trained and untrained hands,
whereas the strength training group reduced tremor in the trained hand only [170]. These results suggest that an element of force control is required during the training protocol if the goal is a contralateral reduction in task variability. However, this work may have been flawed by the selection of the muscles involved in strength training, since the pointing task would primarily involve the shoulder muscles, rather than those of the elbow or wrist. Nonetheless, Keogh and colleagues have re-introduced a valid research question; is the cross education of skill present following a strength training paradigm?

2.5 Methodology to Examine Cross Education

The primary gap in cross education literature is the use of appropriate methodological controls for the determination of contributing mechanisms. This is best evidenced by small number of studies included in previous cross education meta-analyses. The defined inclusion criteria limited the previous meta-analyses to 8 studies in Zhou [6], 13 studies in Munn et al. [4], 16 studies in Carroll et al. [2], 10 studies in Cirer-Sastre et al. [62], and 31 studies in Manca et al. [5]; compared with the 90 studies that a current review uncovered. This discrepancy demonstrates the methodological inconsistencies present in the cross education literature. Detailed in the following sections are the common methodologies and inconsistencies in training paradigms, methodological controls such as familiarization and control groups, the reporting of cross education results, and the use of electromyography for the examination of neural adaptations.

2.5.1 Unilateral Training for the Cross Education of Strength

The effects of training methodologies on the magnitude of cross education have been reviewed in meta-analyses [2–4,6,62]. The greater the training intensity, the greater the magnitude of cross education, with a minimum intensity of approximately 60% maximal
force being required to produce contralateral strength gains [2,6]. However, many studies employ training repetitions of maximal contractions (100% MVC), which may produce large contralateral strength gains, but is impractical for many rehabilitation settings. Of the 90 studies examined in the meta-analysis (manuscript 2), almost half used maximal contractions for their strength-training protocol, with many others using progressive resistance training to maximal force or multiple repetition maximum testing (e.g., 5-RM). Of the 5 studies examining patient populations 2 used maximal contractions, 1 used progressive resistance, 1 used 80% maximal strength, and 1 did not report. This indicates that high-intensity contractions are possible in patient populations, and that the training paradigms of able-bodied participants accurately reflects the training performed in the rehabilitation field.

The specifics of unilateral training programs vary greatly across cross education literature. The presence of cross education has been seen following training programs lasting a minimum of 2 weeks, ranging from 3 times per week to daily training, and across a wide range of reps and sets performed per session. Isometric, isokinetic, and dynamic contractions have been the most widely used, typically at maximal or near-maximal force, with a few studies examining electrical stimulation training. In studies examining multiple forms of training or testing modalities (e.g., contraction type, speed of isokinetic contraction, etc.), it is typically found that the greatest strength gain occurs in the trained modality, with smaller gains generalizing to alternative modalities [11,82,111,171].

2.5.2 Unilateral Practice for the Cross Education of Skill

Just as the specifics of a strength-training program can affect cross education, the factors associated with task acquisition, and the measurement of retention and transfer, can
affect skill transfer of motor learning. Task acquisition for the purpose of learning, rather than immediate performance, has been studied and refined over the past century. Many of the acquisition factors are generally agreed upon to promote learning and can be easily summarized. For instance, the duration of practice is linked to the amount of motor learning [172,173] since cortical adaptations are affected by the amount of practice [174,175]. The frequency of practice favours distribution for learning and massed session for performance [172]. Similarly, the order of practice favours randomization for learning due to the benefits of contextual interference and blocked trials for performance [173]. An external focus of attention proves superior to an internal focus of attention due to the promotion of motor automaticity [176,177]. The difficulty of the task should evoke a certain level of necessary learning and be adapted to the skill level of the individual participant; that is, motor learning will not be seen if a task poses no challenge [173].

The presence of knowledge of results (KR) feedback during motor learning opens some debate for perfecting learning acquisition. There is always a certain amount of inherent feedback when performing a motor task, but the delivery specifics of additional external feedback is somewhat debated. The frequency with which external feedback is provided should be self-selected by each participant for optimal learning [177,178]; however, this may need to be adapted depending on the difficulty of the task [173]. The timing of feedback being delivered after a trial should be delayed for the participant to consolidate before feedback is given; however, this has been debated based on the complexity of the information given [173]. While the suggestion for feedback emphasis is typically to avoid normative or comparative information, this may in fact be motivating for
many participants [177]. It is evident that the factors associated with KR feedback are still debated for the optimization of motor learning.

Following acquisition of a motor skill, the amount of motor learning can be quantified with retention and transfer testing. Kantak and Winstein [179] were the first to identify that consolidation requires a minimum of 4 hours and that retention should be tested after a minimum of 24 hours of consolidation. The presence of feedback during retention is highly debated. Salmoni and colleagues [180] stated that no-KR retention tests were essential for the measurement of motor learning. However, Russell and Newell [181] argue that a no-KR test, when KR feedback was provided during practice, is a completely different task and represents a transfer test more than a retention test. While transfer tests are conventional in motor learning literature, there appears to be no agreed upon definition as to what constitutes a ‘transferrable’ task. However, it is known that a certain amount of complexity and task alteration is necessary for a transfer test to demonstrate motor learning [173]. Lastly, the effect of warm up (familiarization) should always be accounted for, but rarely is, during retention and transfer tests in motor learning [172,179].

2.5.3 Methodological Controls

Carroll and colleagues [2] recognized the lack of familiarization to strength tasks and amended their initial estimate of cross education from an 11% strength gain to a 7.6% strength gain after accounting for familiarization effects. This difference is important due to the ‘quick-jumps-in-strength’ phenomenon that accounts for initial strength gains after only one testing session [182–184]. In the review of 90 cross education studies only 45% report the use of familiarization ranging from one contraction to one session. It is likely that most studies employed a series of ‘familiarization contractions’ rather than a full
familiarization session. Similarly, the literature has great inconsistencies in the presence, examination, and reporting of control groups. It is well documented that control groups (or control limbs) demonstrate strength increases absent of training due to familiarization and repeated testing. However, many cross education studies have been completed without control groups entirely or without control groups completing sham testing. In the review of 90 cross education studies 58% of studies employed a control group, however, only 45% reported the results of the control group thereby demonstrating the inconsistency in reporting and statistical comparison to control group results.

2.5.4 Surface Electromyography

The use of surface EMG in the investigation of cross education neural adaptations is confounded by the necessity to measure muscle activity during voluntary and evoked contractions simultaneously. Voluntary contractions are used to estimate the amplitude of muscle activity during maximal contractions and antagonist co-activation. Alternatively, M-waves, V-waves, and MEPs are used to monitor muscular adaptations and central drive. This presents a need for the maximal, undistorted waves present from monopolar recording over the motor point [185,186], while obtaining maximal muscle activity, with minimal cross talk, as best recorded by bipolar configurations [187,188].

Furthermore, there are large inconsistencies in the reduction and normalization of amplitude measures. In a review of 25 cross education studies employing surface EMG for voluntary contractions there was no consistent method for analyzing muscle activity, with RMS amplitude and integrated EMG (iEMG) being the most common, conducted both with and without normalization to the M-wave. The remaining studies used a variety of methods including mean amplitude value and peak EMG of a moving average. The
discrepancies in data reduction may explain the vast discrepancies in the contralateral muscle activity adaptation results (range -15 to 58% change post-training). Although large changes in contralateral strength may be present following unilateral training, it’s possible that small neural adaptations are being confounded by the discrepancies in data reduction. Arabadzhiev and colleagues [189] using simulated data to demonstrate that a peripheral alteration (i.e., the minor elevation of intracellular action potentials) elicited a greater increase in RMS amplitude of the surface EMG signal than a central alteration (i.e., a 25% increase in MUFR). In addition, the authors demonstrated that normalization to the M-wave almost entirely eliminated the peripheral alteration allowing a more accurate representation of the central adaptation in the normalized RMS amplitude [189].

The practice of amplitude normalization has several debated factors including statistical legitimacy, when normalization should occur, and what the normalizing factor should be [190,191]. This is especially important in cross education literature due to the number of test sessions over weeks or months, potential muscular adaptations or day-to-day variability, and the inability to maintain identical electrode location. Although it appears to be necessary to normalize to examine neural adaptations across a training program, the issues of statistical reliability and normalization factor still need to be addressed. The primary statistical issue with normalization is the reduction in true biological variation [190,192], which decreases the ability to statistically discriminate between participants and thereby decreases the reliability of the measure [193]. For selection of the normalization factor, Halaki and Ginn [194] outlined the common normalization procedures including peak activation during maximal contractions and M-wave peak-to-peak amplitude. Although M-wave normalization was least recommended
based on low within-subject reliability [194], it is widely used in cross education literature because it provides comparable results between repeated test sessions, muscles, and individuals. The likely recommendation for surface EMG use in cross education literature is a consistent choice of normalization and the increased reporting of methodology and measurement reliability.

2.5.5  *Surface Decomposition*

Indwelling electromyography is considered the ‘gold standard’ for the assessment of motor unit behaviour [195], but is not without disadvantages. Aside from the logistical challenges, such as participant discomfort, smaller sample sizes, and the time-consuming decomposition, there are also theoretical disadvantages. The limited pick up volume of indwelling EMG requires repeated insertion to obtain a sufficient amount of motor units [196–198]. However, needle re-insertion can still result in under-sampling and therefore characterize an entire muscle based on an incomplete sample [199]. Therefore, the alternative, surface decomposition, may be the more accessible and potentially more representative solution to motor unit behaviour analysis.

Surface decomposition (dEMG, Delsys Inc.) uses a 5-pin electrode producing 4 channels of differential muscle activity. As described by De Luca et al. [200], Nawab et al. [201,202] and, Chang et al. [203] the Precision Decomposition Algorithm (III) uses artificial intelligence to identify motor unit action potentials using selective amplitude, duration, and inter-pulse interval criteria. Motor unit action potential templates are created and continuously updated (rewighted) as motor unit superpositions are decomposed and firing occurrences are identified. The decomposition results are validated using a built-in
software employing the Decompose-Synthesize-Decompose-Compare procedure [201,204,205].

The Decompose-Synthesize-Decompose-Compare procedure takes the motor unit action potential trains decomposed from the original signal and forms a synthesized signal with the addition of Gaussian noise equal in amplitude to the residual signal. The synthesized signal is then decomposed again according to the initial procedures. The initial decomposition and synthesized decomposition are then compared, and an accuracy rating is given for each motor unit action potential train [204]:

\[
\text{Accuracy} = 1 - \frac{N_{\text{error}}}{N_{\text{truth}}}
\]

Where \(N_{\text{error}}\) is the discrepancies between the initial and synthesized decompositions (i.e., number of false positives identifying a firing when one does not occur, or false negative missing a firing when one does occur) and \(N_{\text{truth}}\) is the number of correctly matched events.

The Delsys surface decomposition algorithms, although internally validated, are not without criticism. Farina and Enoka [206] identified two major issues of the internal validation program. The first is that missed firings will not be included in the synthesized signal and therefore it will not be present for the second decomposition to identify it. This leads to an over-estimation of accuracy as two false positives will be compared and validate each other. Secondly, the validation procedure is “data-driven” in that the double decomposition procedure may be applied to nearly identical signals [206]. Since the synthesized signal includes Gaussian noise equal to the residual amplitude, a highly decomposed signal would have minimal noise added to it, thereby comparing identical signals. The over-decomposition of a signal incorrectly (i.e., identifying noise as motor units) would appear to be as accurate as a correctly decomposed motor unit action potential.
train. Farina and colleagues [206,207] suggest that the only reliable method of validation is a two-source test comparing indwelling to surface signals. Although previous two-source testing has compared indwelling and dEMG-obtained signals, these have only included a small subset of motor units and uses different algorithms for each [206,208]. It’s evident that surface decomposition is still a controversial method requiring further validation techniques.

2.6 Conclusion

Cross education has the potential to be a primary rehabilitation strategy for unilateral injuries and disorders. The cross-body transfer of strength and motor skills can assist in regaining activities of daily living in the affected limb. To best design a rehabilitation program, all contributing mechanisms of cross education should be considered. Furthermore, if motor learning significantly contributes to the cross-body transfer of strength, then coordination training should be included in unilateral training programs thereby making cross education more accessible to clinical populations. The elucidation of cross education mechanisms requires careful methodological controls and a meta-analysis of the field demonstrates the inconsistencies that obfuscate reviews.
References


[24] Urbin MA, Harris-Love ML, Carter AR, Lang CE. High-Intensity, Unilateral Resistance Training of a Non-Paretic Muscle Group Increases Active Range of
Motion in a Severely Paretic Upper Extremity Muscle Group after Stroke. Front Neurol 2015;6.


Adams JA. Historical review and appraisal of research on the learning, retention, and transfer of human motor skills. Psychol Bull 1987;101:41.


[190] Burden A. How should we normalize electromyograms obtained from healthy participants? What we have learned from over 25 years of research. J Electromyogr Kinesiol 2010;20:1023–35.


3 PURPOSE

The overall purpose of this thesis was to further the field of cross education literature. This dissertation evaluated the effect of unilateral resistance training on the cross education phenomenon, while employing the necessary methodological controls to quantify the contributions of neuromuscular mechanisms and motor learning. Specifically, the proposed research will: (1) examine the neural training adaptations in the trained and untrained limb and their persistence after detraining; and (2) examine the effect of unilateral training on motor control and learning in the trained and untrained limbs.

A secondary purpose of the study was to compare the training and cross education responses between sexes (males versus females) and limbs (upper versus lower limb) on the quantity of cross education and the accompanying mechanisms. These factors have been hypothesized in the literature to not have an effect on the cross education phenomenon; however, no study has experimentally examined their impact.

3.1 Manuscript 1

The purpose of this manuscript was to determine the reliability of a novel surface electromyography (sEMG) technique that produces clean signals for both voluntary and evoked contractions. The proposed sEMG configuration and placement was designed to minimize cross talk during voluntary contractions while maintaining an undistorted wave during evoked contractions. The reliability of the sEMG signal is a crucial methodological control for examining the neuromuscular mechanisms of cross education since voluntary and evoked contractions are required simultaneously for multiple measures.
3.2 Manuscript 2

The purpose of this paper was to produce the most comprehensive meta-analyses of cross education to date. The four previous cross education meta-analyses were limited by strict inclusivity parameters. Each previous analysis was comprised of 16 or fewer studies, excluded evoked contraction training modalities (e.g., electromyostimulation), and limited study inclusion to healthy populations and those reporting full data (means and standard deviations) for both the trained and contralateral limbs. The present meta-analysis prioritized inclusivity over selectivity to capture the greatest overview of the cross education field. A total of 83 ‘contralateral strength transfer’ studies, including 4 conducted in patient populations, were included in the analysis, including studies that unintentionally examined cross education by using an untrained contralateral limb as a control for unilateral training. A secondary purpose of the meta-analysis was to examine the effect of training parameters (e.g., modality, intensity, volume) on the magnitude of cross education.

3.3 Manuscript 3

The primary purpose of the present study was to combine and extend previous work on the neural adaptations in the contralateral limb while evaluating the contribution of motor learning to cross education. To accomplish this, cross education was assessed following a 6-week unilateral training program in the upper and lower limbs known to exhibit neural adaptations and was re-assessed following a 6-week detraining period to evaluate retention. Force, motor unit firing rates, surface electromyographic activity, and force variability were monitored over two pre-training sessions, post-training, and a retention testing session.
Flexor carpi radialis surface EMG electrode placement for evoked and voluntary measures

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Abstract

Introduction: The reliability of bipolar electrode recordings, which allow for undistorted compound muscle action potentials (CMAPs) while minimizing cross-talk during voluntary contractions, was evaluated. Methods: Twenty-four men completed maximal voluntary wrist flexion contractions in 4 test sessions. Compound muscle action potentials were also evoked during each session. Surface electromyography was recorded from the flexor carpi radialis (FCR) with the recording electrode (G1) placed on the motor point and a second recording electrode (G2) adjacent to G1. Reliability was assessed using intraclass correlational analysis of variance and standard error of measurement. Results: Root-mean-square (RMS) amplitude and mean power frequency (MPF) were highly reliable ($R = 0.89$ and 0.84, respectively). The CMAPs also exhibited good reliability ($R = 0.75$). Normalization of RMS amplitude reduced the intraclass reliability coefficient ($R = 0.85$).

Conclusion: The electrode placement resulted in reliable measures from voluntary contractions and CMAPs. Normalization can decrease reliability.
Introduction

The use of surface electromyography (sEMG) can be performed using a monopolar or bipolar configuration, where monopolar consists of one electrode on the belly of the muscle and a second electrode on the muscle tendon rather than bipolar where both electrodes are placed, in parallel sequence, on the belly of the muscle. A monopolar electrode configuration with G1 specifically on the motor point is preferred for evoked responses, such as the compound muscle action potential (CMAP) [1]. The peak-to-peak amplitude is greatest at the motor point location, and the waveform is undistorted by dispersion and initial positivity as the potential propagates towards the electrode [2–4]. However, monopolar recordings are highly susceptible to cross-talk from surrounding muscles during voluntary contractions [5,6]. The problem is exacerbated when limb volume is small and there are a large number of muscles in close proximity, such as in the forearm [6,7]. In this case, a bipolar electrode configuration has long been recommended to minimize potential cross-talk from surrounding muscles [8]. For muscles of the forearm, such as the flexor carpi radialis (FCR), bipolar recording away from the motor point poses 2 problems. In some participants, there may not be enough surface area within the muscle borders to record in a bipolar configuration away from the motor point [9], and the CMAP will be highly distorted [10].

The purpose of this paper is to assess the reliability of a novel electrode placement that may be used when the experiment requires both CMAPs and maximal voluntary isometric contractions of the forearm muscle within the same experiment. Maximal isometric contractions were used, because the level of antagonist coactivation increases with the force of the contraction to maintain joint stability [5,11–13]. Thus, maximal
contractions are an ideal experimental manipulation to test the impact of electrode configuration on the presence of cross-talk.

The electrode placement involves a recording electrode (G1) directly on the motor point with a second recording electrode (G2) immediately adjacent to G1 resulting in a 1 cm center-to-center interelectrode distance. The proposed electrode placement has several advantages: (1) cross-talk is minimized by using differential recording [1,5]; (2) peak-to-peak amplitude of the CMAP is greatest over the motor point [4]; (3) electrode placement avoids the borders of small muscles [7,14]; and (4) the CMAPs retain an initial negativity without a leading positivity [2,3]. To this end, we assessed the reliability of the peak-to-peak (P-P) amplitude of the CMAP, the root-mean-square (RMS) amplitude, and the mean power frequency (MPF) of surface electromyographic (sEMG) recordings from the FCR obtained with these electrode placements. Additionally, antagonist muscle activity was recorded from the extensor carpi radialis (ECR) to assess cross-talk. Changes in reliability due to normalization of the FCR RMS amplitude using the P-P amplitude of the CMAP will also be reported. It was hypothesized that the proposed electrode configuration would result in reliable estimates of muscle electrical activity for the FCR during maximal voluntary and evoked muscle contractions.

**Methods**

Twenty-four men volunteered to participate in this study. Participants were aged 18-30 years, right arm dominant, and had no neurologic/orthopedic abnormalities. Procedures were reviewed and approved by the Brock University Research Ethics Board, and participants signed an informed consent form prior to participation.
Each participant completed a series of maximal isometric wrist flexion strength trials and CMAPs in 4 test sessions separated by at least 48 hours. Four test sessions were used to identify trends in the data associated with repeated testing, since individuals may exhibit increases in maximal isometric strength due to task learning. It has previously been shown that maximal isometric strength can continue to increase across 4 consecutive test sessions, but it tends to plateau after the third [15–17]. Changes in maximal isometric strength can then impact the stability of sEMG mean activity across test sessions [18].

All data collection took place inside a Faraday cage. Participants were seated at a testing table, which allowed the right forearm to rest on the surface at 160° elbow extension (see Figure 1). The back of the upper arm rested on a 20° wedge (not shown) to maintain the elbow joint angle. The forearm was in a custom jig to prevent extraneous movement and keep the wrist in a neutral position. Participants performed isometric wrist flexion by pressing the palm against a metal surface attached to a load cell (JR3 Inc., Woodland, CA), which recorded torque.

sEMG recording electrodes were placed on the flexor carpi radialis (FCR) and extensor carpi radialis (ECR). For each test session the motor points of each muscle were located using low-level repeated percutaneous electrical stimulation [18,19]. The motor points were marked with indelible ink, and participants were asked to retain these marks for the 4 sessions. The procedures for maintaining consistent electrode location on the motor point across test sessions have previously resulted in highly reliable sEMG measures [18,20]. Pediatric size Ag/AgCl electrodes (F-E9M 11 mm, GRASS Technologies, Asto-Med, Inc.) were used for the study. The first electrode (G1) was placed directly on the electrically identified motor point. The second electrode (G2) was placed directly adjacent
to the motor point, in line with the muscle fiber orientation, with an interelectrode distance of 1 cm. A self-adhesive ground electrode (5 cm diameter, CF5000, Axelgaard Manufacturing Co., Ltd., Fallbrook, CA) was placed on the dorsum of the hand. All signals were amplified (Grass P511, Astro-Med, Inc., Warwick, RI) to maximize the resolution of the 16-bit analogue-to-digital converter (PCI-6251, DATAQ Instruments, Akron, OH) and band-passed filtered (3-1000 Hz).

The stimulation procedures followed an earlier study by Christie et al. [21] but were modified for the median nerve. Maximal CMAPs were evoked from the median nerve by stimulating at the cubital fossa with the cathode (3.2 cm diameter, 879100, Axelgaard Manufacturing Co., Ltd., Fallbrook, CA) over the brachial pulse at the elbow crease and the anode (5 cm diameter, CF5000, Axelgaard Manufacturing Co., Ltd., Fallbrook, CA) placed directly across from the cathode above the olecranon process. A series of 5 maximal
CMAPs were evoked with 15-s rest between each twitch. Twitches were evoked (Grass Telefactor S88, Astro-Med Inc., West Warwick, RI) using supramaximal stimulation (~110%). After a minimum of 5-minutes rest, participants performed 5 maximal isometric wrist flexion strength trials. Each contraction was 5-s in duration with 2-minute intertrial rest periods. Participants were instructed to contract as hard and as fast as possible and were given visual feedback via a force trace presented on an oscilloscope (VC-6525, Hitachi, Woodbury, NY).

Flexor carpi radialis RMS amplitude and mean power frequency (MPF) were obtained from a 1-s window taken from the center of each maximal voluntary contraction (MVC). The P-P amplitude was determined for each CMAP. Normalized RMS amplitude was calculated for the FCR by dividing the RMS amplitude by CMAP P-P amplitude. While reliability analysis focused on FCR sEMG measures, the sEMG activity from both the FCR and ECR was used to assess the level of common signal using cross-correlation coefficient [22].

Intraclass correlational analysis of variance was used to assess the reliability of each measure across the 4 testing days, along with the standard error of measurement (SEM). Details of the calculations are described in a previous study by McIntosh and Gabriel [18]. Briefly, the statistical model was a 2-way fully nested analysis of variance that allowed for partitioning of total variance into true score error, day-to-day error, and trial-to-trial error. The mean squares from the analysis of variance were then used to calculate the intraclass reliability coefficient and standard error of measurement [18,20]. The standard error of measurement (SEM) was calculated as the square root of the mean square error from the 2-way analysis of variance model as recommended by Weir [23]. The
intrasubject coefficient of variation (CV) was the SEM expressed as a percentage of the grand mean of the data from the 4 test sessions [24].

**Results**

Figure 2 shows representative tracings obtained from a participant during pilot work. This figure illustrates the differences in cross-talk between monopolar and bipolar electrode configurations. In both cases, G1 is on the motor point, but G2 is either on the distal muscle tendon (monopolar) or placed adjacent to G1 (bipolar). Below each recording is the associated scatter plot comparing the agonist and antagonist muscle, and reference line for the cross-correlation coefficient ($R_{xy}$). There was the expected decrease in magnitude associated with bipolar recordings, and the cross-correlation coefficient decreased from 0.61 to 0.12. Common signal ($R^2_{xy}$) between FCR and ECR was 1.4% for the bipolar configuration. Representative FCR CMAPs for monopolar and bipolar configurations are shown in Figure 3. Other than absolute amplitude, the bipolar CMAP retained the initial negativity. The presence of the FCR CMAP in the ECR is also greatly reduced for studies that examine coactivity in the antagonist. For the current sample, the bipolar configuration resulted in common signal that was less than 3%. The following paragraphs describe the reliability of the sEMG variables obtained with the proposed electrode placements in the FCR.
Figure 2. Monopolar (left panels) and bipolar (right panels) sEMG recordings with associated force traces. The upper panels are representative traces obtained during maximal isometric contractions of the wrist flexors: flexor carpi radialis (black) and extensor carpi radialis (light grey). The scatter plots in the lower panels show the associated cross-correlation ($R_{xy}$) between flexor carpi radialis and extensor carpi radialis sEMG signals.

Figure 3. Monopolar (left panel) and bipolar (right panel) CMAPs evoked in the flexor carpi radialis (FCR). The FCR potentials are the top traces, and the volume-conducted potentials in the extensor carpi radialis (ECR) are the bottom traces of each panel.
The table lists the means, standard deviations, and intraclass reliability analysis for all criterion measures. There was a significant increase in maximal isometric wrist flexion strength \((P < 0.01)\). Post-hoc testing (Tukey HSD) revealed significant increases \((P < 0.01)\) across the first 3 test sessions with a plateau between sessions 3 and 4. As a result, the day-to-day error accounted for 27.9\% of the total variance. The trial-to-trial error was 6.8\% of the total variance. The true score error was 65.3\% of the total variance. The participants were very consistent with respect to maximal isometric wrist flexion strength, as demonstrated by a narrow range of scores. Therefore, the SEM was only 1.44 Nm, the intrasubject CV was 9.1\%, and the intraclass reliability coefficient was high \((R=0.90)\).

As might be expected, the increase in maximal isometric wrist flexion strength was associated with a significant increase in means across days for the FCR RMS amplitude \((P < 0.01)\). The day-to-day error accounted for 29.8\% of the total variance. The differences between trials on each day accounted for only 8.5\% of the total variance. Figure 4 (upper panel) shows that participants were highly consistent and generally exhibited a narrow range of scores; this is illustrated by the vertical standard deviation bars for each participant. The SEM was 0.08 mV (or 80 µV) so that the intrasubject CV was 22.2\%. The narrow range of scores resulted in a low degree of overlap in the scores between participants, so that there was a large between-subjects mean squares, a high true score error (61.7\%), and a high intraclass reliability coefficient \((R=0.89)\). Following normalization of the FCR RMS amplitude to CMAP P-P amplitude, day-to-day error increased to 34.8\% of the total variance. Trial-to-trial error also increased to 11.1\% of the total variance. Compared to non-normalized FCR RMS, normalization resulted in a nearly identical intrasubject CV of 22.0\%. However, the normalization procedure redistributed
the variance so that there was increased overlap between participants as seen in Figure 4 (lower panel). There was a commensurate reduction in true score error to 54.2% of the total variance, resulting in a somewhat lower intraclass reliability coefficient ($R=0.85$).

The change in FCR mean power frequency (MPF) means across days accounted for 37.9% of the total variance, with the differences between trials on each day accounting for 9.0% of the total variance (Table 1). Participants were highly consistent with respect to this measure and had a SEM that was 9.8 Hz, leading to an intrasubject CV of 7.5%. The low degree of overlap of scores between participants resulted in a true score error accounted for more than half of the total variance (53.1%), so that the intraclass reliability coefficient was high ($R=0.84$). The fluctuation in CMAP P-P amplitude means across days accounted for 56.1% of the total variance, with trials accounting for 1.8% of the total variance (Table). The SEM was 0.43 mV and the intrasubject CV was 6.5%. Thus, although the day-to-day error was high, the true score error still accounted for a large portion (42.2%) of the total variance. The resulting intraclass reliability coefficient was $R=0.75$. 
Table 1. Means and standard deviations across the 4 testing days. The variance components (percent accounted for), the standard error of the measurement (SEM), the intrasubject coefficient of variation (CV), and the intraclass correlation coefficient ($R$) for each measure.

<table>
<thead>
<tr>
<th>Source</th>
<th>Torque</th>
<th>Flexor Carpi Radialis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean Torque</td>
<td>Root-Mean-Square Amplitude</td>
</tr>
<tr>
<td>Test Day</td>
<td>Nm</td>
<td>mV</td>
</tr>
<tr>
<td>Day 1</td>
<td>13.52 ± 4.67</td>
<td>0.32 ± 0.21</td>
</tr>
<tr>
<td>Day 2</td>
<td>15.77 ± 4.87</td>
<td>0.41 ± 0.35</td>
</tr>
<tr>
<td>Day 3</td>
<td>16.88 ± 5.06</td>
<td>0.38 ± 0.28</td>
</tr>
<tr>
<td>Day 4</td>
<td>17.33 ± 6.26</td>
<td>0.33 ± 0.24</td>
</tr>
<tr>
<td>Grand Mean ± SD</td>
<td>15.88 ± 5.44</td>
<td>0.36 ± 0.28</td>
</tr>
</tbody>
</table>

Variance Components
(Percent Variance Accounted)

<table>
<thead>
<tr>
<th></th>
<th>Trial-to-Trial</th>
<th>Day-to-Day</th>
<th>True Score</th>
<th>SEM</th>
<th>CV</th>
<th>$R$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>6.8%</td>
<td>8.5%</td>
<td>1.8%</td>
<td>11.1%</td>
<td>9.1%</td>
<td>0.90</td>
</tr>
<tr>
<td></td>
<td>27.9%</td>
<td>29.8%</td>
<td>56.1%</td>
<td>34.8%</td>
<td>22.2%</td>
<td>0.89</td>
</tr>
<tr>
<td></td>
<td>65.3%</td>
<td>61.7%</td>
<td>42.2%</td>
<td>4.3%</td>
<td>6.5%</td>
<td>0.75</td>
</tr>
<tr>
<td></td>
<td>1.44</td>
<td>0.08</td>
<td>0.43</td>
<td>0.013</td>
<td>22.0%</td>
<td>0.85</td>
</tr>
<tr>
<td></td>
<td>9.1%</td>
<td>22.2%</td>
<td>6.5%</td>
<td>7.5%</td>
<td>7.5%</td>
<td>0.84</td>
</tr>
</tbody>
</table>
Figure 4. The intraclass correlation coefficient (R), standard error of measurement (SEM), and intrasubject coefficient of variation (CV) for flexor carpi radialis root-mean-square amplitude (top), and root-mean-square amplitude normalized to compound muscle action potential peak-to-peak amplitude (bottom). The grand mean is shown as the dotted line. The means (circles) and standard deviations (vertical bars) are displayed for each participant.

Discussion

It was expected that there would be changes across sessions in FCR muscle activity during maximal voluntary wrist flexor contractions. Repeated strength testing has been shown to increase maximal isometric strength and sEMG activity due to task learning [16,17]. Four test sessions were used, because such increases in strength tend to plateau after the third consecutive test session [16,17], which was the case here. Thus, a decrease in the stability of means across test sessions was expected [18].

Each of the measures exhibited day-to-day variability resulting in a significant difference between means. However, the lack of stability in means was compensated for by
highly consistent scores within participants; that is, each participant had a narrow range of scores around his own mean [18,20]. The consistency of scores is important, because a reliable measure should discriminate between individuals who are truly different with respect to the characteristic being assessed [18]. If participants are consistent at reproducing scores around their own mean, it allows the test to discriminate between individuals, because the scores of one participant do not overlap with those of another. Higher consistency increases the between-subjects means squares (true score error) which in turn increases the intraclass reliability coefficient [18,25].

We used the SEM and CV as adjunct measures to assess the consistency of scores within subjects. When the SEM is expressed as a percentage of the grand mean of all the data, the result is the intrasubject CV that allows a relative comparison to other measures and other studies. The intrasubject CV for sEMG measures ranged from 6.5-22.2%, which was consistent with previous reports. Mathur, Eng, and McIntyre [26] and Larsson et al. [27] reported the SEM and intrasubject CV of the amplitude and frequency of sEMG signals from knee extensors during an 80% and 100% MVC, respectively. The FCR RMS had a CV of 22.2%, which was slightly lower than the range (23.4-31.9%) observed by Mathur et al. [26] but higher than the range (7-10%) reported by Larsson et al. [27]. Likewise, the FCR MPF had a CV of 7.5%, which was within the range (5.3-10.7%) reported by both Mathur et al. [26] and Larson et al. [27].

Taken together, the SEM, CV, and intraclass correlation coefficients for the sEMG measures, indicate that the changes in means did not interfere with the ability to discriminate between individuals. The bipolar electrode placement with G1 electrode in the innervation zone can be used to obtain reliable measures of the sEMG signal during
evoked and voluntary contractions. Furthermore, this electrode configuration resulted in a common signal between the FCR and ECR that less than 3%, which is in exact agreement with Mogk and Keir [22].

The reliability of the FCR CMAP P-P amplitude was lower ($R=0.75$) than what was reported previously using more clinically oriented techniques. Intraclass reliability coefficients of $R=0.97$ and $R=0.84$ have been reported by Christie et al. [20] and Jaberzadeh et al. [28], respectively. Nevertheless, the intraclass reliability coefficient we observed is still considered to be good [29]. Lee and Carroll [30] used a similar experimental set-up and documented variability in FCR CMAP P-P amplitude, even when the position of the forearm remained constant. We suggest that the higher variability reflects differences between clinical versus experimental techniques.

In our experimental setup, participants were not in anatomical position. Higher intraclass reliability coefficients have been observed when participants were supine in anatomical position, which allowed easy, direct access to percutaneous stimulation of the median nerve [10,20,28]. Test position then interacts with the method of delivering peripheral nerve stimulation. The study by Christie et al. [20] used hand-held prong electrodes to elicit CMAPs. We used stimulation pad electrodes, and while great care was taken to place the stimulating pads in the same location across sessions, even slight alterations in arm position could result in displacement of the nerve relative to the pad electrode. The nerve may not have received the same consistent, direct stimulation that is possible with a hand-held probe. In support of this idea, Phadke et al. [31] recently used a custom-fabricated thermoplastic clamp for the stimulating electrodes. The clamp provided compression, which minimized electrode movement and maintained contact with the
median nerve, while their participants were in an upright test position, similar to our study. The author observed an intraclass reliability coefficient of $R=0.85$ for the ratio of slopes for the H-reflex to M-wave (CMAP) stimulus-response curves. An intraclass reliability coefficient of $R=0.88$ was observed for the ratio of the maximum P-P amplitude of the H-reflex and M-wave. Thus, this stimulation method holds promise for future studies [31].

There are a number of potential limitations associated with bipolar recordings within the innervation zone that should be addressed. Bipolar electrodes placed over the innervation zone tend to record lower signal amplitudes due to cancellation caused by bi-directionally propagating waves [1,32]. However, placing G1 on the motor point, and G2 immediately adjacent can minimize cancellation effects [33]. In support, the grand mean FCR RMS amplitude was $358\pm277 \mu V$ (reported in millivolts in the Table), which is comparable to the only other study to report non-normalized results. Mizuno, Secher, and Quistorff [34] observed $420\pm90 \mu V$ with an inter-electrode distance of 3 cm, resulting in a larger pick-up volume and greater signal amplitude [1]. The P-P amplitude of the CMAP in our study was $6.7\pm3.2 \text{ mV}$, which is greater than $4.4\pm0.9 \text{ mV}$ (range 2.2–6.1 mV) reported by Christie et al. [20] who used a monopolar electrode configuration. Jaberzadeh et al. [28] observed a CMAP P-P amplitude of $4.7\pm1.0 \text{ mV}$ (range 3.4–6.8 mV) using a bipolar electrode configuration. Thus, while cancellation effects are a legitimate concern [1], the FCR CMAP P-P amplitude we observed was greater than previous work, regardless of electrode configuration.

Beck et al. [35] provided experimental evidence that spectral estimates within the innervation zone are higher due to cancellation of lower frequency components of the signal. The MPF we observed in was $130\pm33 \text{ Hz}$ at 100% MVC. Roman-Liu and Bartuzi
observed a MPF of 121±28 Hz while participants performed isometric contractions of the wrist flexors at 50% MVC. The higher MPF we observed is most likely due to muscle fiber conduction velocity changes between 50 and 100% MVC [37,38]. In support, a nearly identical MPF of 123±27 Hz was reported for the tibialis anterior at 100% MVC using bipolar electrodes placed away from the motor point [39].

With this electrode placement G2 may still be within the innervation zone [4]. Therefore, it is reasonable to question the variability of amplitude and spectral estimates we obtained [32,35,40]. However, we have provided empirical evidence that FCR RMS amplitude (R=0.89) and MPF (R=0.84) may be assessed across multiple test sessions with reliability coefficients that are considered excellent [29,41,42]. Because electrodes closer to the motor point have been shown to overestimate conduction velocity [40], we are not advocating this placement for muscle fiber conduction velocity analysis.

It has been previously suggested that sEMG should be normalized to the CMAP P-P amplitude. By subtracting out potential changes within the muscle and technical factors that may vary across test sessions, the remaining sEMG signal should be a true estimate of central drive [43,44]. While conceptually appealing, the normalization procedure poses 2 problems. First, the ratio score combines the measurement error of 2 variables and can redistribute the variance. We observed greater trial-to-trial and day-to-day error variances and a lower intraclass reliability coefficient for the normalized versus non-normalized FCR RMS amplitude.

Decreases in the intraclass reliability coefficient associated with normalization have previously been reported by a number of investigators [27,45–49]. The main reason is that normalization results in a greater overlap in the ratio scores between participants,
decreasing the ability to discriminate between individuals (see Figure 4). The lower true score error decreases the intraclass reliability coefficient, which is also reflected in the coefficient of variability. The normalization of sEMG variables has clinical relevance if the measure is being used to discriminate between patient and non-patient populations [50].

A second, more subtle problem that has been ignored is that normalization violates one of the basic tenants of data transformations prior to statistical analysis: the rank of the participants within the sample should not change [51]. Rank-order would only be maintained if there was a perfect correlation \( r = 1.0 \) between sEMG RMS (numerator) and CMAP (denominator) amplitudes. Inglis et al. [52] investigated the statistical effects of normalization and demonstrated that it is more appropriate to use the denominator as a covariate. Otherwise, spurious relationships can emerge in the analysis and interpretation of the data [53,54].

To summarize, CMAP and voluntary sEMG measures were reliable across multiple test sessions, which show the efficacy of the proposed electrode placement. A bipolar electrode configuration may be used to minimize cross-talk during voluntary contractions while still retaining the undistorted CMAPs for experiments requiring both procedures. Normalizing RMS amplitude by P-P amplitude of the CMAP can increase the coefficient of variation and lower the reliability of criterion measures. Careful methodological controls can result in highly reliable measures without resorting to normalization.
References


Contralateral Strength Gains in Young, Older, and Patient Populations: A Meta-Analysis of Cross Education

Lara A. Green and David A. Gabriel

Submitted to: Physical Therapy Reviews
Abstract

**Background:** Cross education is the contralateral strength gain following unilateral training of the ipsilateral limb. This phenomenon provides an ideal rehabilitation model for acute or chronic rehabilitation. Previous cross education meta-analyses have been limited to a handful of studies and excluded many training paradigms resulting in low study inclusion. The present meta-analysis aimed to (1) be as inclusive as possible, (2) compare cross education in young and older able-bodied participants and in patient populations, and (3) detail the impact of methodological controls on the quantification of cross education.

**Methodology:** A review of English literature was conducted to identify all studies that employed unilateral resistance training and reported contralateral strength results, including unilateral training studies that inadvertently observed cross education by employing the contralateral limb as a control. The percent strength gain and effect size were calculated for ipsilateral and contralateral limbs.

**Results and Conclusions:** A total of 90 studies fit the predetermined inclusion criteria and were included in the analysis. The included studies were further divided into 131 units employing separate unilateral training paradigms. These were separated into young, able-bodied (n=118), older, able-bodied (n=7), and neuromuscular patients (n=6). Cross education was an average of 18% (standardized mean difference (SMD) = 0.70) in young, able-bodied participants, 17% (SMD = 0.59) in healthy able-bodied participants, and 29% (SMD = 0.76) in neuromuscular patients. Electromyostimulation training was superior to voluntary training paradigms for both ipsilateral and contralateral strength gains. The magnitude of strength transfer was similar between upper and lower and between males and females.
Introduction

Cross education is the strength gain that is found in the contralateral limb following a unilateral training program on the homologous limb. Cross education was first reported in 1894 by Scripture, Smith, and Brown [1] who determined that task steadiness and muscular strength could be improved in the contralateral limb following unilateral training. This phenomenon is of great importance for clinical applications and rehabilitation and requires further mechanistic investigation. Cross education provides a beneficial rehabilitation model for unilateral injuries or disorders; including, acute injuries or immobilization (casting) of a single limb, and neurologic disorders, such as stroke, affecting the body unilaterally.

Previous research has proposed that cross education can be explained by two distinct, but not necessarily mutually exclusive, hypotheses: ‘cross-activation’ and ‘bilateral access’ [2,3]. The ‘cross-activation’ hypothesis proposes that unilateral activity excites both ipsilateral and contralateral cortical motor areas. With this hypothesis, the unilateral training causes adaptations in both hemispheres, though to a lesser extent in the untrained hemisphere. Alternatively, the ‘bilateral access’ hypothesis suggests that the homologous untrained muscle can access the unilateral adaptations of training through interhemispheric communication from the associated motor areas [2,3].

Previous meta-analyses and systematic reviews have determined that the average contralateral strength gain from cross education is approximately 8-12% [4–7]. This amount corresponds to approximately 35-60% of the strength increase that is found in the ipsilateral (trained) limb [4,6,8]. However, these previous reviews of cross education were limited to 2 [9], 8 [8], 10 [10], 13 [6], 16 [4], and 31 [7] articles. There are several factors that make the review of cross education complicated and limited, including the name
discrepancies confounding the search for studies, and the variety of training paradigms. However, the primary reason for the small ‘sample sizes’ of cross education reviews is the stringency of inclusion criteria. The reviews by Munn et al. [6], Carroll et al. [4], Cirer-Sastre et al. [10], and Manca et al., [7] were limited to the analysis of randomized controlled studies. In addition, only studies with full data (means and standard deviations) for each of the ipsilateral experimental, contralateral experimental, and control limbs were included.

The inconsistent terminology and the unintentional examination of cross education using the contralateral limb as a ‘control limb’ for unilateral training has confounded the analysis of the field. Cross education of strength has been referred to by many names including cross-transfer, cross-over, or contralateral training. Similarly, the cross education of skill following unilateral practice is typically referred to as inter-lateral transfer of learning, bilateral transfer, or intermanual transfer. These studies generally focus on single session practice, rather than training; and the transfer of a skill, rather than strength. Although widely studied, the practice paradigms and the outcome measurements of the cross education of skill vary drastically across studies making them extremely difficult to quantitatively compare. Therefore, this meta-analysis focuses solely on the cross education of strength.

Lastly, variability in training paradigms makes it difficult to compare cross education between studies. There is a considerable variation in the duration (number of sessions), volume (contractions per session), intensity (load), and modality (type of contraction or stimuli) of unilateral training. The reviews by Carroll et al. [4] Munn et al. [6], and Manca et al. [7] limited their analyses to studies employing training intensities greater than 50%
maximal strength for a minimum of 2 weeks. Most notably, the previous meta-analyses included only isometric, isokinetic, and dynamic training [4,6,7,10], specifically excluding ‘alternative’ training via electromyostimulation (EMS), transcranial magnetic stimulation, vibration, or acupuncture.

The present analysis prioritized inclusivity over selectivity to capture the greatest overview of the field. A review of literature was undertaken to include as many ‘contralateral strength transfer’ studies as possible, including studies that unintentionally examined cross education by using an untrained contralateral limb as a control for unilateral training. In order to advance the use of cross education for rehabilitation purposes, the analysis was not limited to healthy populations as long as strength was assessed pre and post intervention.

**Methods**

**Definitions**

For the purpose of this analysis the term study will refer to an article as referenced. The term unit will refer to a training paradigm within a study, while the term limb will be the designated trained, untrained, or control limb of a participant. For example, one study may have two units within it where one unit was assigned to one type of training (e.g., eccentric training, elbow flexion training, low frequency training, etc.) and another unit was assigned to a separate training paradigm (e.g., concentric training, knee flexion training, high frequency training, etc.).

**Literature Search**

The included studies were collected from a review of cross education and unilateral training literature, rather than a targeted search of databases. The reference list of each
study was examined to include previously noted cross education studies. In addition, studies using unilateral training were examined for the unintentional observation of cross education where the contralateral limb was designated as a control limb.

**Inclusion Criteria**

The selection of inclusion criteria was designed to be as inclusive as possible for the broadest review possible.

**POPULATION.** All ages, sexes, and abilities were included in the present review. Units were separated into three groups: (1) young able-bodied (young) participants (< 50 years of age), (2) older able-bodied (older) participants (>50 years of age), and (3) neuromuscular disorder (patient) populations.

**TRAINING PARADIGM.** All training types aimed at improving strength were included in the present study, including EMS training which has been previously excluded from cross education meta-analyses. Training modalities (contraction types) were separated into the following categories: isometric, isokinetic, dynamic (including isotonic), EMS, or ‘other’. If two types of voluntary contractions were performed for training, then the unit was placed in the ‘other’ category. The EMS category consists of stimulation alone or superimposed on a voluntary contraction. Any training intensity (load) was included as long as it was greater than 0% maximal strength (i.e., the intention was strength gain, rather than endurance gain). The criteria for number of sessions was >5 sessions to include training stimuli rather than mechanistic examinations.

**OUTCOMES.** Studies were included if strength was measured and reported in any manner including: pre-training and post-training means, mean gain, or percent gain. Studies were further separated into units only where separate training paradigms were employed, rather
than separate outcomes. Where one training unit had multiple outcomes the single outcome that was homologous to the training modality (i.e., closest in contraction type, joint angle, speed of contraction, etc.) was selected, with the exception of EMS, vibration, or electro-acupuncture training, where a voluntary contraction was selected. When multiple contraction types were used for training as well as testing, the contraction type used most in training was selected as the outcome measure.

SAMPLE SIZE. The inclusion criterion for unit sample size was \( \geq 3 \) to get an appropriate mean and standard deviation for effect size calculation. No control group was required for inclusion in the analysis.

**Analysis**

EFFECT SIZE. Where means and standard deviations were reported effect size was calculated for each limb within a unit using The Cochrane Collaboration Review Manager (RevMan V.5.3) [11]. The standardized mean difference (SMD) and 95% confidence intervals were calculated using inverse variance as the statistical method, and random effects as the analysis model. Statistical significance (Z-score) was calculated in RevMan to determine if the effect was greater than null. Where standard error (SE) was reported it was converted to standard deviation (SD) using the following formula including group sample size (n):

\[
SD = SE \times \sqrt{n}
\]

The effect size was calculated where possible for the experimental limbs (trained and untrained) and the control limb(s). If both limbs of the control group were measured (dominant and non-dominant) then each limb was separately used as a control for the
experimental limb. If only one control limb was tested then it was included as the control for both the trained and untrained experimental limbs.

PERCENT GAIN. Where means were reported the percent gain of the trained and/or untrained limb was calculated according to the following formula:

\[ \% \text{ Gain} = \frac{Post - Pre}{Pre} \times 100 \]

If only percent gain was reported but not pre-training or post-training mean values then the percent gain was included as reported.

CROSS-BODY TRANSFER. The magnitude of cross-body transfer was calculated to determine how much of the training effect was transferred to the untrained limb. The calculation was conducted for each unit as follows:

\[ \text{Cross-body Transfer} = \frac{Untrained \% \text{ Gain}}{Trained \% \text{ Gain}} \times 100 \]

COMPARISONS. Independent sample t-tests were performed using SAS 9.4 (SAS Institute Inc., Cary, NC) with a 0.05 significance level. The magnitude of percent gain in the untrained (cross education) limb and the trained limb was examined between (1) upper versus lower limb, (2) males versus females, and (3) familiarized versus non-familiarized units. The upper limb training consisted of elbow flexion, wrist flexion and extension, and handgrip exercises amongst others. The lower limb training consisted primarily of knee extension and flexion, and secondarily plantar flexion and dorsiflexion exercises. The effect of sex was examined from units that were composed of only males or only females. Lastly, familiarization was taken as reported and included anything from a familiarization contraction or testing procedures familiarization to an entire familiarization session.
Results

Study and Unit Characteristics

A total of 107 studies were identified and 90 studies were included in the analysis. The 17 excluded studies did not fit the following criteria: no strength data reported for untrained limb (4 studies), no strength measure (4 studies), no pre-test data (4 studies), less than 5 training sessions (2 studies), and less than 3 participants (3 studies). The remaining 90 studies included a total of 131 units. Of those, 118 units (from 81 studies [12–91]) included young, able-bodied participants with a median age of 23 years and a median sample size of 11 (range 3–342) participants. Seven (7) units (from 6 studies [13,27,72,92–94]) included older, able-bodied participants with a median age of 73 years and a median sample size of 11 (range 6-14).

The remaining 6 units (from 6 studies [95–100]) were conducted using neuromuscular patient populations with a median sample size of 10 (range 5-21) participants. The neuromuscular disorder breakdown is as follows: stroke patients (3 studies), patients with various neuromuscular disorders (1 study), multiple sclerosis (1 study), and osteoarthritis patients (1 study).

Outcome Measures

The training characteristics are presented in Table 1 for each of the groups. The results of effect size and percent gain for the number of units that fit each criterion are reported for the untrained (cross education) limb in Table 2 and for the trained limb in Table 3. Forest plots are presented for the untrained limb in Figure 1 for the young group and Figure 2 for the older (2A) and patient (2B) groups.
Table 1. Median and range of training characteristics.

<table>
<thead>
<tr>
<th>Training Characteristic</th>
<th>Young</th>
<th>Older</th>
<th>Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of Training Sessions</td>
<td>21 (6 – 84)</td>
<td>27 (6 – 36)</td>
<td>18 (16 – 27)</td>
</tr>
<tr>
<td>Training Volume (Sets x Reps)</td>
<td>30 (3 – 250)</td>
<td>34 (20 – 40)</td>
<td>33 (24 – 42)</td>
</tr>
<tr>
<td>Training Intensity (% maximum)</td>
<td>100 (10 – 100)</td>
<td>100 (70 – 100)</td>
<td>100 (80 – 100)</td>
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</tbody>
</table>

The average percent gain (above baseline strength) in the untrained contralateral limb of young participants following unilateral training in the ipsilateral limb was 18%, as calculated from 118 units. A review of 79 units with adequate cross education data (means and standard deviations of the untrained limb) resulted in an effect size of 0.70 (95% CI: 0.58 – 0.83, p < 0.001). The amount of cross education was similar amongst different training modalities with the exception of EMS training. Electromyostimulation training was employed in 9 units, which demonstrated an average strength gain of 28%. Five units reported enough data to calculate effect size which was large [101] at 1.65 (95% CI: 0.74 – 2.55, p < 0.001). This is greater than the small effect size of 0.08 (95% CI: -0.05 – 0.22, p = 0.21) in the control limb, which corresponded to a mean 3% gain.
Fig. 1 Forest plot of Standardized Mean Difference (SMD) for each young unit included in the analysis for the untrained (cross education) limb. Light grey lines indicate cut-off values for small (0.2), moderate (0.5) and large (0.8) effect sizes.
The average percent gain in the untrained limb of older participants following unilateral training was 17%, as calculated from 7 units. A review of 4 units with adequate cross education data resulted in an effect size of 0.59 (95% CI: 0.17 – 1.01, p < 0.01). The modes of training included: dynamic (3), isokinetic (2), isometric (1), and resistance tubing (1). The amount of cross education in the Patients subgroup was a 29% strength gain (calculated from 6 units), which corresponded to a large effect size of 0.76 (95% CI: 0.21 – 1.31, p < 0.01, calculated from 4 units). Five of the studies employed strength training (resistive exercises) of the less-affected limb, one study [96] employed kicking and tracking movements of the less-affected limb while secured to a tilt-table.

![Fig. 2 Forest plot of Standardized Mean Difference (SMD) for each older (A) and patient (B) unit included in the analysis for the untrained (cross education) limb. Light grey lines indicate cut-off values for small (0.2), moderate (0.5) and large (0.8) effect sizes.](image)

The influence of limb, sex, and familiarization had no influence on the percent gain of the untrained or trained limb, or the cross-body transfer, as presented in Table 4.
Table 2. Effect size (standardized mean difference), percent gain, and controlled percent gain for the **untrained** (contralateral) limb.

<table>
<thead>
<tr>
<th></th>
<th>N (units)</th>
<th>Effect Size (95% CI)</th>
<th>N (units)</th>
<th>% Gain</th>
<th>Cross-Body Transfer</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Young</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Isometric</td>
<td>24</td>
<td>0.72 [0.56, 0.89]**</td>
<td>36</td>
<td>15%</td>
<td>65%</td>
</tr>
<tr>
<td>Isokinetic</td>
<td>23</td>
<td>0.61 [0.41, 0.80]**</td>
<td>31</td>
<td>20%</td>
<td>70%</td>
</tr>
<tr>
<td>Dynamic</td>
<td>21</td>
<td>0.61 [0.37, 0.85]**</td>
<td>35</td>
<td>19%</td>
<td>61%</td>
</tr>
<tr>
<td>EMS</td>
<td>5</td>
<td>1.65 [0.74, 2.55]**</td>
<td>9</td>
<td>28%</td>
<td>76%</td>
</tr>
<tr>
<td>Other</td>
<td>6</td>
<td>0.46 [0.18, 0.74]**</td>
<td>7</td>
<td>12%</td>
<td>80%</td>
</tr>
<tr>
<td><strong>Older</strong></td>
<td>4</td>
<td>0.59 [0.17, 1.01]**</td>
<td>7</td>
<td>17%</td>
<td>52%</td>
</tr>
<tr>
<td><strong>Patients</strong></td>
<td>4</td>
<td>0.76 [0.21, 1.31]**</td>
<td>6</td>
<td>29%</td>
<td>77%</td>
</tr>
<tr>
<td><strong>Control Limb</strong></td>
<td>36</td>
<td>0.08 [-0.05, 0.22]</td>
<td>58</td>
<td>2.4%</td>
<td>--</td>
</tr>
</tbody>
</table>

CI: confidence interval; EMS: electromyostimulation.*p < 0.05, **p < 0.001, for effect size only. Cross-body transfer is the amount of strength gain transferred from the ipsilateral limb to the contralateral limb.

Table 3. Effect size (standardized mean difference), percent gain, and controlled percent gain for the **trained** (ipsilateral) limb.

<table>
<thead>
<tr>
<th></th>
<th>N (units)</th>
<th>Effect Size (95% CI)</th>
<th>N (units)</th>
<th>% Gain</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Young</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Isometric</td>
<td>25</td>
<td>1.11 [0.90, 1.32]**</td>
<td>36</td>
<td>25%</td>
</tr>
<tr>
<td>Isokinetic</td>
<td>20</td>
<td>0.95 [0.79, 1.11]**</td>
<td>30</td>
<td>31%</td>
</tr>
<tr>
<td>Dynamic</td>
<td>19</td>
<td>1.30 [1.00, 1.60]**</td>
<td>33</td>
<td>33%</td>
</tr>
<tr>
<td>EMS</td>
<td>6</td>
<td>2.12 [1.13, 3.11]**</td>
<td>9</td>
<td>37%</td>
</tr>
<tr>
<td>Other</td>
<td>6</td>
<td>0.53 [0.24, 0.81]**</td>
<td>7</td>
<td>15%</td>
</tr>
<tr>
<td><strong>Elderly</strong></td>
<td>4</td>
<td>1.38 [0.80, 1.96]**</td>
<td>7</td>
<td>32%</td>
</tr>
<tr>
<td><strong>Patients</strong></td>
<td>2</td>
<td>0.56 [0.11, 1.01]*</td>
<td>4</td>
<td>29%</td>
</tr>
<tr>
<td><strong>Control Limb</strong></td>
<td>31</td>
<td>0.13 [-0.02, 0.28]</td>
<td>52</td>
<td>3.2%</td>
</tr>
</tbody>
</table>

CI: confidence interval; EMS: electromyostimulation.*p < 0.05, **p < 0.001, for effect size only.
Table 4. The number of units that fall within each category: sex of the unit, the usage of familiarization, the limb involved, and the presence of a control group from the able-bodied participants (N = 125 units).

<table>
<thead>
<tr>
<th></th>
<th>Number of Units</th>
<th>Number of Participants</th>
<th>% Gain (Untrained)</th>
<th>% Gain (Trained)</th>
<th>Cross-Body Transfer</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Limb Trained</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lower</td>
<td>63</td>
<td>1521</td>
<td>19%</td>
<td>28%</td>
<td>70%</td>
</tr>
<tr>
<td>Upper</td>
<td>62</td>
<td>744</td>
<td>17%</td>
<td>30%</td>
<td>63%</td>
</tr>
<tr>
<td><strong>Sex of Unit</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males Only</td>
<td>62</td>
<td>970</td>
<td>16%</td>
<td>26%</td>
<td>65%</td>
</tr>
<tr>
<td>Females Only</td>
<td>25</td>
<td>727</td>
<td>18%</td>
<td>33%</td>
<td>55%</td>
</tr>
<tr>
<td>Both Sexes</td>
<td>29</td>
<td>456</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unknown Sex</td>
<td>9</td>
<td>112</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Familiarized</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>58</td>
<td>1302</td>
<td>19%</td>
<td>30%</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>67</td>
<td>963</td>
<td>17%</td>
<td>29%</td>
<td></td>
</tr>
<tr>
<td><strong>Control Group (Y/N)</strong></td>
<td>74 / 51</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note: the presence of a control group (“Y”) does not indicate the reporting of control results.
*significant difference between unit categories (Lower v Upper; Males v Females; Yes v No Familiarization), p < 0.05.

**Discussion**

The primary aim of the current meta-analysis was to prioritize inclusivity for the largest systematic analysis of cross education. Secondarily, this meta-analysis aimed to further cross education within the rehabilitation field by quantifying the presence of cross education in young and older able-bodied participants, as well as in patient populations. By carefully identifying the crucial inclusion criteria and reducing inclusion selectivity this meta-analysis was able to include data from 90 studies with 131 units of training groups.

The cross education gain was an 18% increase from baseline strength in young, able-bodied adults; a 17% increase in older, able-bodied participants, and a 29% increase in a patient population consisting of post-stroke, neuromuscular disorders, and osteoarthritis patients. The values of cross education are higher than the previous and most widely cited...
estimates of 8% by Carroll et al., [4] and Munn et al., [6], and higher than the recent estimate of 12% reported by Manca et al., [7]. The cross-body transfer to the untrained limb ranged from 52% to 80% of the ipsilateral training effect.

The separation of training modalities allowed for the analysis of cross education and training adaptation from different contraction types with sufficient sample sizes and statistical power. This identified the advanced capabilities of electromyostimulation (EMS) training producing a cross education effect of 28%, of which previous meta-analyses excluded [4,6,7,10]. Compared to cross education produced by isokinetic (20%), dynamic (19%), and isometric (15%) voluntary contractions, it is evident that EMS training produces a superior transfer of strength. The logistical ease of EMS training for varying populations and the associated voluntary strength gains, make it an ideal modality for cross education in rehabilitation settings.

There were numerous methodological deficiencies that were identified by previous meta-analyses including the need for control group data [6] and the lack of familiarization [4]. Both of these methodological controls are instituted for the purpose of minimizing “quick jumps in strength” that would over-estimate the magnitude of cross education. The present meta-analysis included 58 control units reporting an average strength gain of 2.4% (median: 2.3%, range: -6% – 11%). Therefore, the inclusion of a control group is important to account for the over-estimation of cross education due to extraneous factors such as task familiarization.

It has been shown that task familiarity and familiarization contractions can increase force approximately 3-11% within a single session [102–105]. Carroll and colleagues [4] estimated that the effect of familiarization on the overestimation of cross education was
approximately 4%. Therefore, it is surprising that there was no significant difference in the strength gain between groups that were familiarized and those that were not. It was hypothesized that a lack of familiarization would overestimate the magnitude of the cross education and training strength gain. The likely reason for the absence of difference in the strength gain is the lack of reporting in the majority of studies as to what was considered to be ‘familiarization’. Since most studies neglected to detail the method of familiarization, any study which noted that it’s participants were ‘familiarized’, be it a demonstration, a single test contraction, or an entire session, was included in the ‘familiarized’ group.

The large number of units included in the present meta-analysis allowed for the comparison of cross education between upper and lower limbs and between sexes in 125 units of able-bodied participants. Manca et al., [7] separated 31 studies into upper and lower limb training finding a larger magnitude of cross education in the lower limb (16.4%) compared to the upper limb (9.4%). However, the present meta-analysis found no significant difference between cross education in the lower (19%) and upper (17%) limbs.

Similarly, there was no significant difference in the magnitude of cross education between males (16%) and females (18%), However, comparison between sexes in the trained limb revealed slightly larger (p = 0.07) training adaptations in females (33%) compared to males (26%). This resulted in a slightly larger (p = 0.23) cross-body transfer of strength in males compared to females. To date, many studies have assumed an equality between sexes in the magnitude of cross education, often citing the review by Zhou [8], which does not compare sexes. In the literature, only two studies [43,94] included sex comparisons following unilateral training. Both studies also found significant differences between sexes in the magnitude of the training adaptation, but no difference in the
magnitude of cross education. This indicates that there is a difference in the amount of transfer (or ratio between trained and untrained limbs) between the sexes, however previous literature is conflicting. Hubal and colleagues [43] found a significantly higher strength cross-body transfer ratio in females (21%) compared to males (16%). Alternatively, Tracy and colleagues [94] found a significantly lower strength transfer ratio in females (32% transfer) compared to males (36% transfer).

Conclusion

A review of 131 unilateral training units resulted in a cross education strength gain of 18% in young adults, 17% in older adults, and 29% in a patient population, which is higher than previous estimates [4,6,7] of 8% to 12%. The cross education effect was accompanied by a significant moderate to large effect size in each population. The average cross-body transfer ranged from 52% to 77% slightly higher that previous estimates of 35-60% [4,6]. The present analysis identified: the presence of cross education in young and older able-bodied participants as well as patient populations; the efficacy of EMS training over voluntary modalities; and the equivalence in cross education between upper and lower limbs as well as in males and females. The 17-29% magnitude of cross education is promising for the use of unilateral training in rehabilitation.
References


[49] **Kidgell DJ, Stokes MA, Pearce AJ.** Strength training of one limb increases corticomotor excitability projecting to the contralateral homologous limb. Motor Control 2011;15:247–266.


The neuromuscular adaptations contributing to cross education of strength and skill in the upper and lower limbs

Lara A. Green and David A. Gabriel

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Abstract

Cross education is the strength gain or skill improvement transferred to the contralateral limb following unilateral training or practice. The present study examined the transfer of both strength and skill following a strength training program. Forty participants (20M, 20F) completed a 6-week unilateral training program of dominant wrist flexion or dorsiflexion. Strength, force variability, and muscle activity were assessed pre-training, post-training, and following 6-weeks of detraining (retention). Analyses of covariance compared the experimental limb (trained or untrained) to the control (dominant or non-dominant). There were no sex differences in the training response. Cross education of strength at post-training was 6% ($p<0.01$) in the untrained arm and 13% ($p<0.01$) in the untrained leg. Contralateral strength continued to increase following detraining to 15% in the arm ($p<0.01$) and 14% in the leg ($p<0.01$). There was no difference in strength gains between upper and lower limbs ($p>0.05$). Cross education of skill (force variability) demonstrated greater improvements in the untrained limbs compared to the control limbs during contractions performed without concurrent feedback. Significant increases in V-wave amplitude ($p=0.02$) and central activation ($p<0.01$) were highly correlated with contralateral strength gains. There was no change in agonist amplitude or motor unit firing rates in the untrained limbs ($p>0.05$). The neuromuscular mechanisms mirrored the force increases at post-training and retention supporting central drive adaptations of cross education. The continued strength increases at retention identified the presence of motor learning in cross education, as confirmed by force variability.
Introduction

The cross education of strength and skill learning was first discovered in 1894 by [1], Smith and Brown who determined that muscular strength and task steadiness could be improved in the contralateral limb following unilateral training. Cross education of strength refers to the strength gain that is transferred to the contralateral limb following a unilateral training program in the ipsilateral limb. The mechanisms behind the cross education of strength include cortical and spinal adaptations, which alter the neural drive to the contralateral, untrained limb [2,3]. The same is true for the cross education of skill following unilateral motor task practice, which is typically examined following 1-2 sessions of unilateral motor task practice.

Leung and colleagues [4] found no difference in transcranial magnetic stimulation (TMS) responses following a single session of unilateral metronome-paced elbow flexion (strength training) and visuomotor tracking of the upper limb (skill practice), indicating the similarity of mechanisms contributing to the cross education of strength and skill. However, most studies in this field examine the transfer of strength and skill separately following unique training paradigms [3,5]. Since all resistive exercise (i.e., strength training) includes a skill component, and therefore the potential for motor learning [6–8], it is likely that a strength training program results in the cross education of both strength and skill. Therefore, the present study investigated the presence of skill transfer (cross education of skill) following unilateral strength training.

Early meta-analyses of cross education estimated that the contralateral strength gain was approximately 7.8% [9,10]. However, these analyses were limited to few studies, and have been expanded by Manca and colleagues [11] resulting in an estimated contralateral strength gain of 11.9%. Furthermore, a comprehensive review of the literature including 79
studies, undertaken by the authors, estimates the contralateral strength gain to be an estimated 18%. The potential of cross education for clinical purposes is evident; however, the improvement of motor skills is an important clinical aspect that has been widely overlooked following strength training. Keogh and colleagues [5] examined motor output variability after unilateral task-specific coordination training (joint angle trajectory matching) versus general strength training (unilateral bicep curls and wrist flexion training). Although only the coordination training group had improved variability in the contralateral limb, both groups improved bilateral co-activation, indicating that the ‘task to assess variability’ (postural tremor during pointing) may have been too dissimilar from the strength training movements.

While a change in co-activation is a neural adaptation associated with coordination, changes in task variability demonstrate a motor learning dimension that represent adaptation of the internal model associated with the task [12,13]. Previous cross education studies examining coactivation in the contralateral limb, have demonstrated improvements in co-activation [14–18]. However, to our knowledge the variability of a voluntary contraction has yet to be examined in the contralateral limb following unilateral strength training. In addition, no unilateral training study has examined the changes in co-activation following a period of detraining, to determine the persistence of motor learning.

There are discrepant findings in cross education literature with respect to changes in neural drive to the homologous muscle. Despite the evidence that unilateral training increases contralateral neural drive and corticospinal excitability [3,19–22], previous studies examining the cross education of strength have found equivocal results in measures of central drive. The amplitude of contralateral muscle activity following unilateral training
has been found to increase approximately 28% (range from 3 to 59%) in the majority of studies; however, 8 studies have found no significant change in agonist activity [15–17,19,23–31].

Studies assessing the ‘completeness of activation’ as assessed by twitch interpolation have shown a significant increase [30,32], or no change [33]. Similarly, V-wave amplitude has been examined in two cross education studies with Colomer-Poveda [24] demonstrating no change, and Finland and colleagues [16] demonstrating a non-significant 29% increase. In the latter case, the authors concluded that the high variability of the measure was the reason for the non-significant finding. Lastly, an increase in motor unit firing rates (MUFRs) would be expected to accompany an increase in neural drive to the muscle [34,35]. However, in two studies examining unilateral training, there was no increase in MUFRs in the contralateral limb at post-training [29,36]. This result is surprising and deserves additional examination.

Thus, there are a number of unresolved issues that motivate a re-examination of cross education to assess both strength and skill following a unilateral strength training program. Therefore, 20 males and 20 females completed a 6-week unilateral training program in the upper and lower limbs, with assessment at pre- and post-training, as well as following a 6-week detraining period to evaluate retention. It was hypothesized that unilateral strength training would increase contralateral strength and central drive, as demonstrated with increased central activation and V-wave amplitude. Furthermore, it was hypothesized that strength training would result in the cross education of skill, as demonstrated by decreased force variability in the contralateral limb.
Methods

Experimental approach to the problem

The measurement scheduled was designed to explore both the strength and motor learning aspects of strength training. There were two pre-training sessions (familiarization and baseline), separated by a minimum of 48 hours and a maximum of 7 days. The first pre-test session was used to subtract initial gains in strength due solely to task familiarity, so that all comparison started from baseline. Training adaptations and motor learning were assessed immediately following 6-weeks of unilateral strength training and with a retention test after 6-weeks of detraining. We have previously shown that decreases in antagonist co-contraction and force variability for maximal effort contractions decreased following practice and were retained after 2-weeks and 3-months of detraining [7].

All four limbs of each participant were tested at each session, regardless of group assignment. Unilateral training effects of strength training were evaluated in the following way. The dominant arm of the leg-training group, served as the control for the dominant (trained) arm of the arm-training group (see Figure 1a, comparison 1). Conversely, the dominant leg of the arm-training group was the control for the dominant (trained) leg of the leg-training group (see Figure 1a, comparison 2). Cross education was evaluated by comparing the non-dominant limbs. The non-dominant arm from leg-training group served as the control for the non-dominant (untrained) arm of the arm-training group (see Figure 1b, comparison 3). Similarly, the non-dominant leg from the arm-training group served as the control for the non-dominant (untrained) leg of the leg-training group (see Figure 1b, comparison 4). Surface EMG (sEMG) was monitored in both dominant and non-dominant limb while testing regardless of the limb performing the movements, to ensure that contralateral adaptations were due to cross education rather than force irradiation (i.e.,
postural stability during testing). Instructions for performing maximal voluntary contractions (MVCs) and ramp contractions were kept consistent across participants and sessions to minimize experimenter influence.

Figure 1. Statistical comparisons for the training effects of the trained arm (1) and leg (2), and cross education effects of the untrained arm (3) and leg (4) compared to the relevant dominant and non-dominant controls. The experimental (A: ipsilateral and B: contralateral) limbs of the training group are noted in black.

Participants

Forty participants (20 males, 20 females) reviewed and signed informed consent documents as approved by the Brock University Research Ethics Board and conducted according to the principles expressed in the Declaration of Helsinki. All participants were young adults (24 ± 3 years), recreationally-moderately active, and free of self-reported neurological or orthopaedic abnormalities as ruled out by the Physical Activity Readiness Questionnaire (PAR-Q+) from the Canadian Society for Exercise Physiology. Handedness and footedness were determined using an adapted version of the Lateral Preference Inventory [37]. Participants were randomly assigned to either the arm-training group
(ATG; wrist flexion training) or the leg-training group (LTG; dorsiflexion training) with an equal number of males and females within each group. The dorsiflexors and wrist flexors were selected due to their involvement in activities of daily living and gait, and the importance of cross education to post-stroke rehabilitation. The strength of the dorsiflexor muscles were selected due to their importance in gait and posture and the common occurrence of post-stroke weakness (i.e., drop foot) [25,38,39].

**Experimental set up**

All testing took place inside a Faraday cage in the Electromyographic Kinesiology Laboratory at Brock University. Participants were seated in a jig to isolate the intended muscles and minimize extraneous movement. A computer screen was placed in front of the participant to display their force. For dorsiflexion contractions participants were seated with the hip, knee, and ankle at 90° angles. Isometric contractions were performed by contracting against a metal bar, which was lowered over the top of the foot (dorsiflexion), and pressing down upon the footplate (plantar flexion). For wrist flexion contractions participants were seated at a table with their arm resting on the surface at 160° of elbow extension. The forearm was in a neutral (half-supinated) position and the hand was fixed between metal bars to contract isometrically against the palmar (wrist flexion) or dorsal (wrist extension) bar.

**Surface electromyography**

The tibialis anterior (TA), soleus, flexor carpi radialis (FCR), and extensor carpi radialis on both the right and left sides were prepared for sEMG. The skin was shaved, cleansed with isopropyl alcohol and lightly abraded. The motor point of each muscle was found using low-level electrical stimulation passed over the surface of the skin at a rate of
1.5 pulses per second. The motor point was determined as the point at which a barely visible contraction was elicited with the least amount of stimulation. Once found, the motor point was marked with indelible ink. Pediatric sized electrodes (5 mm electrode diameter, F-E9 20 mm, GRASS Technologies, Asto-Med, Inc., Warwick, RI) were placed in a bipolar configuration with one electrode directly over the motor point and the second electrode placed immediately adjacent, in line with the muscle fibres, for an interelectrode distance of 1 cm. This electrode configuration and placement has been previously demonstrated to minimize cross-talk during voluntary contractions while maintaining the compound muscle action potential shape during evoked contractions, with a high inter-session reliability (ICC=0.85) [40]. By electrically identifying the motor point we could maintain consistent electrode placement across sessions. Ground electrodes were placed on the medial malleolus and patella for the lower leg recordings, and on the back of the hand and olecranon process for the forearm recordings. Surface EMG was amplified 500-2,000 times (Grass P511, Astro-Med, Inc., Warwick, RI) to maximize the resolution of the 16-bit analogue-to-digital converter (DI-720, DATAQ Instruments, Akron, OH). The signals were band-pass filtered (3-1,000 Hz) before digitization at 2,000 Hz (WinDaq Acquisition, DATAQ Instruments, Akron, OH). Skin-electrode impedance was recorded pre- and post-testing and confirmed to be below 10 kΩ before testing began. Skin temperature was and recorded pre- and post-testing for each limb.

Surface decomposition EMG was collected using a 5-pin sensor (dEMG System, Delsys, Inc., Boston, MA) with an interelectrode distance of 5 mm, which results in 4 differential channels of data. The dEMG sensor was secured to the skin over the FCR and TA muscles directly adjacent to the sEMG electrodes and affixed with tape and a strap
around the limb to maintain slight pressure on the skin. An additional ground electrode was placed on the olecranon process (forearm testing) or the patella (lower leg testing). The dEMG signals were amplified 1,000 times and filtered between 20 and 450 Hz to maximize the resolution of the 16-bit analogue-to-digital converter before digitization at 20,000 Hz (Bagnoli-16 and EMGworks 4.2, Delsys, Inc., Boston, MA). The force signals from the load cells (MB-100 and SSMH, Interface Inc., Scottsdale, AZ) were sampled without amplification or filtering through the Bagnoli-16 A/D board at 20,000 Hz as well as the DI-720 A/D board at 2,000 Hz.

**Testing protocol**

The order of testing (upper versus lower, dominant versus non-dominant) was balanced across participants and groups, and kept consistent across sessions for each participant. Testing for each limb was performed as follows (see Figure 2 for protocol).

To begin, a series of three maximal M-waves were elicited from the median (for FCR stimulation) or common fibular nerve (for TA stimulation) while the participant was at rest with a 1 ms square-wave pulse using a hand-held stimulation probe (Grass S88 Stimulator and SIU8T, Astro-Med Inc.). Participants then performed three isometric MVCs of the agonist muscles (wrist flexion and dorsiflexion) each lasting 4-seconds in duration with 2-minute inter-trial rest periods. Participants were instructed to contract ‘hard-and-fast’ and hold their maximum steady, which was assisted by a force trace presented to participants on an oscilloscope. An interpolated twitch was evoked in the middle of each contraction at a supramaximal stimulation level corresponding to approximately 110% of the M-wave stimulation level.
Participants performed three isometric MVCs in the ‘opposite’ direction (wrist extension and plantar flexion contractions) lasting 4-seconds in duration with 2-minute inter-trial rest periods. Participants then performed a 6-second ramp contraction (wrist flexion or dorsiflexion) to 20% MVC force, which was completed as a signal quality check for the dEMG electrodes. Three isometric ramp contractions to 60% MVC force were performed with 2-minute intertrial rest periods. The ramps increased at 10% MVC per second for a rise of 6-seconds, held a steady plateau at 60% MVC for 6-seconds, and decreased force at 10% MVC per second for a drop of 6-seconds. A computer screen placed in front of the participants displayed a ‘target’ trapezoidal ramp contraction, which participants were asked to follow with their force trace resulting in concurrent knowledge of results (KR).

During the retention test only (12-week test), the ramp contractions were performed without feedback (noKR) to serve as a retention test. Three blinded ramps (screen turned off) were performed immediately after the three contractions with feedback. Participants were reminded of their goal to increase force to what they believed to be 60% of maximal strength over 6-seconds, hold it steady for 6-seconds, and release consistently for 6-seconds. The timing was counted out for the participants as follows “ramp up-5-4-3-2-1, hold steady-5-4-3-2-1, ramp down-5-4-3-2-1.” No feedback from the investigator was given between trials or between limbs.
Figure 2. Testing protocol including evoked potentials, maximal voluntary contractions (MVCs) of the wrist flexors and dorsiflexors with an interpolated twitch, MVCs of the wrist extensors and plantar flexors, and 60% trapezoidal contractions of the wrist flexors and dorsiflexors.

Training

Training was performed over a period of 6 weeks outside the laboratory and included dynamic contractions performed using a pulley cable (for dorsiflexion) or a dumbbell (for wrist flexion) set at 80% of the participants MVC force. Participants trained 4 times per week, completing three sets of 10-12 reps per session. At week three, participants returned to the lab to complete three sets of three MVCs on the trained limb in order to increase the prescribed weight to reflect the participant’s current strength (new 80% MVC weight). To minimize mirror contractions participants were instructed to focus on the dominant limb while resting the non-dominant limb with their knee flexed and foot resting on the floor (for dorsiflexion contractions) or with their elbow bent and their hand resting in their lap (for wrist flexion contractions). Participants were instructed not to specifically train the contralateral (non-dominant) side or the alternate muscle group (i.e., dorsiflexors if in ATG and wrist flexors if in LTG), nor begin any new training regimen over the course of their
study involvement. During detraining, participants were instructed to resume normal activity, with the exception of specifically training the wrist flexors or dorsiflexors.

Data reduction

Force and root-mean-square (RMS) amplitude of the sEMG signal were calculated from a 1-second window at the center of the MVC, terminating before the interpolated twitch where applicable. Agonist RMS sEMG was then normalized to the peak-to-peak amplitude of the corresponding M-waves evoked at rest (RMS/Mmax). Surface EMG activity of the limb at rest, was obtained from the same data window, to monitor postural stabilizing activity. The amount of co-contraction during ramp contractions was calculated as the antagonist activity normalized to its RMS amplitude when the muscle was maximally contracting as an agonist [14]. The amount of cross talk between muscles was measured from the M-wave peak-to-peak amplitude present in the antagonist and by using the cross-correlation between agonist and antagonist sEMG during an MVC.

Wrist flexion and dorsiflexion MVCs included twitch interpolation for the purpose of identifying V-waves and calculating central activation. V-waves were identified from the sEMG signal as a distinct wave occurring approximately 20-50ms following the M-wave, as evoked by the twitch interpolation. If no V-wave could be identified on a minimum of 2 of the 3 contractions, that limb was removed from the V-wave analysis. The V-wave was expressed as a ratio to the immediately preceding Mmax (V/M ratio) and averaged for the identified contractions. The central activation ratio (CAR) was calculated as the force increase obtained by the interpolated twitch compared to the maximal voluntary force using the formula \[\text{CAR} = 1 - \left(\frac{\text{interpolated twitch force}}{\text{maximal voluntary force}}\right) \times 100\] [34,41], where maximal voluntary force was the mean force taken from 1-second
immediately prior to the interpolated twitch, and interpolated twitch force was force increase resulting from the stimulation.

Motor units from the 60% ramp contractions were decomposed using the Precision Decomposition Algorithm III in the dEMG Analysis software (version 1.1, Delsys, Inc., Boston, MA), which identifies each motor unit and the firing instances [42–44]. The decomposition results were then validated using a built-in validation software employing Decompose-Synthesize-Decompose-Compare technique [44,45]. The MUFRs were calculated as the inverse of the inter-pulse interval and smoothed using a Hanning window of 0.95 seconds. The firing rate for each motor unit was then calculated as the mean firing rate occurring in a 1-second window at the center of the plateau portion (60% maximal force) of the ramp contraction. The coefficient of variation (σ / μ × 100) was calculated for each MUFR. Motor unit firing rates were included in the analysis if they met the following criteria: (1) the motor unit firing instances met a minimum decomposition accuracy of 90%; (2) the trial had a minimum of 5 motor units with >90% decomposition accuracy; and (3) the motor unit firing rate had less than a 20% coefficient of variation.

The variance ratio was calculated for each set of three ramp contractions, which determines the reproducibility of the forces traces, on a point-by-point basis [46]. By interpolating the force traces to the identical number of points, the variance ratio measures how consistently the shape of each ramp is performed [6]. This measures the variability of the motor pattern between trials, with a lower score indicating less variability, thus a comparison of identical trials would produce a variability ratio score of zero. The root-mean-square-error (RMSE) was calculated as the standard deviation during the plateau
portion of each contraction. It was normalized to the average force level to account for strength differences, effectively resulting in the coefficient of variation of the ramp plateau.

Statistics

All statistical analyses were performed in SAS (SAS 9.4, SAS Institute Inc., Cary, NC). The data were first screened to determine if they met the statistical assumptions prior to performing analysis of covariance (ANCOVA) procedures. When normality was violated, appropriate transformations were performed. Preliminary analyses were also performed to determine if there were any significance differences between males and females. There were no significant sex × session interaction terms, so the data were collapsed for further analyses. Significant differences were evaluated using a two-way repeated measures ANCOVA with one between factors (experimental versus control) and one within factor (baseline, post-training, retention). Baseline scores were used as a covariate to subtract out initial discrepancies in strength. Further, for measures calculated during the 60% ramp contractions, there were 4 sessions included in the analysis to examine the ramp contractions performed with no feedback during the retention: baseline, post-training, retention (feedback), retention (‘no feedback’).

To assess the training adaptations the following ANCOVAs were performed: (1) trained arm of the ATG versus dominant control arm of the LTG; and (2) trained leg of the LTG versus dominant control leg of the ATG. Similarly, to assess cross education in the untrained limb the following ANCOVAs were performed: (1) untrained arm of the ATG versus non-dominant control arm of the LTG; and (2) untrained leg of the LTG versus non-dominant leg of the ATG. Contrasts were performed using least squares means with a Bonferroni adjustment to compare the difference between sessions (baseline to post-
training, and baseline to retention), as well as the magnitude of training adaptations between the experimental limbs (trained versus untrained) at each session. Effect sizes were calculated according to Cohen [47], for each limb separately across time points (baseline to post-training, baseline to retention, and baseline to ‘no feedback’ where applicable). As suggested by Cohen [47], $d = 0.2$ is considered a small effect, 0.5 a medium effect, and 0.8 a large effect.

For measures calculated during the ‘no feedback’ contractions (force variability and antagonist co-contraction) a separate ANOVA was performed. Since it was expected that the ‘no feedback’ contractions would result in large changes from baseline for all groups, the ANOVA was run to compare the percent change score from baseline to ‘no feedback’ between groups (experimental versus control).

To determine the correlation between neuromuscular adaptations and strength improvements due to training and cross education, correlations were calculated using the repeated measures analysis of variance technique [48] for force of the trained or untrained limb and the neuromuscular measures (RMS/M, MUFRs, V/M ratio, CAR).

The contralateral transfer of strength in the experimental limbs (E: trained or untrained) is presented as percent strength gain accounting for the control limbs (C: dominant or non-dominant) according to the following formula [10]:

$$\frac{E_{Post} - E_{Baseline}}{E_{Baseline}} - \frac{C_{Post} - C_{Baseline}}{C_{Baseline}}$$

The strength gain magnitude was compared between upper and lower limbs and between trained and untrained limbs using change scores:

$$Post\ Gain = \frac{Post - Baseline}{Baseline} \quad Retention\ Gain = \frac{Retention - Baseline}{Baseline}$$
The strength gain in the upper and lower limbs was compared using an ANOVA, with post hoc testing (least square means) to examine the limbs at post-training and at retention. The strength gain in the trained and untrained limbs was examined using a regression (X: trained by Y: untrained) separated by upper and lower limbs and by post-training and retention testing.

**Results**

*Methodological Controls*

Participant demographics are detailed in Table 1. Resting limb EMG activity was < 3% of maximal activity recorded during MVCs. Cross talk, as measured from the M-waves, was approximately 9% of the agonist activity being recorded in the antagonist signal. Cross talk, as examined in the flexion MVCs using the cross correlation function, was less than 1%. Skin-electrode impedance and skin temperature stayed consistent within and between sessions.

Across limbs the average change in wrist flexion and dorsiflexion strength from familiarization to baseline testing was a 6% increase, while the agonist activity increased by 3%. The wrist extension and plantar flexion MVCs increased by 10% from familiarization to baseline. The V/M ratio increased 8%, but central activation increased less than 1% due to the high initial levels of central activation achieved in the familiarization session (97 ± 3.5%). The initial motor learning was evident in the large decreases in VR (up to 100%) and RMSE (up to 41%).

The following results are presented for the trained dominant limb (‘trained’), the contralateral non-dominant limb (‘untrained’), the dominant limb of the alternate group (‘dominant control’), and the non-dominant limb of the alternate group (‘non-dominant..."
The F-ratios of each ANCOVA are presented in text for the main effect of sessions (Baseline, post-training, retention) for each group. To further examine significant main effects, contrasts examining baseline to post-training, and baseline to retention were performed and are presented in the tables and figures. All values presented are calculated from the least squares means. Percent change scores and effect sizes are presented in Tables 2 (upper limb) and 3 (lower limb).

Table 1. Participant demographics, baseline strength, and training compliance.

<table>
<thead>
<tr>
<th></th>
<th>Arm Training</th>
<th>Leg Training</th>
</tr>
</thead>
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<tr>
<td></td>
<td>N = 20</td>
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<tr>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
<td></td>
</tr>
<tr>
<td>Age (y)</td>
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<td>23 ± 2</td>
</tr>
<tr>
<td>Height (cm)</td>
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<tr>
<td>Weight (kg)</td>
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<tr>
<td>Strength (N)</td>
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<td></td>
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<tr>
<td>Dominant Arm</td>
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<tr>
<td>Non-dominant Arm</td>
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<td>121.3 ± 33.0</td>
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<tr>
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<tr>
<td>Non-dominant Leg</td>
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</tr>
<tr>
<td>Compliance (%)</td>
<td>96 ± 6</td>
<td>93 ± 7</td>
</tr>
</tbody>
</table>

*Significant difference between groups (p < 0.05). §Strength is average force from baseline MVCs. ¶Arm strength for the leg-training group is calculated from a subset of participants (N=12) due to a slight change in equipment configuration.

Strength

Maximal strength significantly increased in the trained arm (F_{(2,75)} = 25.27, p < 0.01), trained leg (F_{(2,76)} = 12.45, p < 0.01), untrained arm (F_{(2,76)} = 10.54, p < 0.01) and untrained leg (F_{(2,76)} = 8.91, p < 0.01). The training stimulus resulted in strength increases of 24% (d = 0.84) at post-training and 16% (d = 0.67) at retention in the trained arm, 17% (d = 0.69)
at post-training and 17% ($d = 0.70$) at retention in the trained leg. Cross education resulted in strength increases of 6% ($d = 0.32$) at post-training and 15% ($d = 0.45$) at retention in the untrained arm, and 13% ($d = 0.40$) at post-training and 14% ($d = 0.58$) at retention in the untrained leg as demonstrated in Figure 3. There was no increase in any of the control limbs ($p$’s > 0.05).

Strength was compared between the trained and untrained limbs using contrasts at each session. At baseline there was no difference between trained and untrained strength in the arms ($p = 0.95$) or legs ($p = 0.98$). At post-training the trained and untrained arms were significantly different ($p < 0.01$) demonstrating that the trained arm increased strength significantly more than the untrained arm. At post-training the trained and untrained legs were not significantly different ($p = 0.35$), indicating that the magnitude of cross education was equal to the magnitude of training. At retention testing, the trained and untrained arm strength had equalized ($p = 0.84$), as did the trained and untrained leg strength ($p = 0.79$).
The gain scores between upper and lower limb strength (as reported above) were compared for the trained and untrained limbs. There was no significant difference in the limb (upper vs lower) × session (post gain and retention gain) interaction for the trained ($F_{(1,38)} = 1.72, p = 0.20$) or the untrained ($F_{(1,38)} = 0.41, p = 0.53$) limbs.

The strength gain in the trained (X) versus untrained (Y) limb are presented in Figure 4. The regression analysis demonstrated a significant correlation between trained and untrained strength for the lower limb at post-training ($R = 0.73, p < 0.01$) and retention ($R = 0.89, p < 0.01$). However, the correlation was not significant in the upper limb at post-training ($R = 0.17, p = 0.47$) or retention ($R = 0.29, p = 0.21$).
Extensor muscle strength (wrist extension and plantar flexion) was measured to assess the specificity cross education. Wrist extension strength increased for the trained arm ($F_{(2,76)} = 4.63, p = 0.01$) and untrained arm ($F_{(2,76)} = 7.88, p < 0.01$), but also increased for the dominant control arm ($F_{(2,76)} = 3.26, p = 0.04$) and the non-dominant control arm ($F_{(2,76)} = 5.43, p < 0.01$). Plantar flexion strength also increased for the trained leg ($F_{(2,75)} = 14.88, p < 0.01$) and untrained leg ($F_{(2,76)} = 6.02, p < 0.01$), but also increased for the dominant control leg ($F_{(2,75)} = 4.95, p < 0.01$) and non-dominant control leg ($F_{(2,76)} = 4.47, p = 0.01$).
**Agonist EMG Amplitude and Motor Unit Firing Rates**

The agonist RMS/Mmax ratio, demonstrated in Figure 5, significantly increased in the trained leg ($F_{(2,76)} = 16.19, p < 0.01$), with an increase at post-training (33%, $d = 0.78$) and retention (40%, $d = 0.89$). There was no significant session main effect in the trained arm ($F_{(2,76)} = 1.77, p = 0.20$) despite increases of 23% ($d = 0.68$) at post-training and 15% ($d = 0.39$) at retention testing. The session main effect was not significant for the untrained arm ($F_{(2,76)} = 0.81, p = 0.45$) with small increases at post-training (6%, $p = 0.55, d = 0.16$) and retention (13%, $p = 0.21, d = 0.34$). Although not significant, the effect sizes in the untrained arm are larger than in the non-dominant control arm ($d = -0.04 – 0.14$). The session main effect approached significance in the untrained leg ($F_{(2,76)} = 2.74, p = 0.07$), with small increases in TA activity at post-training (15%, $p = 0.051, d = 0.43$) and significantly at retention (15%, $p = 0.04, d = 0.46$). There was no increase in any of the control limbs ($p$’s > 0.05).
Figure 5. Least squares means (error bars: SE) of the agonist root-mean-square activity of the flexor carpi radialis (upper) and tibialis anterior (lower) calculated from the maximal voluntary contractions at baseline, post-training, and retention.

*Significance between experimental and control limb (p < 0.05); N = 20.

There was no significant change in the MUFRs of the trained arm (F(2,76) = 0.06, p = 0.94), trained leg (F(2,76) = 1.03, p = 0.36), untrained arm (F(2,76) = 0.46, p = 0.64) or untrained leg (F(2,76) = 0.97, p = 0.38). The average MUFR was 16.30 (SD 2.04) in the FCR and 15.09 (SD 2.06) in the TA, with an average change over sessions of 3.6%.

**Evoked contractions**

Interclass correlation analysis revealed that the V/M ratio was unreliable for the upper limb (ICC’s < 0.34), and therefore was only calculated for the lower limb (ICC’s > 0.80). V-waves could be identified on 16 participants in the trained and untrained limbs, and 19 participants in the dominant and non-dominant control limbs. There was a significant increase in V/M ratio in the trained leg (F(2,66) = 8.70, p < 0.01) and the untrained leg (F(2,66) = 3.97, p = 0.02) as illustrated in Figure 6. At post-training the V/M
ratio increased 59% \((d = 1.11)\) in the trained leg and 41% \((d = 0.65)\) in the untrained leg. This increase was primarily retained following detraining in the trained \((44\%, d = 1.08)\) and untrained legs \((34\%, d = 0.57)\). Neither of the control legs had a significant session main effect \((p’s > 0.05)\); however, the V/M ratio in the dominant control leg was significantly higher at post-training than baseline \((F_{(1,66)} = 4.29, p = 0.04)\). Although significant, the V/M ratio increase in the dominant control limb \((29\%, d = 0.47)\) was smaller than that of the trained or untrained limb, and returned to baseline values by retention testing.

![Figure 6. Least squares means (error bars: SE) of the V-wave to M-wave (V/M) ratio at baseline, post-training, and retention. *Significance between experimental and control limb \((p < 0.05)\); for the trained and untrained limbs N = 16, for the control limbs N = 19.]

Central activation in the lower limb was, on average, 99.7% at baseline. Therefore, the room for improvement was extremely limited \((< 0.3\%)\) so this variable was excluded. The upper limb demonstrated a slight dominance effect at baseline, with an average central activation ratio (CAR) of 97% in the dominant arm compared to 95% in the non-dominant arm as shown in Figure 7. There was a significant increase in the trained arm \((F_{(2,75)} = 5.22, p < 0.01)\) from 97.5% CAR at baseline to 98.5% CAR at post-training and retention, and in the untrained arm \((F_{(2,75)} = 4.92, p < 0.01)\) from 95.0% CAR at baseline to 96.1% and
96.6% at post-training and retention, respectively. There was no increase in any of the control limbs (p’s > 0.05) with an average change over sessions of 0.15%.

*Correlations*

To determine the relationship between force gains and neuromuscular adaptations in the trained and untrained limbs, repeated measures correlations ($r_{rm}$) were calculated from a repeated measures ANOVA. The correlation between force and RMS amplitude normalized to M-wave amplitude was poor in the trained ($r_{rm} = 0.17, p = 0.28$) and untrained ($r_{rm} = 0.13, p = 0.41$) arms. The relationship was slightly better in the lower limb with a significant correlation in the trained ($r_{rm} = 0.60, p < 0.01$) and untrained ($r_{rm} = 0.32, p = 0.04$) legs. The V/M ratio was significantly correlated with force gains in the trained ($r_{rm} = 0.66, p < 0.01$) and untrained ($r_{rm} = 0.59, p < 0.01$) legs. The CAR was also significantly correlated with force gains in the trained ($r_{rm} = 0.45, p < 0.01$) and untrained ($r_{rm} = 0.68, p < 0.01$) arms. The correlation between strength gains and changes in MUFs was poor ($r_{rm} = 0.04 – 0.19$) and non-significant ($p > 0.05$) for all limbs.
**Variability and Co-contraction of Ramp Contractions**

There was no consistent pattern of change in the antagonist co-contraction and no significant changes in the untrained limb at any session, as assessed by ANCOVAs ($p$'s > 0.05). As a result, the motor skill findings will focus on the variance ratio and RMSE. The only significant changes in the variance ratio at post-training or retention occurred in the trained leg at retention (-28%, $p = 0.03$), and untrained leg at post-training (-30%, $p = 0.03$). The RMSE demonstrated no significant changes in any limb at post-training or retention testing ($p$'s > 0.05). As expected, there was an increase in variability during the ‘no feedback’ contractions for all limbs due to the removal of concurrent feedback, therefore the percent changes (baseline to ‘no feedback’) were statistically compared (ANOVA) between experimental (trained or untrained) and control (dominant or non-dominant) limbs. The force variability measures calculated during retention contractions without feedback (‘no feedback’) are presented in Figure 8 as a percent change from baseline. A smaller increase relative to baseline indicates a greater amount of learning.

Overall, the variance ratio and RMSE had greater increases in the upper limb than the lower limb, indicating that the wrist flexion ramp was a more difficult task without feedback than the dorsiflexion, and/or participants found it more difficult to estimate 60% of their maximum in wrist flexion. In the upper limb, the variance ratio did not indicate that there was any training or cross education adaptations contributing to motor skill improvement. However, the RMSE demonstrated a significantly smaller increase from baseline in the untrained (29%, $d = 0.66$) compared to the non-dominant control (97%, $d = 2.14$) arm, and a smaller, but non-significant increase in the trained arm (55%, $d = 1.27$) compared to the dominant control (71%, $d = 1.63$) arm. In the lower limb, the variance
ratio demonstrated a motor learning adaptation with a significantly smaller increase from baseline in the untrained (42%, \( d = 0.81 \)) compared to the non-dominant control (136%, \( d = 1.68 \)) leg, and a non-significantly smaller increase in the trained (83%, \( d = 1.11 \)) compared to the dominant control (151%, \( d = 1.50 \)) leg. The RMSE in the lower legs demonstrated a very consistent increase across all legs (18-25%, \( d = 0.45-0.68 \)).

Figure 8. Percent change from baseline to ‘no feedback’ calculated from least squares means of the variance ratio (left) and root-mean-square error (right) calculated from the force trace of the trapezoidal contractions; \( N = 20 \). Note: a lower percent change indicates better performance (i.e., less variability) during ‘no feedback’ contractions.

Discussion

The purpose of this study was to evaluate the neuromuscular mechanisms contributing to strength and skill improvements following a 6-week unilateral, strength training program. In addition, the persistence of neuromuscular adaptations was examined following 6-weeks of detraining. The unilateral training study successfully induced cross education strength gains, of magnitudes in line with previous work [9–11]. The
experimental design of the present study ensured that the contralateral strength gain was due to cross education of unilateral training. Carroll and colleagues [10] showed that the lack of a familiarization session over-estimated the cross education strength gain by up to 4%. In agreement, the present study found a strength increase from familiarization to baseline of 4% in the contralateral arm and 7% in the contralateral leg. Therefore, the exclusion of a familiarization session would have greatly over-estimated the cross education effect. Furthermore, the presence of a control group ensured that the strength gain was due to the cross education of unilateral training rather than a training effect due to repeated testing or increased familiarity with the testing equipment. The effect sizes of cross education at post-training and retention ($d = 0.32$-$0.58$) indicate a small to moderate effect, which has potential for clinically relevant gains.

Few studies have experimentally compared cross education between males and females, and between the upper and lower limb. Therefore, 20 males and 20 females were randomly allocated to an arm-training (wrist flexion) or leg-training (dorsiflexion) group. No sex difference was observed in the patterns of change, confirming the results of Hubal and colleagues [49]. To date, the authors are not aware of any unilateral training study that statistically compared the magnitude cross education between upper and lower limbs following matched training protocols. Two meta-analyses have pooled upper and lower limb data to determine the effect of limb on cross education. Munn and colleagues [9] estimated the cross education strength gain to be 3.8% in the upper limb compared to 10.4% in the lower limb, however, this difference was non-significant ($p = 0.16$). Alternatively, Manca and colleagues [11] found a significant ($p = 0.006$) difference between the estimated 9.4% strength gain in the upper limbs and the 16.4% strength gain in
the lower limbs. The present study demonstrated a slightly smaller contralateral strength gain at post-training in the upper limb (6%) compared to the lower limb (13%), but there was no significant difference between the magnitude of cross education in the upper and lower limbs. However, inspection of Figure 4 demonstrates that the magnitude of cross-transfer (strength gain from trained to untrained) in the lower limb is somewhat larger than in the upper limb.

Cross education has been demonstrated to be highly specific to the training contraction type [17,49,50] and to the homologous muscle [17,51–53]. Therefore, the magnitude of cross education demonstrated in the present study may have been underestimated due to the different contraction types used for training (dynamic) versus testing (isometric). The use of dynamic contractions was selected to be most applicable to everyday movement (i.e., activities of daily living) and logistical for rehabilitation programs, while isometric testing was selected for the purpose of recording motor unit firing rates. The specificity of training also applies to the task, flexion versus extension flexion contractions. An increase in extension force at post-training in all limbs was not likely a feature of training or cross education. Rather, the fewer number of total contractions for extension resulted in continued familiarization in the extension direction.

The correlations between contralateral strength gains and the neuromuscular mechanisms provide linkage between central adaptations and cross education. The discrepant findings in the correlation of RMS/Mmax and force between the trained and untrained limbs, as well as between the upper and lower limbs, follows the conflicting results of previous research. Interestingly, there was no significant correlation between FCR activity and wrist flexion force, but there was a significant correlation between TA
activity and dorsiflexion force in both the trained and untrained limbs. Assessment of FCR activity scatter plots reveals that participants with high RMS/M-wave amplitude at baseline demonstrated decreases at post-training and retention, indicating an inflated value at baseline. However, these participants were not removed as their values did not meet the threshold criteria for outliers. There was no significant correlation between motor unit firing rates and force for any limb due to the lack of change in motor unit firing rates. Alternatively, both V/M ratio and CAR demonstrated strong correlations with contralateral force, confirming the increase in central drive to the contralateral limb following unilateral training [16,52,54]. It is not surprising, that the neuromuscular adaptations mirrored the training and cross education force increases at post-training and retention.

There were small increases (6-15%) in the contralateral limb’s agonist activity at post-training; however, these were not accompanied by any change in MUFs. Previous cross education studies examining muscle activity have found ambiguous results, which are highly variable depending on the training paradigm. Similar to the present study, two studies investigating the change in motor unit firing rates following unilateral training found no significant change in either the trained or untrained limbs despite ipsilateral and contralateral strength gains [29,36]. This lack of change in MUFs may indicate a methodological limitation of recording firing rates during the ramps contractions (at 60% maximal force) rather than during the MVCs. Previous work by Kamen and Knight [55] found a significant increase in maximal contraction MUFs with familiarization despite no significant change in 10% and 50% force contractions. It is likely that small adaptations may have been present if contractions were performed at the same absolute force as baseline (i.e., 60% baseline force) rather than a percentage of the increased strength.
Despite a lack of adaptation in the MUFRs, the increase in agonist activity was accompanied by increases in central drive, as assessed by the V-wave [16,54,56]. Two previous cross education studies examining V-wave amplitude found no significant increase in the contralateral limb following training [16,24]. The present study demonstrated moderate to large increases in the V/M ratio in the untrained limb after 6-weeks of training (41%, $d = 0.65$) and persisting after 6-weeks of detraining (34%, $d = 0.57$). Similarly, the central activation ratio was used to evaluate the “completeness of skeletal muscle activation” [34,57,58], and quantify the amount of efferent drive to the muscle [30]. Previous work has demonstrated equivocal results in contralateral voluntary activation following unilateral training [30,32,33]. In the present study, the central activation ratio of the lower limb was already > 99% at baseline. However, the upper limb demonstrated moderate to large increases in the central activation ratio of the contralateral arm at post-training (1.2%, $d = 0.51$) and a continued increase to retention testing (1.7%, $d = 0.70$).

The persistence of strength and neuromuscular mechanisms following detraining in the present study is a novel finding in the field of cross education. Previous unilateral training studies (range 3-10 weeks training) which examined cross education following a period of detraining (range 2-12 weeks), all reported a loss in cross education strength following detraining [28,30,59–64]. Interestingly, the present study demonstrated a continued increase in contralateral strength following detraining. The continued force increase resulted in a total cross education strength gain of 18% for the contralateral arm, and 22% for the contralateral leg. This continued increase resulted in the untrained limb strength being equal to the trained limb strength at retention. This was accompanied by the
maintenance or continued increase of the neuromuscular adaptations (RMS/Mmax, V/M ratio, central activation ratio) in the contralateral limbs. This novel finding indicates the contribution of motor learning to cross education.

The permanency of motor learning was assessed during retention testing using force variability calculated during ‘no feedback’ (noKR) contractions. The variance ratio was calculated to evaluate the stability of motor performance by measuring the participant’s ability to reproduce a consistent motor pattern [7]. The RMSE was calculated to evaluate task performance by measuring the ability to match a target and calculating force steadiness across a targeted plateau [7]. Surprisingly, the variance ratio and RMSE did not significantly change in the trained and untrained limbs at post-training and retention. However, some evidence of motor learning in the untrained limb was present in the ‘no feedback’ contractions demonstrated in the RMSE for the untrained arm and in the variance ratio for the untrained leg, indicating that the contralateral limbs were able to reproduce the target task more consistently and steadily than the control limbs. The lack of improvement in variability at post-training and retention are likely due to the different contraction types used in training (dynamic self-paced to 80% force) versus testing (isometric ramps to 60% force). However, the presence of motor learning during the ‘no feedback’ contractions suggest that the cross education of skill is robust in the face of specificity.

The presence of motor learning following strength training demonstrates the skill component of strength training and the cross education of skill without a specific coordination task. These results suggest that the theories of cross education involving the bihemispheric ‘storage’ and inter-hemispheric access of motor engrams [2,3,65,66] may be
generalizable to tasks performed by the homologous muscle beyond the practiced exercise (i.e., contraction type). Additionally, the changes in strength and neuromuscular adaptations were similar between the trained and untrained limbs, providing support for the hypothesis that unilateral training induces adaptations of a motor center (such as the premotor cortex) that provides common drive to both hemispheres [3].

**Conclusion**

In the present study we experimentally evaluated the contributions of neuromuscular adaptations and motor learning to cross education with the following conclusions:

1. There was no difference between sexes or upper and lower limbs in the magnitude of training or cross education.
2. Cross education of strength totalled 15% in the untrained wrist flexors and 14% in the untrained dorsiflexors.
3. Agonist RMS amplitude, V-wave amplitude, and central activation ratio confirmed the neuromuscular adaptations associated with cross education.
4. There was no change in motor unit firing rates at 60% of maximal force.
5. A continued increase in contralateral strength at retention demonstrated the persistence of cross education following 6-weeks of detraining.
6. Force variability measures demonstrated the presence of motor learning in the contralateral limbs.
Table 2. Change scores calculated from least squares means of the upper limb. Strength, EMG amplitude, and central activation calculated from maximal voluntary contractions; MUFRs, variance ratio, and RMSE calculated from the ramp contractions to 60% maximal force. Variability is also reported for the ‘no feedback’ ramps performed during the retention testing.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Trained Arm</th>
<th>Dominant Control Arm</th>
<th>Untrained Arm</th>
<th>Non-Dominant Control Arm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Δ Post Raw (%)</td>
<td>Δ Retention Raw (%)</td>
<td>Δ Post Raw (%)</td>
<td>Δ Retention Raw (%)</td>
</tr>
<tr>
<td>Strength (N)</td>
<td>27.6 (27%)</td>
<td>19.7 (19%)</td>
<td>3.5 (3%)</td>
<td>3.3 (3%)</td>
</tr>
<tr>
<td></td>
<td>*d = 0.84</td>
<td>d = 0.67</td>
<td>*d = 0.07</td>
<td>d = 0.06</td>
</tr>
<tr>
<td>Agonist EMG (RMS/M Ratio)</td>
<td>0.017 (23%)</td>
<td>0.011 (15%)</td>
<td>-0.002 (-3%)</td>
<td>0.000 (0%)</td>
</tr>
<tr>
<td></td>
<td>*d = 0.68</td>
<td>d = 0.39</td>
<td>*d = -0.02</td>
<td>*d = -0.01</td>
</tr>
<tr>
<td>Central Activation (%)</td>
<td>0.90 (0.9%)</td>
<td>0.95 (1.0%)</td>
<td>0.21 (0.2%)</td>
<td>0.03 (0.0%)</td>
</tr>
<tr>
<td></td>
<td>*d = 0.64</td>
<td>d = 0.65</td>
<td>*d = 0.10</td>
<td>d = 0.02</td>
</tr>
<tr>
<td>MUFRs (pps)</td>
<td>0.05 (0%)</td>
<td>-0.11 (-1%)</td>
<td>0.78 (5%)</td>
<td>0.11 (1%)</td>
</tr>
<tr>
<td></td>
<td>*d = 0.03</td>
<td>d = -0.06</td>
<td>*d = 0.44</td>
<td>*d = 0.06</td>
</tr>
<tr>
<td>Variance Ratio</td>
<td>-0.002 (-12%)</td>
<td>0.001 (6%)</td>
<td>-0.003 (-17%)</td>
<td>-0.006 (-29%)</td>
</tr>
<tr>
<td></td>
<td>*d = -0.30</td>
<td>d = 0.10</td>
<td>*d = -0.30</td>
<td>*d = -0.37</td>
</tr>
<tr>
<td>Δ No Feedback</td>
<td>0.062 (385%), *d = 2.52</td>
<td>0.040 (198%), *d = 1.72</td>
<td>0.040 (170%), *d = 1.99</td>
<td>0.044 (179%), *d = 2.37</td>
</tr>
<tr>
<td>RMSE (%)</td>
<td>-0.20 (-7%)</td>
<td>0.03 (1%)</td>
<td>0.18 (6%)</td>
<td>-0.11 (-4%)</td>
</tr>
<tr>
<td></td>
<td>*d = -0.15</td>
<td>d = 0.03</td>
<td>*d = 0.23</td>
<td>*d = -0.02</td>
</tr>
<tr>
<td>Δ No Feedback</td>
<td>1.59 (55%), *d = 1.27</td>
<td>2.00 (71%), *d = 1.63</td>
<td>0.96 (29%), *d = 0.66</td>
<td>2.64 (97%), *d = 2.14</td>
</tr>
</tbody>
</table>

Δ Post: change from baseline to post-training. Δ Retention: change from baseline to retention (detraining). EMG: electromyography; RMS/M Ratio: the root-mean-square amplitude normalized to the M-wave peak-to-peak amplitude; MUFRs: motor unit firing rates; RMSE: root-mean-square error of the force trace. Cohen’s *d* is given for effect size of the change from baseline. N=20. *significant change from baseline (*p* < 0.05). ‡Significant difference between experimental and control (for No Feedback only).
Table 3. Change scores calculated from least squares means of the lower limb. Strength, EMG amplitude, and V/M ratio calculated from maximal voluntary contractions; MUFRs, variance ratio, and RMSE calculated from the ramp contractions to 60% maximal force. Variability is also reported for the ‘no feedback’ ramps performed during the retention testing.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Trained Leg</th>
<th></th>
<th>Dominant Control Leg</th>
<th></th>
<th>Untrained Leg</th>
<th></th>
<th>Non-Dominant Control Leg</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Δ Post</td>
<td>Δ Retention</td>
<td>Δ Post</td>
<td>Δ Retention</td>
<td>Δ Post</td>
<td>Δ Retention</td>
<td>Δ Post</td>
<td>Δ Retention</td>
</tr>
<tr>
<td></td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
<td>Raw (%)</td>
</tr>
<tr>
<td>Strength (N)</td>
<td>52.4 (20%)*</td>
<td>58.4 (22%)*</td>
<td>7.6 (3%)</td>
<td>15.0 (6%)</td>
<td>37.6 (15%)*</td>
<td>55.0 (22%)*</td>
<td>5.7 (2%)</td>
<td>19.3 (7%)</td>
</tr>
<tr>
<td></td>
<td>$d = 0.69$</td>
<td>$d = 0.70$</td>
<td>$d = 0.08$</td>
<td>$d = 0.15$</td>
<td>$d = 0.40$</td>
<td>$d = 0.58$</td>
<td>$d = 0.06$</td>
<td>$d = 0.19$</td>
</tr>
<tr>
<td>Agonist EMG (RMS/M Ratio)</td>
<td>0.026 (33%)*</td>
<td>0.032 (40%)*</td>
<td>0.007 (9%)</td>
<td>0.009 (12%)</td>
<td>0.011 (15%)*</td>
<td>0.012 (15%)*</td>
<td>0.003 (4%)</td>
<td>0.009 (11%)</td>
</tr>
<tr>
<td></td>
<td>$d = 0.78$</td>
<td>$d = 0.89$</td>
<td>$d = 0.30$</td>
<td>$d = 0.31$</td>
<td>$d = 0.43$</td>
<td>$d = 0.46$</td>
<td>$d = 0.15$</td>
<td>$d = 0.29$</td>
</tr>
<tr>
<td>V/M Ratio§</td>
<td>0.18 (59%)*</td>
<td>0.13 (44%)*</td>
<td>0.09 (29%)*</td>
<td>0.07 (22%)</td>
<td>0.14 (41%)*</td>
<td>0.11 (34%)*</td>
<td>0.06 (16%)</td>
<td>0.001 (0%)</td>
</tr>
<tr>
<td></td>
<td>$d = 1.11$</td>
<td>$d = 1.08$</td>
<td>$d = 0.47$</td>
<td>$d = 0.33$</td>
<td>$d = 0.65$</td>
<td>$d = 0.57$</td>
<td>$d = 0.24$</td>
<td>$d = 0.02$</td>
</tr>
<tr>
<td>MUFRs (pps)</td>
<td>-0.65 (-4%)</td>
<td>-0.05 (0%)</td>
<td>0.67 (5%)</td>
<td>0.68 (5%)</td>
<td>0.67 (5%)</td>
<td>0.09 (1%)</td>
<td>0.46 (3%)</td>
<td>0.04 (0%)</td>
</tr>
<tr>
<td></td>
<td>$d = -0.39$</td>
<td>$d = -0.02$</td>
<td>$d = 0.32$</td>
<td>$d = 0.30$</td>
<td>$d = 0.36$</td>
<td>$d = 0.05$</td>
<td>$d = 0.20$</td>
<td>$d = 0.02$</td>
</tr>
<tr>
<td>Variance Ratio</td>
<td>-$0.007 (-21%)*</td>
<td>-$0.009 (-28%)*</td>
<td>-$0.006 (-22%)*</td>
<td>-$0.006 (-20%)*</td>
<td>-$0.010 (-30%)*</td>
<td>-$0.003 (-9%)*</td>
<td>-$0.001 (2%)</td>
<td>-$0.006 (21%)*</td>
</tr>
<tr>
<td></td>
<td>$d = -0.25$</td>
<td>$d = -0.61$</td>
<td>$d = -0.13$</td>
<td>$d = -0.31$</td>
<td>$d = -0.81$</td>
<td>$d = -0.47$</td>
<td>$d = 0.05$</td>
<td>$d = 0.34$</td>
</tr>
<tr>
<td>Δ No Feedback</td>
<td>.028 (83%), $d = 1.11$</td>
<td>.042 (151%), $d = 1.50$</td>
<td>.014 (42%), $d = 0.81$</td>
<td>$\ddagger$</td>
<td>.041 (136%), $d = 1.68$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RMSE (N)</td>
<td>-0.19 (-5%)</td>
<td>-0.35 (-10%)</td>
<td>-0.37 (-12%)</td>
<td>-0.32 (-10%)</td>
<td>-0.33 (-9%)</td>
<td>-0.28 (-8%)</td>
<td>-0.46 (-13%)</td>
<td>-0.36 (-10%)</td>
</tr>
<tr>
<td></td>
<td>$d = -0.01$</td>
<td>$d = -0.11$</td>
<td>$d = -0.28$</td>
<td>$d = -0.28$</td>
<td>$d = -0.16$</td>
<td>$d = -0.12$</td>
<td>$d = -0.28$</td>
<td>$d = -0.19$</td>
</tr>
<tr>
<td>Δ No Feedback</td>
<td>.76 (22%), $d = 0.59$</td>
<td>.68 (22%), $d = 0.68$</td>
<td>.62 (18%), $d = 0.45$</td>
<td></td>
<td>.87 (25%), $d = 0.51$</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Δ Post: change from baseline to post-training. Δ Retention: change from baseline to retention (detraining). Δ No Feedback: change from baseline to retention (ramps performed without feedback). EMG: electromyography; RMS/M Ratio: the root-mean-square amplitude normalized to the M-wave peak-to-peak amplitude; V/M ratio: V-wave amplitude normalized to M-wave amplitude; MUFRs: motor unit firing rates; RMSE: root-mean-square error of the force trace. Cohen’s $d$ is given for effect size of the change from baseline. N=20. §V/M Ratio: N=16 for trained and untrained, N=19 for control legs. *Significant change from baseline ($p < 0.05$). ‡Significant difference between experimental and control (for No Feedback only).
References


Cross education is the improvement in strength or skill that is found in the contralateral limb following a unilateral training program or motor practice on the homologous limb. This phenomenon provides a potential rehabilitation model for unilateral injuries or disorders, including acute injuries or immobilization (casting) of a single limb, and neurologic disorders, such as stroke, affecting the body unilaterally. The cross education of skill (or skill transfer) literature primarily focuses on skilled tasks (i.e., goal-oriented to a target or timed), which are practiced and tested within a single session. While the cross education is typically defined by an increase in contralateral strength, skill transfer is defined by contralateral task improvement. Although both thoroughly examined, these phenomena are not examined concurrently due to the inherent differences in training (or practice) paradigms. However, as previously demonstrated for unilateral strength training, all resistive exercise includes a skill component, and therefore the potential for motor learning [1–3]. The combined benefits of skill acquisition and resistance training may be crucial for rehabilitation involving decrements in both function and strength.

The overall purpose of this thesis was to further the field of cross education by examining the neuromuscular adaptations contributing to the cross-body transfer of strength and skill, while employing the necessary methodological controls that have been underused and under-examined in previous works. A comprehensive review of literature was necessary to identify the literature gaps, which are primarily based on methodological and experimental design concerns. The neuromuscular adaptations of cross education have been thoroughly studied, but are plagued with methodological inconsistencies leading to an inability to compare across the literature. Therefore, manuscript 1 examined a novel surface
EMG electrode placement and configuration designed to allow for reliable recording of both voluntary and evoked contractions, which is necessary in the examination of cross education. Manuscript 2 was a meta-analysis of cross education literature for the purpose of identifying deficiencies in previous experimental designs and methodologies in order to best inform the specifics of a cross education study. Lastly, manuscript 3 was a unilateral training study designed to identify the neuromuscular adaptations of cross education and examine the previously unidentified presence of skill transfer following a strength training protocol.

7.1 Summary of Findings

Manuscript 1 demonstrates the reliable and effective placement and configuration of sEMG electrodes for evoked and voluntary contractions. The novel configuration consisted of placing one electrode on the electrically identified motor point and the second electrode directly adjacent in a bipolar configuration. The configuration was developed to reap the benefits of both monopolar and bipolar configurations as follows: (1) cross-talk is minimized by using differential recording [4,5]; (2) peak-to-peak amplitude of the CMAP is greatest over the motor point [6]; (3) electrode placement avoids the borders of small muscles [7,8]; and (4) the CMAPs retain an initial negativity without a leading positivity [9,10].

Both voluntary isometric wrist flexion contractions and evoked stimulations of the median nerve were performed to assess the flexor carpi radialis (FCR) activity using the novel configuration. We assessed the reliability of the peak-to-peak amplitude of the compound muscle action potential (CMAP), the root-mean-square (RMS) amplitude, and the mean power frequency of surface electromyographic recordings. In addition, antagonist
muscle activity was recorded from the extensor carpi radialis (ECR) to assess cross-talk. The normalization of RMS amplitude to CMAP peak-to-peak amplitude was also evaluated for reliability. Using the novel configuration, the cross-correlation coefficient was 0.12 and the common signal ($R^2_{xy}$) between FCR and ECR was 1.4%. The measures were highly reliable and consistent (ICC = 0.75 – 0.89) across the four test sessions. The normalization procedure slightly lowered the RMS amplitude intraclass correlation coefficient from 0.89 (non-normalized) to 0.85 (normalized to CMAP), but demonstrated that normalization using the novel configuration is reliable when necessary for repeated testing.

Both evoked and voluntary sEMG measures were reliable across multiple test sessions, which show the efficacy of the proposed electrode placement in cross education research. A bipolar electrode configuration may be used to minimize cross-talk during voluntary contractions while still retaining the undistorted CMAPs for experiments requiring both procedures. As cross education research requires both voluntary and evoked contractions to be performed simultaneously, and over multiple test sessions, the novel electrode configuration will allow for the reliable assessment of neuromuscular mechanisms.

Manuscript 2 provided a comprehensive estimation of the cross education effect and identified some of the methodological inconsistencies specific to cross education research. The field of cross education is long standing with a large breadth and depth of studies examining the cross-body transfer of unilateral training adaptations. However, meta-analyses in the field have included only a select number of studies. Upon examination, it was clear that methodological inconsistencies and strict inclusion criteria were the primary reason that a comprehensive literature review of the field did not exist. Furthermore, the
meta-analysis by Carroll and colleagues [11] identified a potential for over-estimation due to a lack of familiarization and experimental control groups. Therefore, the goal of this manuscript aimed to include as many cross education of strength studies as possible to identify the true impact of unilateral training, while identifying the methodological issues that plague the field.

A review of 90 studies resulted in 131 distinct unilateral training groups of young able-bodied participants (n = 118 units), older able-bodied participants (n = 7 units), and patient populations (n = 6 units). In the young group, the cross education strength gain was estimated to be 18%, which had a moderate effect size of 0.70 (95% CI: 0.58 – 0.83). This corresponded to approximately 67% of the direct training adaptation in the trained limb (29% gain; effect size = 1.14; 95% CI: 0.99 – 1.29). The older group had an average cross education strength gain of 17%, with a moderate effect size of 0.59 (95% CI: 0.17 – 1.01). The patient group had the largest cross education strength gain of 29% with a moderate effect of 0.76 (95% CI: 0.21 – 1.31). A review of control limbs identified the potential for a 3% over-estimation of the cross education strength gain.

Interestingly, the strength gain was not significantly different between groups that were familiarized and those that were not. This is likely due to the criterion of ‘familiarization’ which ranged from a single contraction to a full session. The robustness of the present meta-analysis allowed for the examination of factors involved in training program design. The meta-analysis identified: the improved efficacy of EMS training over voluntary modalities, the absence of a correlation between the amount of training stimulus and cross education, the equivalence in cross education between upper and lower limbs, and the transfer discrepancy between males and females.
The use of unilateral training of the unaffected limb is not prevalent in unilateral disorder rehabilitation, potentially due to the underestimation of the cross education effect and the lack of information for maximizing benefits. Manuscript 2 contributed to the rehabilitation field by examining training program factors for impact and efficacy. First and foremost, previous meta-analyses have greatly underestimated the cross education effect of an approximately 18% strength gain in the contralateral limb. This is likely due to the exclusion of electromyostimulation (EMS) training from previous meta-analyses, which proved to be the effective training methodology. Additionally, there was no correlation between the intensity of training (% maximal force) and the contralateral strength gain. These results can inform a more accessible rehabilitation program that may not require the heavy loads and voluntary contractions previously thought to be necessary. This work is imperative for informing future unilateral training programs for maximizing cross education in patient populations.

Manuscript 3 experimentally evaluated the neuromuscular adaptations contributing to the cross education of both strength and skill with the following conclusions. The novel contribution of this paper was the evaluation of motor learning (cross education of skill) following a unilateral resistive exercise training program. To accomplish this, force variability was assessed pre- and post-training, as well as during a retention session following 6-weeks of detraining and during ‘no feedback’ contractions. The cross education of strength was evident following 6-weeks of unilateral training as an 11% strength gain in the contralateral wrist flexors and a 15% strength gain in the contralateral dorsiflexors.
Increases in V-wave amplitude of the untrained leg and central activation ratio of the untrained arm at post-training demonstrated neuromuscular adaptations associated with cross education. These measures were highly correlated with the strength gains in both the trained and contralateral limbs, further confirming their contribution to training and cross education adaptations. This work helped clarify the equivalent V-wave adaptations found by Fimland and colleagues [12] and Colomer-Poveda and colleagues [13], and confirm the results of Tøien and colleagues [14]. We found a significant 41% increase in V/M ratio in the contralateral limb at post training. Despite increases in central drive, there was little change in contralateral agonist RMS amplitude, and no change in motor unit firing rates obtained at 60% of maximal force. This lack of change in MUFRs may be due to the recording of firing rates during the submaximal contractions (at 60% maximal force) rather than during the MVCs. Kamen and Knight [15] found a significant increase in maximal contraction MUFRs with familiarization despite no significant change in 10% and 50% force contractions. The results of this study confirm the neuromuscular adaptations identified in previous works, as estimated following familiarization and compared to an experimental control group.

A novel finding was the continued increase in strength following 6-weeks of detraining such that the total cross education was 18% in the wrist flexors and 22% in the dorsiflexors. This persistent gain indicated the presence of motor learning, which was supported by the force variability results. There was no difference in force variability at post-training and retention; however, the variance ratio and root-mean-square error calculated during the ‘no feedback’ contractions at retention testing demonstrated improved transfer of motor learning in the contralateral limb compared to the control limb. These
results suggest that improvements in motor skill performance were transferred from the trained to the untrained limb, and likely underestimate the amount of motor learning since training was performed using dynamic contractions and force variability was assessed during isometric contractions. Furthermore, these results indicate the importance of using a transfer test (e.g., contractions performed without knowledge of results (noKR)), since the presence of motor learning was only discernable during the ‘no feedback’ contractions.

The assessment of cross education following a period of detraining was designed to evaluate the permanency of adaptations and the presence of motor learning. The delayed retention of a skill demonstrates the ability to retrieve the learned movement, indicating that consolidation of motor memory has occurred [16,17]. The stages of learning are often separated into fast and slow, as characterized by the activation patterns and blood flow to different cortical regions [18]. The ‘fast’ stage of learning is most often examined in motor learning literature within a single session; however, the timelines are highly dependent on the complexity of the task [18,19]. Lehéricy and colleagues [20] demonstrated that blood oxygenated level dependent (BOLD) signal changes present in early learning (day 1) of a finger movement sequence continued to persist at 4-weeks of training. Although it is likely that the contralateral strength increase at retention we reported is due to the consolidation of the motor task during 6-weeks of detraining, it may also be possible that the ‘early’ stage of learning was not completed during the training period.

The presence of motor learning has been assessed via representational organization in the motor cortex following strength and skill training in rats. Remple and colleagues [21] examined forelimb representations (activation patterns) in the motor cortex following skill training (reaching for 1 pasta strand), strength training (reaching for pasta bundles of
increasing sizes), and control (no reaching) conditions. The ‘strength trained’ rats increased their maximal strength (bundle size) over 4-weeks, and both the strength and skill reaching groups increased their accuracy. Interestingly, the alteration in forelimb representations was nearly identical between the skill and strength groups. The authors concluded that the movement representations within the motor cortex were sensitive to the skill component of the task but not the strength component, since the strength training group showed no additional adaptations in movement representations [21]. This suggests that the skill component of dynamic training likely induced the motor learning adaptations demonstrated in manuscript 3, and that a training program with greater skill demands would likely enhance the neural adaptations associated with strength and skill transfer.

This dissertation provided insight and future directions for cross education, in both experimental research and applied rehabilitation. By systematically reviewing the field, numerous methodological issues in cross education experimental research were identified and examined with suggestions for future research. First and foremost is the requirement for randomized, controlled research designs and the inclusion of a familiarization session. A control group is necessary to account for the potential gains that could result from testing alone. This is evidenced by the 6% strength gain from session 1 to session 2 (familiarization), and further 3% strength gain at post-testing found in manuscript 3. This also indicates the need for familiarization, not only to control for pre-training strength increases present from session 1 to 2, but also to control for the improvement in task performance as evidenced by large decreases in force variability from session 1 to 2.

A second methodological issue is the collection and reporting of neuromuscular results. For example, the large range in percent adaptations in agonist muscle activity is
likely due to the large discrepancies in data analysis, namely the selection of averaging and
the use of normalization. Manuscript 1 details a novel configuration ideal for simultaneous
recording of voluntary and evoked contractions as necessary in cross education research.
The configuration proved to be reliable in both contraction types, while limiting the volume
of cross talk and maintaining the evoked potential shape and amplitude. The pros and cons
of RMS amplitude to CMAP normalization were also identified, and although not
recommended in the ideal situation, it is demonstrated that normalization can be reliable
when required.

Lastly, this dissertation provides insight for designing a rehabilitation program for
unilateral training of the unaffected limb to improve strength and motor performance on the
affected limb. Most notably, the presence of motor learning in the contralateral limb
compared to the control limb indicates that motor skill improvements can be gained in the
contralateral limb with resistive exercise training and should be further investigated. The
dynamic exercise training prescribed in manuscript 3 had a skill component, as
demonstrated by improvements in force variability, however, skill transfer could likely be
enhanced by increasing the skill component of the rehabilitation program. Further, the
results of the meta-analysis indicate that a more accessible training programme (e.g., EMS
training or lower intensity contractions) will provide the same benefits of cross education
for patient populations.

7.2 Limitations

The primary advantage and limitation of this dissertation is the examination of motor
unit firing rates using surface electromyography. The novel Delsys dEMG system allows
for the identification of motor units through surface electromyography. Surface EMG
recordings are superior to indwelling in pick-up volume, ease of use, and participant comfort. To date, only two studies have examined motor unit firing rates (MUFRs) in the contralateral limb following unilateral training [22,23]. Both studies employed indwelling EMG during submaximal [22] or maximal [23] contractions and found no increase in contralateral MUFRs at post-training. It is surprising that no cross education study has attempted to examine MUFRs since. The use of sEMG for motor unit decomposition allowed us to reliably examine MUFRs in a large sample of participants across multiple days.

Our results confirmed those of the previous work, with no change in the trained or contralateral MUFRs occurring. However, there are limitations to the use of sEMG decomposition for the evaluation of MUFRs. The lack of change in MUFRs found in manuscript 3 may indicate a methodological limitation of recording firing rates during the ramp contractions (at 60% maximal force) rather than during maximal force. Previous work by Kamen and Knight [15] found a significant increase in maximal contraction MUFRs with familiarization despite no significant change in 10% and 50% force contractions. Additionally, the validation of surface decomposition has been criticized. Therefore, further examination is necessary to determine the validity of the MUFRs recorded from sEMG.

The cross education of skill following strength training was a novel contribution of manuscript 3; however, the discrepancy between training and testing modalities may have impeded the ability to fully examine skill transfer. The equivocal results in the variance ratio and RMSE during the no feedback contractions were likely due to the differences between the ‘testing’ task (isometric trapezoids to 60% force) compared with the ‘practice’
task (dynamic self-timed contractions at 80% force). Additionally, motor learning studies typically track the practice effects (e.g., continuous testing within and between sessions) to create a learning curve, but the present study was limited to testing at pre- and post-training. Future work should examine the learning curve of the trained limb to identify a performance plateau, however, the nature of cross education limits testing of the contralateral limb in order to minimize any ‘direct’ training effect due to repeated testing, thereby limiting the ability to track the cross education of skill.

7.3 Future Directions

The present work has identified the experimental design and methodological controls necessary for the evaluation of cross education. Therefore, the primary future direction of the present work is the inclusion of patient populations. To date, only a few studies have examined cross education in patient populations with limited success. However, the differentiation between statistically significant and clinically significant indicated promising results of unilateral training in the unaffected limb. The identification of more accessible training options (e.g., EMS training) should be examined in able-bodied and patient population for the motor learning and neuromuscular adaptations. It’s possible that the addition of EMS to voluntary contractions enhances the cortical and spinal adaptations of cross education.

In a comprehensive analysis of the cross education literature the overwhelming majority of studies use isokinetic, isometric, and dynamic exercises for unilateral training. Although all resistive exercises have a skill component, it’s possible that unilateral coordination training may be more beneficial to the improvement in contralateral function than high-intensity contractions. The strength gains from resistance training are highly
specific to the joint angle, contraction type, and velocity [24–26]. Therefore, resistance training is ideal in the early stages of a rehabilitation program when the primary goal is a return in strength [27], but should be supplemented with coordination or proprioception training in order to improve functional activities [28,29]. A future direction is the development of a training program that incorporates high-intensity contractions with coordinated movements for the maximization of motor learning adaptations.
References


APPENDIX A

Pilot Study for Manuscript 3

Methodology

Twenty (10 male, 10 female) healthy college-aged (18-30 years) participants volunteered for the pilot study. Prior to study involvement, participants signed a consent form outlining all methods and materials as approved by Brock University Research Ethics Board. The experimental setup and measures are described below.

Protocol

The protocol was performed for both the dominant wrist and ankle for each participant. To begin, five maximal M-waves were elicited from the agonist muscle. After 2 minutes of rest, participants performed three maximal isometric voluntary contractions (MVC) with twitch interpolation. Each MVC lasted 3 seconds in duration and were separated by 2 minutes of rest. Participants were encouraged to contract “hard and fast” to maximum force and then maintain a steady trace. Twitch interpolation was performed in the middle of each MVC. An average force level from a steady plateau during the MVCs was used to determine force levels for subsequent contractions. After 2 minutes of rest, participants completed a 5-second contraction at 20% MVC force for signal quality analysis of the dEMG sensors. This was performed one to three times based on necessity, if multiple trials were needed a minimum of 1 minute rest was given. Following the signal quality check, participants performed ramp contractions to 80% MVC force. The ramp up was 8 seconds in duration (rate of 10% force per second), followed by a 4 second plateau held at 80% force, then an immediate release to relaxation.
Data Analysis

The unfiltered mean force, RMS amplitude, and MPF of the sEMG signal were calculated from a 500-ms window before the center of each MVC contraction (Farina and Merletti 2000). V-waves were identified from the sEMG signal as a distinct wave occurring approximately 20-50ms following an M-wave during the twitch interpolation. If no V-wave could be identified on a minimum of 2 of the 3 contractions in a session, that participant was removed from the V-wave analysis for a total of 7 removed from the wrist flexor V-wave analysis and 8 removed from the dorsiflexor V-wave analysis.

Motor unit firing rates (MUFs) were obtained from the 4-second plateau at the 80% MVC force level of the ramp contraction. Motor units were decomposed using EMGworks (v4.1.7, Delsys, Inc., Boston, MA), which identified each motor unit and the firing instances. The decomposition results were then validated using the validation software (v42, Delsys, Inc., Boston, MA). Motor units meeting the criteria of 85% accuracy were included in the analysis. This resulted in the exclusion of 23% of wrist flexion contractions and 32% of dorsiflexion contractions being excluded due to a lack of motor units meeting the criteria. Therefore, the reliability of the motor unit firing rates was performed on participants that had accurate MUs for 2 trials occurring on 2 sessions rather than 3 trials across 3 sessions. Using a 2 x 2 model, there were 18 participants included for wrist flexion MUFs and 16 participants included for dorsiflexion MUFs.

Statistics

An intraclass correlational analysis of variance was performed for each measure to assess its reliability across the three testing sessions, with the exception of MUFs.
assessed across two testing sessions. The statistical model was a 2-way fully nested ANOVA, which subdivided the variance into true error, day-to-day error, and trial-to-trial error. The mean squares and total sums of squares was then used to calculate the intraclass correlation coefficient (R) and the standard error of the measurement (SEM). The SEM was calculated according to [242], taking the square root of the trial-by-trial mean square error.

Results and sample size estimation calculated from the pilot study are detailed in Appendix X.

**Sample Size Estimation**

Sample size estimation was conducted using Cohen’s (1998) case four formula. This calculation requires the *a priori* establishment of the following values, which were taken from the pilot reliability study:

- Level of significance (α),
- The appropriate power (β),
- The mean (\( \bar{x} \)) and standard deviation (\( \sigma \)) of the criterion measure,
- The intraclass coefficient (R) measure of the criterion measure,
- The effect size (ES), which is deemed important or non-trivial by the investigator.

To balance the risk of Type I and Type II errors, Cohen (1969) suggests using a 4:1 ratio. To satisfy this ratio, \( \alpha \) of 0.05 and \( \beta \) of 0.20 are selected, where power is 0.80 (1-\( \beta \)). The means and standard deviations and interclass correlation coefficients for each measure can be found in Tables A1-A2. The effect size for each measure was determined based on previous literature. The sample size calculation was performed on the wrist flexion and dorsiflexion force and sEMG RMS amplitude taken from 100% MVC contractions, and the MUFRs taken from the 4-second plateau of the ramp contractions:
Mean force (wrist flexion and dorsiflexion), effect size of 8% change,
Agonist RMS amplitude (FCR and TA), effect size of 20% change,
Antagonist RMS amplitude (ECR and Soleus), effect size of 10% change,
Agonist V-waves (FCR and TA), effect size of 30% change,
Agonist MUFRs (FCR and TA), effect size of 15% change.

Since the aim of the pilot study was to assess reliability, rather than an intervention, the
differences between means is minimal and therefore the pilot appears under-powered.
Based on the estimations from the sample size calculations and importance of each
measure, the sample size for the training study was established at 20 participants per group.
Table A1. Intraclass correlation analysis of variance including grand means, standard error of measurement (SEM), and the intraclass correlation coefficients (R) calculated according to [234]. Root mean square (RMS) amplitude of the flexor carpi radialis (agonist) and extensor carpi radialis (antagonist), mean force, and V-waves were taken from wrist flexion maximal voluntary contractions. Motor unit firing rates (MUFRs) were taken from wrist flexion ramp contractions. Also reported are the calculated sample sizes based on a power of 0.80 and the resulting power for a sample size of 20 (per group), as estimated from the figures below.

<table>
<thead>
<tr>
<th>Source</th>
<th>Mean Force</th>
<th>Agonist Amplitude</th>
<th>Antagonist Amplitude</th>
<th>V-waves (N=13)</th>
<th>MUFRs (N=18)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grand Mean</td>
<td>50.62 N</td>
<td>246.07 uV</td>
<td>51.97 uV</td>
<td>592.4 uV</td>
<td>18.75 Hz</td>
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<tr>
<td>SEM</td>
<td>6.38 N</td>
<td>48.7 uV</td>
<td>14.0 uV</td>
<td>646.7 uV</td>
<td>1.95 Hz</td>
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<td>R</td>
<td>0.979</td>
<td>0.925</td>
<td>0.948</td>
<td>0.887</td>
<td>0.443</td>
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<td>Test Day</td>
<td>M ± SD</td>
<td>M ± SD</td>
<td>M ± SD</td>
<td>M ± SD</td>
<td>M ± SD</td>
</tr>
<tr>
<td>1</td>
<td>49.76 ± 27.02</td>
<td>271.5 ± 221.5</td>
<td>52.1 ± 36.9</td>
<td>724.7 ± 1243.5</td>
<td>18.71 ± 4.3</td>
</tr>
<tr>
<td>2</td>
<td>52.15 ± 30.09</td>
<td>237.1 ± 149.5</td>
<td>50.9 ± 41.6</td>
<td>604.1 ± 1333.5</td>
<td>18.79 ± 4.1</td>
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<tr>
<td>3</td>
<td>49.93 ± 24.88</td>
<td>229.6 ± 129.0</td>
<td>52.9 ± 40.9</td>
<td>448.5 ± 516.1</td>
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</tr>
</tbody>
</table>

| Estimated Sample size (Power = 0.80) | 19 | 16 | 60 | 50 | 24 |
| Estimated Power (N = 20)             | 0.82 | 0.88 | 0.35 | 0.40 | 0.70 |
Table A2. Intraclass correlation analysis of variance including grand means, standard error of measurement (SEM), and the intraclass correlation coefficients (R) calculated according to [234]. Root mean square (RMS) amplitude of the tibialis anterior (agonist) and soleus (antagonist), mean force, and V-waves were taken from dorsiflexion maximal voluntary contractions. Motor unit firing rates (MUFRs) were taken from dorsiflexion ramp contractions. Also reported are the calculated sample sizes based on a power of 0.80 and the resulting power for a sample size of 20 (per group), as estimated from the figures below.

<table>
<thead>
<tr>
<th>Source</th>
<th>Mean Force (N=18)</th>
<th>Agonist Amplitude (N=15)</th>
<th>Antagonist Amplitude (N=18)</th>
<th>V-waves (N=15)</th>
<th>MUFRs (N=16)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grand Mean</td>
<td>136.49 N</td>
<td>199.0 uV</td>
<td>17.3 uV</td>
<td>727.6 uV</td>
<td>16.77 Hz</td>
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<td>SEM</td>
<td>9.48 N</td>
<td>24.2 uV</td>
<td>2.4 uV</td>
<td>415.9 uV</td>
<td>2.29 Hz</td>
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<td>R</td>
<td>0.971</td>
<td>0.922</td>
<td>0.861</td>
<td>0.845</td>
<td>0.649</td>
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<tr>
<th>Test Day</th>
<th>Mean ± SD</th>
<th>Mean ± SD</th>
<th>Mean ± SD</th>
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<th>Mean ± SD</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>131.17 ± 45.79</td>
<td>200.4 ± 82.7</td>
<td>16.8 ± 6.5</td>
<td>755.7 ± 686.9</td>
<td>16.9 ± 3.2</td>
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<td>2</td>
<td>138.81 ± 44.19</td>
<td>194.2 ± 76.1</td>
<td>17.2 ± 6.9</td>
<td>661.9 ± 499.2</td>
<td>16.6 ± 3.3</td>
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<td>3</td>
<td>139.50 ± 39.00</td>
<td>202.4 ± 94.2</td>
<td>18.1 ± 8.0</td>
<td>765.3 ± 584.7</td>
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</tbody>
</table>

| Estimated Sample size (Power = 0.80) | 14 | 11 | 55 | 18 | 12 |
| Estimated Power (N = 20)             | 0.93 | 0.96 | 0.38 | 0.84 | 0.92 |
APPENDIX B

Ethics Approval for Manuscript 1

Certificate of Ethics Clearance for Human Participant Research

DATE: 7/10/2013

PRINCIPAL INVESTIGATOR: GABRIEL, David - Kinesiology

FILE: 12-281 - GABRIEL

TYPE: Masters Thesis/Project

STUDENT: Jessica McQuire

SUPERVISOR: David Gabriel

TITLE: Proprioeptive neuromuscular facilitation of the wrist flexors

ETHICS CLEARANCE GRANTED

Type of Clearance: NEW

Expiry Date: 7/31/2014

The Brock University Bioscience Research Ethics Board has reviewed the above named research proposal and considers the procedures, as described by the applicant, to conform to the University’s ethical standards and the Tri-Council Policy Statement. Clearance granted from 7/10/2013 to 7/31/2014. Continued clearance is contingent on timely submission of reports.

The Tri-Council Policy Statement requires that ongoing research be monitored by, at a minimum, an annual report. Should your project extend beyond the expiry date, you are required to submit a Renewal form before 7/31/2014. Continued clearance is contingent on timely submission of reports.

To comply with the Tri-Council Policy Statement, you must also submit a final report upon completion of your project. All report forms can be found on the Research Ethics web page at http://www.brocku.ca/research/policies-and-forms/research-forms.

In addition, throughout your research, you must report promptly to the REB:

a) Changes increasing the risk to the participant(s) and/or affecting significantly the conduct of the study;

b) All adverse and/or unanticipated experiences or events that may have real or potential unfavourable implications for participants;

c) New information that may adversely affect the safety of the participants or the conduct of the study;

d) Any changes in your source of funding or new funding to a previously unfunded project.

We wish you success with your research.

Approved:

Brian Roy, Chair

Bioscience Research Ethics Board

Note: Brock University is accountable for the research carried out in its own jurisdiction or under its auspices and may refuse certain research even though the REB has found it ethically acceptable.

If research participants are in the care of a health facility, at a school, or other institution or community organization, it is the responsibility of the Principal Investigator to ensure that the ethical guidelines and clearance of those facilities or institutions are obtained and filed with the REB prior to the initiation of research at that site.
Ethics Approval for Manuscript 3

Brock University
Research Ethics Office
Tel: 905-688-5550 ext. 3035
Email: reb@brocku.ca

Bioscience Research Ethics Board

Certificate of Ethics Clearance for Human Participant Research

DATE: 2/12/2016
PRINCIPAL INVESTIGATOR: GABRIEL, David - Kinesiology
FILE: 15-103 - GABRIEL
TYPE: Ph. D. STUDENT: Lara Green
SUPERVISOR: David Gabriel
TITLE: The Examination of Potential Mechanisms Underlying the Cross Education Phenomenon

ETHICS CLEARANCE GRANTED

Type of Clearance: NEW Expiry Date: 2/28/2017

The Brock University Bioscience Research Ethics Board has reviewed the above named research proposal and considers the procedures, as described by the applicant, to conform to the University’s ethical standards and the Tri-Council Policy Statement. Clearance granted from 2/12/2016 to 2/28/2017.

The Tri-Council Policy Statement requires that ongoing research be monitored by, at a minimum, an annual report. Should your project extend beyond the expiry date, you are required to submit a Renewal form before 2/28/2017. Continued clearance is contingent on timely submission of reports.

To comply with the Tri-Council Policy Statement, you must also submit a final report upon completion of your project. All report forms can be found on the Research Ethics web page at http://www.brocku.ca/research/policies-and-forms/research-forms.

In addition, throughout your research, you must report promptly to the REB:

a) Changes increasing the risk to the participant(s) and/or affecting significantly the conduct of the study;
b) All adverse and/or unanticipated experiences or events that may have real or potential unfavourable implications for participants;
c) New information that may adversely affect the safety of the participants or the conduct of the study;
d) Any changes in your source of funding or new funding to a previously unfunded project.

We wish you success with your research.

Approved: ____________________________
Sandra Peters, Chair
Bioscience Research Ethics Board

Note: Brock University is accountable for the research carried out in its own jurisdiction or under its auspices and may refuse certain research even though the REB has found it ethically acceptable.

If research participants are in the care of a health facility, at a school, or other institution or community organization, it is the responsibility of the Principal Investigator to ensure that the ethical guidelines and clearance of those facilities or institutions are obtained and filed with the REB prior to the initiation of research at that site.
APPENDIX C

Sample Traces (Manuscript 3)
Figure 1. Representative figure of one participant’s tibialis anterior activity during dorsiflexion maximal voluntary contractions performed at baseline (left panel) and post-training (right panel).
Figure 2. Representative figure of one participant’s force traces during wrist flexion maximal voluntary contractions performed at baseline (grey) and post-training (black). The right panel is zoomed in to identify the force obtained from maximal voluntary contractions (A) and twitch interpolation (B). The central activation ratio (CAR) is demonstrated on the right panel.

\[
CAR = 1 - \frac{\text{Interpolated twitch force (B)}}{\text{Maximal voluntary force (A)}} \times 100
\]
APPENDIX D

Training Forms and Instructions (Manuscript 3)

Wrist Flexion Training

Starting position: full wrist extension

Mid-curl: close fingers as wrist flexes

Ending position: full wrist flexion
Dorsiflexion Training

Cable pulley may need to be lowered to the lowest setting (pin nearest floor).

A step may be sat on, or placed under the calf to elevate the heel off the floor.
Starting position: full plantar flexion (remove shoe if plantar flexion inhibited by back edge).

Ending position: full dorsiflexion.
Training Notes

1. Training should occur **4 times per week**, ideally spaced out throughout the week.

2. Each session should consist of **3 sets of 10-12 reps**
   a. Week 1 perform 10 reps, weeks 2 and 3 increase to 12 reps
   b. Week 4 perform 10 reps at the new weight, week 5 and 6 increase to 12 reps

3. The weight is set at approximately 80% of your isometric maximum and will be readjusted at week 3 during the mid-training test session.

4. Each contraction should last approximately **6 seconds**, count 3 seconds for full flexion (concentric) and 3 seconds for full extension (eccentric) back to rest

5. Perform each contraction through your **full range of motion**

6. Allow for **3-5 minutes of rest** between each set.

7. Train only the limb (dominant ankle OR wrist) as specified by the experimenter.

8. Do NOT begin any NEW training program, sport, or fitness routine.

9. When completing training on the same day as other activity, perform the strength training **BEFORE** any endurance training.
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<thead>
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<th>Week</th>
<th>Dates</th>
<th>Sets</th>
<th>Reps</th>
<th>Weight</th>
<th>Notes</th>
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<td>Testing Session: Sept 21&lt;sup&gt;st&lt;/sup&gt; 10:00 am</td>
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