EFFECT OF WHEY PROTEIN AND HIGH-VOLUME RESISTANCE TRAINING ON
MUSCLE MASS AND STRENGTH IN POSTMENOPAUSAL WOMEN

A Thesis
Submitted to the Faculty of Graduate Studies and Research
In Partial Fulfillment of the Requirement
For the Degree of

Master of Science
in
Kinesiology and Health Studies
University of Regina

By

Krissy Dawn Weisgarber
Regina, Saskatchewan
July 30, 2014

Copyright 2014: K.D. Weisgarber
UNIVERSITY OF REGINA

FACULTY OF GRADUATE STUDIES AND RESEARCH

SUPERVISORY AND EXAMINING COMMITTEE

Krissy Dawn Weisgarber, candidate for the degree of Master of Science in Kinesiology & Health Studies, has presented a thesis titled, *Effect of Whey Protein and High-Volume Resistance Training on Muscle Mass and Strength in Postmenopausal Women*, in an oral examination held on June 24, 2014. The following committee members have found the thesis acceptable in form and content, and that the candidate demonstrated satisfactory knowledge of the subject material.

External Examiner: *Dr. Jennifer Copeland, University of Lethbridge*

Supervisor: Dr. Darren Candow, Faculty of Kinesiology & Health Studies

Committee Member: Dr. John Barden, Faculty of Kinesiology & Health Studies

Committee Member: Dr. Paul Bruno, Faculty of Kinesiology & Health Studies

Chair of Defense: Prof. Wes Pearce, Faculty of Fine Arts

*via Tele-conference*
Abstract

The purpose was to examine the combined effects of whey protein supplementation and high-volume resistance training (HVRT) on muscle mass and strength in healthy postmenopausal women. Postmenopausal women (n=12, age: 57 ± 4.3 years, weight: 72 ± 13.4 kg, height: 163 ± 5.7 cm, body mass index: 27 ± 5.4) consumed whey protein (40 grams) or placebo immediately following unilateral resistance training 2 days per week (Monday, Thursday) and consumed the opposite beverage after training the other side of the body on alternating days (Tuesday, Friday) for 10 weeks. For training, participants performed 3 sets at 30% baseline 1-repetition maximum (1RM) to volitional muscle fatigue for 4 exercises (leg curl, biceps curl, leg extension, triceps extension). Prior to and following training, assessments were made for upper and lower limb lean tissue mass (dual energy x-ray absorptiometry), muscle thickness of the elbow and knee flexors and extensors (ultrasound), and muscle strength (1RM leg curl, biceps curl, leg extension, triceps extension). There was a significant increase over time for muscle strength (biceps curl, leg extension, triceps extension; P=0.006) and muscle thickness (elbow flexors and extensors; P=0.022) with no differences between whey protein and placebo. High-volume resistance training improves indices of muscle mass and strength in postmenopausal women with no greater benefits observed from whey protein.
Acknowledgements

I would like to thank my supervisor Dr. Darren Candow for his expertise, patience, time and knowledge through this thesis process as well as my committee members, Dr. John Barden and Dr. Paul Bruno for their comments and input. I would like to thank my study participants for their time and energy spent on the study, without them the study would cease to exist. I would also like to thank my parents and boyfriend who have been a wonderful support system, helping and encouraging me along the way. This thesis would not have been able to happen without your love, support and motivation. Thanks for making this possible.
Table of Contents

Abstract ..................................................................................................................... ii
Acknowledgements..................................................................................................................... iii
Table of Contents........................................................................................................................ iv
List of Tables ............................................................................................................................. vii
List of Figures ........................................................................................................................... viii
List of Appendices...................................................................................................................... ix
List of Abbreviations ................................................................................................................... x

CHAPTER ONE: Introduction ....................................................................................... 1

1.1 Muscle protein turnover ................................................................................................. 4

1.1.1 Mammalian Target of Rapamycin (mTOR)................................................................. 4

1.1.2 Estrogen ......................................................................................................................... 5

1.1.3 Growth hormone, insulin-like growth factor & dehydroepiandrosterone ............... 6

CHAPTER TWO: Potential Benefits of Resistance Training............................................ 7

2.1 Training intensity ............................................................................................................. 9

2.1.1 High volume resistance training ................................................................................. 9

CHAPTER THREE: Whey Protein Supplementation..................................................... 10

3.1 Protein ............................................................................................................................. 10

3.1.1 Muscle protein synthesis ......................................................................................... 11

3.2 Protein source ................................................................................................................. 13

3.2.1 Leucine ....................................................................................................................... 13

3.3 Whey protein and resistance training .......................................................................... 14

3.4 Purpose and hypothesis ............................................................................................... 15
APPENDIX A: Participant Information and Consent Form .......................................... 52

APPENDIX B: PAR-Q.................................................................................................. 56

APPENDIX C: PARMED-X........................................................................................... 57

APPENDIX D: 3 Day Food Log Instructions ................................................................... 61
List of Tables

Table 2.1 Summary of studies involving traditional resistance training programs in postmenopausal women .................................................................8

Table 5.1 Muscle strength values at baseline and 10 weeks ........................................... 26

Table 5.2 Muscle thickness values at baseline and 10 weeks ........................................... 28

Table 5.3 Muscle quality values at baseline and 10 weeks ............................................. 30
List of Figures

Figure 3.1 Schematic diagram of increased translational efficiency from additional dietary amino acids leading to greater muscle mass and strength involving resistance training ................................................................. 12
List of Appendices

<p>| | | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Participant Information and Consent Form</td>
<td>52</td>
</tr>
<tr>
<td>B</td>
<td>PAR-Q</td>
<td>56</td>
</tr>
<tr>
<td>C</td>
<td>PARMED-X</td>
<td>57</td>
</tr>
<tr>
<td>D</td>
<td>3 Day Food Log Instructions</td>
<td>61</td>
</tr>
</tbody>
</table>
## List of Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-RM</td>
<td>One Repetition Maximum</td>
</tr>
<tr>
<td>4E-BP1</td>
<td>4E Binding Protein</td>
</tr>
<tr>
<td>cm</td>
<td>Centimeter</td>
</tr>
<tr>
<td>g</td>
<td>Gram</td>
</tr>
<tr>
<td>ACSM</td>
<td>American College of Sports Medicine</td>
</tr>
<tr>
<td>CSA</td>
<td>Cross Sectional Area</td>
</tr>
<tr>
<td>DHEA</td>
<td>Dehydroepiandrosterone</td>
</tr>
<tr>
<td>DXA</td>
<td>Dual Energy X-ray Absorptiometry</td>
</tr>
<tr>
<td>EAA</td>
<td>Essential Amino Acids</td>
</tr>
<tr>
<td>eIF-3</td>
<td>Eukaryotic Initiation Factor 3</td>
</tr>
<tr>
<td>eIF-4E</td>
<td>Eukaryotic Translation Initiation Factor 4E</td>
</tr>
<tr>
<td>eIF-4G</td>
<td>Eukaryotic Translation Initiation Factor 4 Gamma Protein</td>
</tr>
<tr>
<td>ERs</td>
<td>Estrogen Receptors</td>
</tr>
<tr>
<td>GH</td>
<td>Growth Hormone</td>
</tr>
<tr>
<td>HVRT</td>
<td>High Volume Resistance Training</td>
</tr>
<tr>
<td>IGF-1</td>
<td>Insulin-Like Growth Factor 1</td>
</tr>
<tr>
<td>IL-1</td>
<td>Interleukin-1 Cytokine</td>
</tr>
<tr>
<td>IL-6</td>
<td>Interleukin-6 Cytokine</td>
</tr>
<tr>
<td>Kg</td>
<td>Kilogram</td>
</tr>
<tr>
<td>MHz</td>
<td>Megahertz</td>
</tr>
<tr>
<td>MPB</td>
<td>Muscle Protein Breakdown</td>
</tr>
<tr>
<td>MPS</td>
<td>Muscle Protein Synthesis</td>
</tr>
<tr>
<td>MQ</td>
<td>Muscle Quality</td>
</tr>
<tr>
<td>mRNA</td>
<td>Messenger Ribonucleic Acid</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Description</td>
</tr>
<tr>
<td>-------------</td>
<td>--------------------------------------------------</td>
</tr>
<tr>
<td>mTOR</td>
<td>Mammalian Target of Rapamycin</td>
</tr>
<tr>
<td>mTOR-C1</td>
<td>Raptor Mammalian Target of Rapamycin</td>
</tr>
<tr>
<td>mTOR-C2</td>
<td>Rictor Mammalian Target of Rapamycin</td>
</tr>
<tr>
<td>NEAA</td>
<td>Non-Essential Amino Acids</td>
</tr>
<tr>
<td>p70 s6k</td>
<td>70-kDA S6 protein kinase</td>
</tr>
<tr>
<td>PAR MED-X</td>
<td>Physical Activity Readiness Medical Examination</td>
</tr>
<tr>
<td>PAR-Q</td>
<td>Physical Activity Readiness Questionnaire</td>
</tr>
<tr>
<td>PASE</td>
<td>Physical Activity Scale For The Elderly</td>
</tr>
<tr>
<td>rRNA</td>
<td>Ribosomal Ribonucleic Acid</td>
</tr>
<tr>
<td>S6K11</td>
<td>Protein S6 Kinase</td>
</tr>
<tr>
<td>S6K1</td>
<td>Ribosomal Protein S6 Kinase Beta-1</td>
</tr>
<tr>
<td>S6K2</td>
<td>Ribosomal Protein S6 Kinase Beta-2</td>
</tr>
<tr>
<td>tRNA</td>
<td>Transfer Ribonucleic Acid</td>
</tr>
</tbody>
</table>
CHAPTER ONE: Introduction

Aging is associated with a decrease in skeletal muscle mass (i.e. sarcopenia) and strength (Doherty, 2003; Hunter, McCarthy & Bamman, 2004; Lenk, Schuler & Adams, 2010) which subsequently decreases the ability to perform tasks of daily living (Short & Nair, 2001) such as carrying groceries and climbing stairs. Sarcopenia has been shown to contribute to insulin resistance, type 2 diabetes mellitus, dyslipidemia, hypertension (Hunter, McCarthy & Bamman, 2004; Karakelides & Nair, 2005), rheumatoid- and osteoarthritis, vascular disease and osteoporosis (Breen & Phillips, 2011). There is typically a 0.5% reduction in muscle mass per year after 50 years of age with an accelerated deterioration in muscle tissue after 65 years of age (Karakelides & Nair, 2005; Lenk, Schuler & Adams, 2010). Females typically experience an accelerated decline in muscle mass (0.6% per year) after menopause (Candow, Chilibeck, Weisgarber, Vogt & Jones, 2013), possibly because of a decrease in estrogen production (Maltais, Desroches, & Dionne, 2009) and an imbalance between muscle protein synthesis (MPS) and muscle protein breakdown (MPB) (Boire, 2009). Subsequent to aging muscle loss, there is also a 20-50% decrease in maximal muscle strength (Doherty, 2003). Furthermore, strength expressed relative to muscle mass (i.e. muscle quality = strength/muscle mass) is a better indicator of muscle function than muscle strength alone (Dutta, Hadley & Lexell, 1997). While the exact mechanisms explaining sarcopenia remain to be elucidated, one main mechanistic contributor involves muscle protein kinetics (Breen & Phillips, 2011). Resistance exercise promotes numerous adaptations in skeletal muscle, many of which may help prevent or reverse sarcopenia (Hunter, McCarthy & Bamman, 2004; Karakelides & Nair, 2005; Lenk, Schuler & Adams, 2010;

It is generally perceived that gains in muscle hypertrophy are achieved with heavier loads. However, emerging research indicates that HVRT may result in similar or even greater rates of MPS, which may lead to increased muscle hypertrophy over time (Bemben, Fetters, Bemben, Nabavi, & Koh, 2000; Burd et al., 2010; Ogasawara, Loenneke, Thiebaud & Abe, 2013). For example, Bemben et al. (2000) compared the effects of work-matched, low load, high-volume resistance training (40% 1-RM, 16 repetitions) to high load, low-volume resistance training (80% 1-RM, 8 repetitions) for 12 exercises (3 sets: leg extension, leg curl, leg press, shoulder press, biceps curl, triceps extension, seated row; 1 set: hip extension, hip flexion, hip abduction, hip adduction; 3 days per week for 6 months) in 25 postmenopausal women (41-60 years). Results showed that both resistance training protocols resulted in similar gains in muscle mass and strength. Similarly, Taaffe et al. (1996) found no significant difference in thigh muscle strength, fiber cross sectional area (CSA) and tissue compositions between work-matched high intensity (7 repetitions; 80% 1-RM) and low intensity (14 repetitions; 40% 1-RM) resistance exercise programs performed 3 times per week (3 exercises: leg press, knee extension, knee flexion) for 52 weeks in healthy 65-79 year old women. Results across studies suggest that low-load resistance training may be an alternative to high-load resistance training for aging adults for improving muscle mass and strength. From a practical standpoint, aging adults may feel more comfortable in using lighter loads during a resistance training program due to certain physiological ailments (i.e. arthritis, osteopenia, osteoporosis, inflammation) associated with aging (Bemben et al., 2000;
Breen & Phillips, 2011; Loenneke & Pujol, 2011). Furthermore, adaptations from low load, high-volume resistance training may occur at a faster rate compared to high load, low-volume resistance training (Bemben et al., 2000).

In addition to the potential beneficial effects of HVRT, aging muscle mass and strength may be further increased with the addition of whey protein since two of the main contributing factors of ‘anabolic resistance’ with aging (defined as a blunted muscle protein synthetic response; Kumar et al., 2009) are resistance training and exogenous amino acids (Breen & Phillips, 2011; 2009; Loenneke & Pujol, 2011). Several studies have shown that whole-body protein turnover (i.e. changes in the rates of MPS and MPB), mixed MPS (i.e. sarcoplasmic, mitochondrial, myofibrillar), myofibrillar protein synthesis, and myosin heavy chain synthesis may be affected by aging (Karakelides & Nair, 2005). Whey protein, defined as a series of globular proteins derived from cheese and milk manufacturing from dairy cows, has a high essential amino acid profile, specifically leucine (Ha & Zemel, 2003), which is quickly absorbed leading to rapid amino acid delivery to skeletal muscles (Burd, Tang, Moore & Phillips, 2009; Phillips, Hartman, & Wilkinson, 2005), and increases the rates of MPS at rest and following resistance exercise (Burd et al., 2009; Pennings et al., 2011; Tang, Moore, Kujbida, Tarnopolsky, & Phillips, 2009). Therefore, the purpose of this thesis was to determine whether whey protein and HVRT could overcome ‘anabolic resistance’ and improve muscle mass and strength in postmenopausal women.
1.1 Muscle Protein Turnover

The constant and simultaneous synthesis and breakdown of skeletal muscle protein is referred to as muscle protein turnover (Breen & Phillips, 2011). If the rates of MPS are greater than the rates of MPB, net muscle protein accretion occurs potentially leading to muscle hypertrophy over time (Breen and Phillips, 2011). The cellular mechanisms helping explain muscle protein turnover include gene transcription, cell signalling, and enzymes involved in various proteolytic pathways (Burd et al., 2009).

1.1.1 Mammalian Target of Rapamycin. The mammalian target of rapamycin (mTOR) pathway is considered the main governing regulator of messenger ribonucleic acid (mRNA) translation and subsequent MPS (Little & Phillips, 2009; Schoenfeld, 2010; Weinhert, 2009) and has been shown to play a key role in the regulation of mRNA translation and MPS for muscle hypertrophy (Bemben et al., 2000). mTOR has two distinct complexes, raptor-mTOR and rictor-mTOR, which are more commonly referred to as mTOR-C1 and mTOR-C2 (Weinhert, 2009; Wang & Proud, 2006). Research shows that activation of mTOR and its downstream kinases, 4E binding protein 1 (4E-BP1), 70-kDA S6 protein kinase (p70 s6k), and protein S6 kinase (S6K11), are involved in the muscle protein synthetic response to resistance training, as distribution of rapamycin (an mTOR inhibitor) to individuals prior to exercise inhibits MPS (Fry et al., 2011; Little & Phillips, 2009). mTOR regulates cell growth and is responsible for changes in several major processes including mRNA translation, ribosome biogenesis, nutrient metabolism, autophagy, regulation of muscle hypertrophy and myoblast fusion (Sarbassov, Ali & Sabatini, 2005). With regards to mRNA translation there are two major mammalian proteins of mTOR, S6 Kinase and 4E-BP1 (Sarbassov, Ali & Sabatini, 2005). S6 kinase
has two genes found in mammals, ribosomal protein S6 kinase beta-1 (S6K1) and ribosomal protein S6 kinase beta-2 (S6K2) (Wang & Proud, 2006). S6K1 and S6K2 are activated by mTOR, but how they exactly regulate cell size is still unclear (Sarbassov, Ali & Sabatini, 2005). S6K1 activity is controlled by amino acids (especially leucine) and insulin and has been shown to play a key role in the regulation of the eukaryotic initiation factor 3 (eIF-3) (Wang & Proud, 2006). 4E-BP1, in a non-phosphorylated state, suppresses mRNA translation by binding to eukaryotic translation initiation factor 4E (eIF-4E) preventing interaction with the eukaryotic translation initiation factor 4 gamma (eIF-4G) protein. When 4E-BP1 is phosphorylated by mTOR, eIF-4E is released and used to restore translation (Sarbassov Ali & Sabatini, 2005). It is possible that muscle atrophy with aging is due to insufficient stimulation of mTOR and subsequent MPS following anabolic stimuli, (i.e. resistance training or feeding; Evans, 1995). For example, Fry et al. (2011) found that following an acute bout of resistance training, up-regulation of MPS and associated translational signalling through the MTORC1 pathways occurred at several time segments in younger subjects but not in older adults; indicating a blunted anabolic response to resistance training with aging.

### 1.1.2 Estrogen

The biological process of menopause (i.e. cessation of menstruation) may lead to accelerated muscle and strength loss as postmenopausal women experience a reduced anabolic response to resistance training (Bamman et al., 2003) and dietary proteins (Smith et al., 2008). Mechanistically, estrogen is involved in the muscle protein synthetic process, and during and following menopause estrogen levels substantially decrease (Pannemans, Halliday & Westerterp, 1995). Therefore, the cessation of sufficient estrogen production may have a negative effect on muscle mass
and strength in postmenopausal women. Estrogen may have anabolic effects on muscle, possibly as a result of its conversion to testosterone (Doherty, 2003; Karakelides & Nair, 2005; Thomas, 2007). Together, estrogen and testosterone may inhibit the production of interleukin-1 (IL-1) and interleukin-6 (IL-6) cytokines, which may create an anticatabolic environment (Doherty, 2003; Thomas, 2007). IL-1 blocks the release of luteinizing hormone which decreases testosterone levels and enhances the sarcopenic process in muscle (Morely, 2001). IL-6 has been associated with a decline in functionality and strength in older adults (Morley et al., 2001). There is further evidence that skeletal muscle has estrogen receptors (ERs) at the mRNA level under the form of ERα and ERβ in the nuclei of muscle fibers and capillaries (Brown, 2008). The number of ERs on muscle fibers is diminished in postmenopausal women (Maltais et al., 2009). Additionally, skeletal muscle has a higher proportion of ERα on type II (fast twitch) muscle fibres. This may explain the greater loss of type II muscle fiber size with age as well as the rapid decrease in muscle mass and strength during the perimenopausal years (Brown, 2008). Estrogen may also exhibit indirect effects on skeletal muscle mass and function. Estrogen has a strong behavioral effect on spontaneous activity, with low levels decreasing the desire to be physically active (Brown, 2008). When running distances were longitudinally compared between rats with ovaries intact and rats with ovariectomies, running distances drastically decreased in the estrogen deficient rats. However, estrogen replacement restored running capacity (Brown, 2008).

1.1.3 Growth Hormone, Insulin-Like Growth Factor & Dehydroepiandrosterone. In addition to estrogen, menopause also decreases growth hormone (GH), insulin-like growth factor (IGF-1), a main governor of MPS, and
dehydroepiandrosterone (DHEA) (Maltais et al., 2009), all contributing factors to the age-related loss of muscle mass (Karakelides & Nair, 2005) and muscle performance (Doherty, 2003). IGF-1 is a protein mediated by growth-hormone releasing hormone (GH-RH) that works with GH to activate MPS and inhibit MPB mainly on the PI3K/AKT pathway (Maltais et al., 2009).

CHAPTER TWO: Potential Benefits of Resistance Training

It is well established that traditional resistance training (i.e. > 30% 1RM) programs have a positive effect on aging muscle mass and strength in postmenopausal women (Table 2.1). Mechanistically, muscle hypertrophy from resistance training may be caused by an increase in MPS (Schulte & Yarasheski, 2001), satellite cell activation and proliferation (Verdijk et al., 2009), anabolic hormone production (Smilios, Pilianidis, Karamouzis, Parlavantzis & Tokmakidis, 2007) and a decrease in catabolic cytokine activity (Cornish & Chilibeck, 2009).
Table 2.1 Summary of studies involving traditional resistance training programs in postmenopausal women.

<table>
<thead>
<tr>
<th>First Author, Year</th>
<th>Study Population</th>
<th>Intervention</th>
<th>Duration</th>
<th>Outcome Measure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bamman et al. 2003</td>
<td>N=14 (9 male; 5 female) non-RT healthy older adults; women postmenopausal; Age 68.7 ± 1.6 yrs; 66.2 ± 1.4 yrs; height: 177.9 ± 2.0 cm; 165.6 ± 1.5 cm Weight: 76.5 ± 4.1 kg; 69.4 ± 5.7 kg</td>
<td>Randomly assigned: 2 sets of 10 repetitions at 65-80% 1RM 3 times/week for elbow flexor and extension, seated row, seated overhead press, back extension, leg extension and curl, bench press and sit ups. 3 women randomized to leg press and 2 women to squats.</td>
<td>26 wks</td>
<td>Overall % change in FFM=4.3%. Overall % change in muscle CSA=7.1%.</td>
</tr>
<tr>
<td>Bemben et al. 2000</td>
<td>N=25; non-RT postmenopausal women; Age 41-60 yrs</td>
<td>Randomly assigned: High load (HL; 80%1RM, 8 reps) or high repetition (HR; 40% 1RM, 16 reps) for 3 sets of quadriceps extension, hamstring curl, leg press, shoulder press, biceps curls, triceps extension and seated row with 1 set of hip extension, hip flexion, hip abduction and adduction 3 times per week.</td>
<td>24 wks</td>
<td>Overall % change for MS for HL and HR =30% and 27%. Overall % change for muscle CSA for HL and HR=26.5% and 24%.</td>
</tr>
<tr>
<td>McCartney et al. 1995</td>
<td>N=142 (males=63; females=79) non-RT elderly individuals; Age 60-80 years</td>
<td>Randomly assigned: Exercise (EX) or control (CON) groups (60-70 yrs: 38 males, 36 females; 70-80 years: 25 males, 43 females). Training progressed from 2 sets 50% 1RM to 3 sets 80% 1RM for leg press, ankle plantar flexion, military press, bilateral bench press, biceps curl, ankle dorsiflexion and ad curls. Ten reps performed for arm exercises and 12 for leg.</td>
<td>42 wks</td>
<td>% change for muscle CSA for knee extensors in 60-70=5.6% and 70-80=1.8%; for all women=4.7%. Overall % change for MS for females 60-70=36.8% and for 70-80=55.3%; for all women % change MS =46.05%.</td>
</tr>
<tr>
<td>Nelson et al. 1994</td>
<td>N=39; non-RT postmenopausal women; Age 50-70yrs</td>
<td>Randomly assigned: High intensity (80% 1RM) RT 2 days/week for hip and knee extension, lateral pull-down, back extension, and abdominal flexion.</td>
<td>52 wks</td>
<td>Overall % change for MS=54%. Total body MM by 1.2 ± 0.4 kg.</td>
</tr>
<tr>
<td>Orsatti et al. 2008</td>
<td>N=43; non-RT postmenopausal women; Age 45-75yrs</td>
<td>Randomly Assigned: RT 3 times/week at 60-80% 1RM for 3 sets of 8-12 reps for 2 exercises for each major muscle group (chest, back, thigh) and 1 for each minor muscle group (biceps and triceps).</td>
<td>16 wks</td>
<td>Overall % change for MS =17.5%. Overall % change in muscle mass=9.6%.</td>
</tr>
<tr>
<td>Rhodes et al. 2000</td>
<td>N=44; non-RT elderly women; Age 65-75 yrs</td>
<td>Randomly Assigned: PRT performed 3 times/week, 3 sets of 8 reps at 75% 1RM for chest press, leg press, biceps curl, triceps extension, quadriceps extension and hamstring curl.</td>
<td>52 wks</td>
<td>Overall % change for MS=29%.</td>
</tr>
<tr>
<td>Ryan et al. 1998</td>
<td>N=27; healthy postmenopausal women; Mean age 61 ± 1 yrs</td>
<td>Randomly assigned: RT 3 times/week at 90% 3RM for leg press, chest press, leg curl, latissimus pull down, elbow flexion and extension, leg extension, upper back row, military press, hip abductor and adductor and abdominal curls. Weight increased when 1 set of upper body or 2 sets lower body of 12-15 reps completed.</td>
<td>16 wks</td>
<td>Overall % change for MS=58%</td>
</tr>
<tr>
<td>Taaffe et al. 1996</td>
<td>N=36; healthy elderly women; Age 65-79 yrs</td>
<td>Randomly assigned: Two exercise training intensities; high intensity (HI) 80 1RM for 7 reps or low intensity (LO) 40% 1RM for 14 reps for leg press, knee extension and knee flexion.</td>
<td>52 wks</td>
<td>Overall % change for MS for HI=67% and LO=57%. Overall % change for muscle CSA for HI=25% and LO=14%.</td>
</tr>
</tbody>
</table>

RT=resistance training; 1RM=1 repetition maximum; PRT=progressive resistance training; FFM=fat free mass; CSA=cross sectional area; MS=muscle strength; MM=muscle mass
2.1 Training Intensity

Typically, resistance training programs designed to increase muscle mass and strength focus on training at an intensity of 60-80% 1-RM, for 8-12 repetitions, 2-3 times per week for > 8 weeks (Garber et al., 2011). The American College of Sports Medicine (ACSM) suggests that a load of 70% 1-RM is needed to achieve muscle hypertrophy (Loenneke & Pujol, 2011). However, older adults, especially postmenopausal women, may find it difficult to train at high load (i.e. > 60% 1-RM) intensities due to age-related co-morbidities such as inflammation, arthritis and joint pain (Bemben et al., 2000; Breen and Phillips, 2011). Furthermore, sex differences may exist with regards to optimal training regimes in order to increase muscle mass and strength in aging individuals (Hunter, McCarthy & Bamman, 2004). Postmenopausal females may show a greater hypertrophic response to non-traditional resistance training programs which involve reduced exercise intensity and/or frequency of training (Hunter, McCarthy & Bamman, 2004).

2.1.1 High Volume Resistance Training. The variation in training intensity, based on a 1-RM continuum, allows for specific training adaptations (i.e. muscle hypertrophy, strength) to be achieved (Harris, Debeliso, Spitzer-Gibson & Adams, 2004). Recent evidence indicates that the rates of MPS are maximized at 60% 1-RM in older adults with no further increase at higher training intensities (i.e., 75–90% 1-RM) (Kumar et al., 2009); suggesting a theoretical exercise intensity ceiling. Two recent studies have further corroborated this theory by showing that HVRT (i.e. performing each working set at 30% 1-RM to volitional fatigue) is safe and potentially more effective for increasing and expanding the rates of MPS compared to high load, low-volume resistance training
(i.e. 80-90% 1-RM) (Mitchell et al., 2012; Breen and Phillips, 2011; Burd et al., 2010). For example, Burd et al., (2010) showed that 4 sets of HVRT knee extension contractions (30% 1-RM; 24 ± 3 repetitions) performed to volitional fatigue in young men resulted in a significant increase in myofibrillar MPS at 4 and 24 hours post exercise whereas knee extension contractions performed at 90% 1-RM (5 ±1 repetitions) only increased MPS 4 hours post exercise. More recently, Mitchell et al., (2012) compared the effects of 10 weeks of knee extension contractions performed at 30% 1-RM and 80% 1-RM to volitional fatigue (3 sets, 3 days per week) in young men. Results showed that both training intensities increased muscle cross-sectional area and strength over time. The authors speculate that the greater muscle protein synthetic response from HVRT is related to increased volume of exercise performed and greater muscle fiber activation, especially type II muscle fibers (Burd et al., 2010). Collectively, these findings across studies suggest that multiple set, HVRT (i.e. 30% 1-RM), performed to volitional fatigue is effective for increasing muscle hypertrophy and strength in young adults. However, the effects of HVRT exercise, performed over multiple sets, during a resistance exercise program in postmenopausal women are unknown. Aging muscle exhibits ‘anabolic resistance’ to resistance training, which may be overcome when the volume of exercise is increased (Kumar et al., 2009).

CHAPTER THREE: Whey Protein Supplementation

3.1 Protein

Proteins are organic compounds comprised of amino acids (Krieder & Campbell, 2009). Proteins are found in every cell of the body and aid in growth, recovery,
metabolic and hormonal functions (Powers & Howley, 2007). There are 20 amino acids used to make proteins, 9 essential (EAA) which must be obtained in sufficient quantities from the diet and 11 non-essential (NEAA) which the body can adequately produce or synthesize to maintain nitrogen balance. When protein needs are not adequately met through habitual dietary intake, skeletal muscle is degraded (i.e. transamination) to meet metabolic demands (Kreider & Campbell, 2009). Muscle hypertrophy occurs when adequate amounts of energy and dietary protein intake (i.e. nitrogen intake) exceeds nitrogen excretion resulting in a positive protein balance (i.e. protein synthesis > protein breakdown= positive nitrogen balance) (Campbell, Wilborn, & Bounty, 2010). The mechanical stimulus from resistance training causes an increase in both the rates of MPS and MPB. In the post-absorptive period following exercise, the rates of MPB exceed that of MPS leading to net negative nitrogen balance (Burd et al., 2009). However, an increase in amino acid availability through dietary protein ingestion reverses the catabolic response so that MPS exceeds the rates of breakdown, thereby creating a positive nitrogen balance (Burd et al., 2009). Theoretically, the repeated ingestion of dietary protein following resistance training sessions should lead to small net increases in muscle protein and muscle tissue over time (Weisgarber, Candow and Vogt, 2012).

3.1.1 Muscle Protein Synthesis. Amino acids are selected for protein synthesis by binding with transfer ribonucleic acid (tRNA). The information and order of amino acid sequence for each protein is governed by mRNA that is produced from DNA through transcription. An increase in amino acid availability through additional dietary protein supplementation could potentially increase the combination of tRNA with an amino acid for translocation to ribosomal ribonucleic acid (rRNA) (i.e. increased
translational efficiency) located on the ribosome organelle where protein synthesis occurs. Increased translational efficiency, in the presence of increased amino acid availability, should increase MPS following resistance exercise training sessions (Welle, Thornton, Statt, McHenry, 1994) leading to greater muscle mass and strength (Figure 3.1).

**Figure 3.1** Schematic diagram of increased translational efficiency from additional dietary amino acids leading to greater muscle mass and strength.
3.2 Protein Source

The type of protein ingested may play a pivotal role in the delivery of amino acids to exercising muscles for subsequent MPS (Breen and Phillips, 2011). For example, proteins that are quickly absorbed lead to rapid amino acid availability for protein synthesis (Dreyer & Volpi, 2005; Kreider & Campbell, 2009). Whey protein, a series of globular proteins derived from cheese and milk manufacturing from dairy cows, has a high essential amino acid profile, specifically leucine (Ha & Zemel, 2003), is quickly absorbed leading to rapid amino acid delivery to skeletal muscles (Burd et al., 2009; Phillips, Hartman, & Wilkinson, 2005), and increases the rates of MPS at rest and following resistance training (Burd et al., 2009; Pennings et al., 2011; Tang et al., 2009). Research has shown that the signalling pathways for stimulating MPS are up-regulated when whey protein is consumed after resistance training (Burd et al., 2009; Tang et al., 2009) which may lead to significant muscle accretion over time in older adults (Candow, Chillibeck, Facci, Abeysekara & Zello, 2006).

3.2.1 Leucine. Leucine is an insulin secreting amino acid (Paddon-Jones & Rasmussen, 2009) which stimulates mTOR and pathways involved in MPS (Little & Phillips, 2009; Paddon-Jones & Rasmussen, 2009) and exclusion of leucine results in the inactivation of mTOR signalling (Wang & Proud, 2006). Long-term studies in rodents indicate that leucine enhances phosphorylation of mTOR and p70S6K and decreases protein breakdown (Eley, Russell & Tisdale, 2007). Therefore, leucine appears to be the driving force behind the purported beneficial effects of whey protein.
3.3 Whey protein and resistance training

Resistance training typically increases the rates of MPS and MPB (Phillips, Hartman, & Wilkinson, 2005). Although the mTOR pathway is up-regulated during and after exercise (Beelen et al., 2008), net muscle protein balance remains negative until amino acids are consumed (Philips et al., 2005). Similar to the blunted response of aging muscle to resistance training, older adults also appear less responsive to hyperaminoacidemia (increased amounts of amino acids in the bloodstream; Loenneke & Pujol, 2011; Paddon-Jones, Short, Campbell, Volpi & Wolfe, 2008). Protein is considered an important nutrient required for the maintenance of muscle mass in older adults (Millward, 2008). Dietary protein provides amino acids that support muscle hypertrophy, primarily by stimulating MPS. EAA play the predominant role in promoting accrual of muscle proteins (Borsheim, Tipton, Wolfe & Wolfe, 2002; Volpi, Kobayashi, Sheffield-Moore, Mittendorfer & Wolfe, 2003) with leucine appearing to be most important for initiating molecular events (i.e., mTOR signalling pathway) associated with MPS (Fujita et al., 2007). The stimulatory effect of EAA/leucine on MPS may be blunted with aging, a contributing factor of ‘anabolic resistance’ (Burd et al., 2011; Rennie, 2009). Over time, ‘anabolic resistance’ to dietary proteins is hypothesized to contribute to an overall reduction in muscle protein balance and therefore precipitate loss of aging muscle mass. Therefore, speculation exists that aging adults may require increased dietary protein, preferably whey protein, following resistance training sessions to overcome ‘anabolic resistance’ and stimulate MPS to levels necessary for muscle accretion to occur. For example, whey protein (26 grams) supplementation consumed in close proximity to resistance exercise sessions for 12 weeks increased knee
extensor muscle size compared to carbohydrate (placebo) in healthy older men but had no
effect on muscle size of the elbow flexors and extensors or muscle strength (Candow et
al., 2006); possibly indicating that the dose of whey protein was too low to produce
meaningful muscle accretion and strength. Recently, 40 grams of whey protein
immediately following resistance training was shown to increase the rates of MPS to a
much greater extent than 10-20 grams of whey protein in older adults (Yang et al., 2012).
Due to the potential ‘anabolic resistance’ to dietary protein ingestion (Smith et al., 2008),
it seems plausible that postmenopausal women may require additional protein per serving
(i.e., 40 grams) following each training session to maximize the potential synergetic
effects from protein and resistance training.

3.4 Purpose and Hypotheses

The purpose of this thesis was to examine the effects of whey protein ingestion
and supervised unilateral HRVT in postmenopausal women. It was hypothesized that the
ingestion of whey protein and HRVT would increase lean tissue mass, muscle thickness
and strength more than placebo and HRVT.

CHAPTER FOUR: Methodology

4.1 Participants

Seventeen females, who verbally confirmed that they were postmenopausal (i.e.
defined as having their last menstrual cycle at least 1 year prior to the start of the study)
and were not performing supervised resistance training for 3 months prior to the start of
the study were enrolled. Non-resistance trained postmenopausal women were selected to
potentially maximize the physiological adaptations from resistance exercise (Candow et
al., 2013; Candow et al., 2012). We have previously shown that untrained older adults experience a 0.4 cm greater increase in knee extensor muscle size from whey protein (0.3g•kg⁻¹) and resistance exercise (12 weeks) compared to placebo (Candow et al., 2006). With an effect size of 0.7 (expected minimum change from whey protein and resistance exercise), an alpha level of 0.05, power of 80%, and correlation between the means of 0.9, 17 participants were required.

All participants were required to read, understand, and sign the informed consent form (approved by the University of Regina Research Ethics Board; Appendix A), before participating in the study. This form informed participants about the purpose of the study, the possible risks and adverse effects, contact information of the researchers, experimental methods, confidentiality agreements, and voluntary withdrawal instructions. Once the participant met all the qualifications to take part in the study, they were required to fill out a Physical Activity Readiness Questionnaire (PAR-Q; Appendix B) which assessed their readiness to participate in the resistance training program and included questions related to heart conditions, angina at rest or during physical exercise, as well as balance and bone or joint problems that may affect exercise performance. If the participant indicated any of the above conditions, they were given a Physical Activity Readiness Medical Examination (PAR MED-X; Appendix C) form to be filled out by their family physician. Only 1 participant required a PAR MED-X. Participants were excluded if they had a history of fragility fractures, were taking medications (i.e. bisphosphonates, hormone replacement therapy, prednisone, glucocorticoids) that affect muscle metabolism in the past 6 months, suffered from severe osteoarthritis, had consumed ergogenic aids (i.e. creatine supplements) for ≤ 6 weeks prior to the start of the
study, if they were vegetarians, if they were smokers, and if they had pre-existing kidney or liver abnormalities. Habitual dietary intake was assessed at baseline and at the end of the study using 3-day food diaries (Appendix D) to determine whether the participants changed their diet during the intervention. Participants were instructed not to change their diet or engage in additional physical activity that was not part of their normal daily routine during the study period intervention, to refrain from food or drink for one hour post-supplementation so that a valid estimate of the effects of the protein supplementation could be made (water was permitted ad libitum), and not to consume any ergogenic aids (i.e. creatine monohydrate) or non-steroidal anti-inflammatory drugs (i.e. ibuprofen) during the study as these interventions could affect muscle protein turnover (Candow et al., 2013; 2012) and would potentially influence the outcome measures.

4.2 Research Design

The study was a double-blind, repeated measures, within-subject (placebo control) design where postmenopausal women were randomized to consume whey protein or placebo during unilateral (dominant side of the body was randomized) resistance training 2 days per week (i.e. Monday, Thursday) and consume the opposite beverage while training the other side of the body on alternating days (i.e. Tuesday, Friday) for 10 weeks. This unique design allowed for the direct comparison of whey protein vs. placebo within the same individual, which increased our statistical power and internal validity. However, one potential limitation of this unilateral training design is cross education, where training of one muscle group can lead to neurally mediated strength gains in the untrained contralateral muscle group (Mitchell et al., 2012). Since both sides of the body performed 20 training sessions over 10 weeks using the same
exercises, and the side of the body was randomly allocated, the potential inter-limb effects of cross-education should be minimal. Recent data using a similar unilateral design found no correlation between strength gains in the left vs. right leg (r=0.33) when performing the same amount of resistance training at different training intensities (i.e. 30% vs. 80% 1-RM to volitional fatigue; Mitchell et al., 2012). At baseline and after the study, the primary dependent variables assessed included: (1) upper and lower limb lean tissue mass (DXA), (2) muscle thickness of knee and elbow flexors and extensors (ultrasound), and (3) unilateral muscle strength (1-RM leg curl, biceps curl, leg extension, triceps extension). In addition, participants filled out a 3-day food diary at baseline and after 10 weeks of training.

### 4.3 Nutritional Supplementation

Protein (lemon iced tea flavour powder; 40 grams. Amino acid composition per 40 grams: Alanine 2.0g, Arginine 0.8g, Aspartic Acid 4.4g, Cysteine 1.0g, Glutamine 6.2, Glycine 0.6g, Histidine 0.8g, Isoleucine 2.6g, Leucine 4.2g, Lysine 4.0g, Methionine 0.8g, Phenylalanine 1.2g, Proline 2.6g, Serine 1.8g, Threonine 2.8g, Tryptophan 0.6g, Tyrosine 1.2g, Valine 2.4g) and placebo (30 grams of cornstarch maltodextrin and 10 grams of lemon iced tea flavour powder) were identical in energy content, taste, texture, volume, and appearance. Protein and placebo were provided to each participant in separate, clearly labeled plastic bags with detailed instructions. Participants consumed the protein or placebo in the presence of the exercise supervisor to ensure 100% compliance. The whey protein dosage of 40 grams was chosen as this dosage maximizes the rates of MPS following resistance training in older adults compared to lower whey protein dosages (Yang et al., 2012). Participants consumed 25% of their beverage following
each exercise (4 exercises in total per day). Protein ingestion occurred immediately following exercise, as post-exercise protein ingestion is crucial for improving muscle hypertrophy in aging adults (Esmarck et al., 2001). Protein was not consumed prior to exercise and we have previously found no beneficial effect from this strategy in young adults (Weisgarber et al., 2012), possibly because compromised energy status during exercise blunts the anabolic response to amino acids (Rose & Richter, 2009). We chose a carbohydrate-based placebo for comparison to protein as carbohydrate intake has a minimal effect on MPS (Burd et al., 2009; Gelfand and Barrett, 1987).

4.4 Resistance Training

Prior to the start of supplementation and training, each participant had their unilateral 1-RM strength assessed for the leg curl, biceps curl, leg extension and triceps extension. Following 5-minutes of cycling on a stationary cycle ergometer at a self-selected intensity, participants performed two warm-up sets in order: 1 set of 10 repetitions using a weight determined by each participant to be comfortable and 1 set of 5 repetitions using increased weight. Two-minutes following the warm-up sets, weight was progressively increased for each subsequent 1-RM attempt with a 2-minute rest interval. 1-RM’s were performed in order: right leg curl, left leg curl, right biceps curl, left biceps curl, right leg extension, left leg extension, right triceps extension, left triceps extension.

Following determination of baseline 1RM strength, but prior to the start of supplementation and training, participants familiarized themselves with the machine-based resistance training equipment under direct supervision of a Canadian Society for Exercise Physiology-Certified Personal Trainer, in the Aging Muscle and Bone Health
Laboratory, University of Regina. We chose to use machine-based resistance training equipment because they are considered safer and easier to learn than free weights (Ratamass et al., 2009) and the use of machine-based equipment can lead to greater improvements in machine-based strength tests (Boyer, 1990). An important aspect of the study was that the personal trainer (exercise supervisor) ensured that each participant performed each set to volitional fatigue (defined as the inability to perform the concentric and eccentric phase of a muscle contraction). During the 3-familiarization resistance-training sessions, participants were properly shown how to breathe, use the equipment, and perform repetitions to volitional fatigue using 30% baseline 1-RM.

At least 4 days following the last familiarization session, participants performed 3 sets at 30% baseline 1-RM to volitional muscle fatigue for each exercise in order (i.e. leg curl, biceps curl, leg extension, triceps extension). Participants were instructed to perform the concentric phase in 2 seconds, pause for 2 seconds and then perform the eccentric phase in 2 seconds (i.e. 6 seconds of time under tension) for each muscle contraction. There was a 2 second pause between the concentric and eccentric movements to help reduce the stretch-reflex of muscle shortening (Bosco, Tarrka & Komi, 1982) and potential momentum which may have influenced the results. Participants trained their right extremities on Monday and Thursday and left extremities on Tuesday and Friday. Participants maintained daily training logs where average training volume per session (weight x sets x repetitions) was determined for each participant and monitored by the personal trainer following each resistance training session. Only complete muscle contractions were recorded and used for training volume calculations.
4.5 Primary Dependent Variables

4.5.1 Body Composition.  Limb bone mineral-free lean tissue mass was measured by DXA (Hologic Wi System, Christie Group, Manitoba, Canada) in array mode.  Before scanning, participants were required to take off all removable objects containing metal (i.e. jewellery, glasses, clothing with buttons, and/or zippers).  Scans were performed with participants lying in a supine position along the scanning table’s centerline longitudinal axis.  Feet were taped together at the toes (i.e. phalanges) to immobilize the legs while the hands were maintained in a pronated position within the scanning region.  All scans were performed by the same Nuclear Medicine Technologist.

4.5.2 Muscle Size.  Muscle thickness of the elbow flexors (i.e. biceps), elbow extensors (i.e. triceps), knee flexors (i.e. hamstrings) and knee extensors (i.e. quadriceps) were measured using B-Mode ultrasound (Aloka SSD-500 Tokyo, Japan).  To measure elbow flexor and extensor muscle thickness, a small mark was drawn on the lateral side of each arm to indicate 65% of the distance down from the acromion process to the olecranon process (Farthing & Chilibeck, 2003).  To measure elbow flexor muscle thickness, each participant placed their arms flat on the table with the belly of the biceps facing upwards and the forearm supinated.  To measure elbow extensor muscle thickness, participants stood with their back facing the researcher and with both arms extended and relaxed.

To measure knee flexor and extensor muscle thickness, a small mark was drawn on the lateral side of each leg to indicate 70% of the distance down from the greater trochanter to the lateral epicondyle of the tibia (Abe, Fukashiro, Harada & Kowamoto,
To measure knee flexor muscle thickness, each participant laid prone on the table with both legs extended and relaxed. For knee extensor muscle thickness, each participant sat upright on a table with both legs extended and relaxed.

A 5-MHz scanning transducer head was placed perpendicular to the muscle area interface. The scanning head was coated with water-soluble transmission gel to provide acoustic contact with the muscle surface. When the image produced on the screen was visible, the image on the monitor was frozen. With the image frozen, a cursor was enabled to quantify muscle thickness (cm) at three sites: the proximal, the mid, and the distal sites, as determined by divisions (1 cm) on the monitor. The distal and proximal sites were 6 cm apart, with the mid-site located 3 cm between them. The mid-site corresponds to where the reference mark is drawn over the measured muscle. Muscle thickness measurements were extrapolated from the monitor screen by measuring the distance from the bottom of the subcutaneous adipose layer to the surface of the humerus for the elbow flexors and extensors and to the surface of the femur for the knee flexors and extensors. Two muscle thickness measurements were taken at each site and averaged to give a muscle thickness value for that site. For each muscle thickness measurement, precise markings on the skin were taken using overhead transparency film to ensure that identical sites were measured on each occasion. All measurements were performed by the same experienced researcher.

4.5.3 Muscle Strength. Unilateral leg curl, biceps curl, leg extension and triceps extension strength was assessed using a 1-RM protocol on machine-based resistance training equipment (Pulse Fitness Systems Inc, Winnipeg, Manitoba, Canada). Following 5-minutes of cycling on a stationary cycle ergometer at a self-selected intensity,
participants performed two warm-up sets in order: 1 set of 10 repetitions using a weight determined by each participant to be comfortable and 1 set of 5 repetitions using increased weight. Two-minutes following the warm-up sets, the weight was progressively increased for each subsequent 1-RM attempt with a 2-minute rest interval. The 1-RM was achieved in 6 sets or less. All measurements were collected by the same experienced researcher.

4.5.4 Muscle Quality. Muscle quality was determined by dividing maximal muscle strength (kg) of each exercise by their respective muscle thickness measurement (cm): Leg curl (kg) / knee flexor (cm); Biceps curl (kg) / elbow flexor (cm); Leg extension (kg) / knee extensor (cm); Triceps extension (kg) / elbow extensor (cm).

4.5.5 Diet. Dietary intake was recorded prior to and immediately following supplementation and training to determine whether habitual dietary intake changed over time. Participants used a 3-day food booklet to record what they ate for 2 weekdays and 1 weekend day. Participants were instructed to record all food items, including portion sizes consumed for the 3 designated days. The Interactive Healthy Eating Index (Center for Nutrition Policy and Promotion, United States Department of Agriculture; http://www.cnpp.usda.gov/MyPyramidTracker.htm) was used to analyze 3-day food records. Each food item was entered and the program provided a total energy consumption average over the 3 days as well as energy from carbohydrates, fats, and proteins individually.
4.6 Statistical Analyses

A 2 (Limbs: whey protein side vs. placebo side) x 2 (Time: pre vs. post) x 4 (Exercise task/ Muscle group: leg curl/knee flexors; biceps curl/elbow flexors; leg extension/knee extensors; triceps extension/elbow extensors) repeated measures ANOVA was used to determine differences between sides of the body for muscle thickness, unilateral strength and muscle quality. A 2 (Limbs: whey protein side vs. placebo side) x 2 (Time: pre vs. post) x 2 (Muscle region: arm vs. leg) repeated measures ANOVA was conducted for lean tissue mass. Post hoc analyses (univariate ANOVA) of simple effects were used where significant interactions were found. Repeated measures ANOVA was used to assess changes in dietary intake over time. To determine if differences in training volume impacted the results, paired t-tests were used to assess differences in training volume between the right and left side of the body and between upper and lower muscle groups over 10 weeks of training. All results are expressed as means ± standard deviation. Statistical analyses was performed using SPSS version 18.0 for Windows XP (SPSS Chicago, IL). Significance was set at p < 0.05.

CHAPTER FIVE: Results

5.1 Participants

Of the 17 participants who were initially enrolled, 12 completed the study (age: 57 ± 4.3 years, weight: 72 ± 13.4 kg, height: 163 ± 5.7 cm, body mass index: 27 ± 5.4). Three participants withdrew due to the time constraints and one participant withdrew due to health complications not related to the study. There were no adverse effects reported from the resistance training, protein or placebo. The average 30% 1RM loads used over
the 10 weeks of training were: Leg curl (Protein: 27.4 ± 5.2kg, Placebo: 27.5 ± 5.1kg),
Biceps curl (Protein: 8.2 ± 1.7kg, Placebo: 8.6 ± 1.3kg), Leg extension (Protein: 26.7 ± 6.7kg, Placebo: 27.7 ± 7.0kg), and Triceps extension (Protein: 5.2 ± 0.9kg, Placebo: 5.2 ± 0.9kg).

5.2 Muscle Strength

There was a significant main effect of exercise task (p=0.001) and time (p=0.001) and a significant exercise task x time interaction (p=0.006) for strength. Post hoc analysis showed that muscle strength increased over time for the biceps curl (p=0.035), leg extension (p=0.024) and triceps extension (p=0.001), with no change for the leg curl (Table 5.1).
### Table 5.1. Muscle strength measurements (1-RM) for the biceps curl, leg extension, triceps extension and leg curl in postmenopausal women before and after 10 weeks of supplementation and resistance training.

<table>
<thead>
<tr>
<th>Muscle Group</th>
<th>Pre</th>
<th>Post</th>
<th>%</th>
<th>Pre</th>
<th>Post</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biceps Curl</td>
<td>18.1±3.7</td>
<td>24.0±4.8*</td>
<td>33.1</td>
<td>19.0±4.4</td>
<td>22.5±4.1*</td>
<td>18.4</td>
</tr>
<tr>
<td>Leg Extension</td>
<td>58.8±14.9</td>
<td>63.4±11.2*</td>
<td>7.8</td>
<td>61.3±15.3</td>
<td>65.7±13.0*</td>
<td>7.2</td>
</tr>
<tr>
<td>Triceps Extension</td>
<td>11.4±2.1</td>
<td>21.4±4.9*</td>
<td>87.7</td>
<td>11.4±2.0</td>
<td>21.0±5.7*</td>
<td>84.2</td>
</tr>
<tr>
<td>Leg Curl</td>
<td>60.2±11.4</td>
<td>60.1±7.4</td>
<td>-0.2</td>
<td>60.4±11.3</td>
<td>60.6±7.5</td>
<td>0.3</td>
</tr>
</tbody>
</table>

Values are mean (kg) ± standard deviation. %= percent change over time.
*Significantly different after training (p<0.05).
5.3 Muscle Size

There was a significant main effect of muscle group (p=0.001) and time (p=0.009) and a significant muscle group x time interaction (p=0.006) for muscle thickness. Muscle size of the elbow flexors (p=0.035) and extensors (p=0.006) increased over time with no change for the knee flexors or extensors (Table 5.2). There were no differences between whey protein or placebo.
Table 5.2. Muscle thickness measurements (cm) for the elbow and knee flexor and extensor muscle groups in postmenopausal women before and after 10 weeks of supplementation and resistance training.

<table>
<thead>
<tr>
<th>Muscle Group</th>
<th>Protein Pre</th>
<th>Protein Post</th>
<th>%</th>
<th>Placebo Pre</th>
<th>Placebo Post</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Elbow Flexors</td>
<td>2.4±0.5</td>
<td>2.5±0.6*</td>
<td>4.1</td>
<td>2.4±0.5</td>
<td>2.6±0.4*</td>
<td>8.3</td>
</tr>
<tr>
<td>Elbow Extensors</td>
<td>3.2±0.8</td>
<td>3.7±0.6*</td>
<td>15.6</td>
<td>3.3±0.4</td>
<td>3.7±0.5*</td>
<td>12.1</td>
</tr>
<tr>
<td>Knee Flexors</td>
<td>4.4±0.6</td>
<td>4.5±0.7</td>
<td>2.3</td>
<td>4.6±0.7</td>
<td>4.6±0.7</td>
<td>0.0</td>
</tr>
<tr>
<td>Knee Extensors</td>
<td>3.2±0.9</td>
<td>3.3±0.8</td>
<td>3.1</td>
<td>3.2±0.9</td>
<td>3.3±0.9</td>
<td>3.1</td>
</tr>
</tbody>
</table>

Values are mean (cm) ± standard deviation. %= percent change over time.
*Significantly different after training (p<0.05).
5.4 Muscle Quality

There was a significant main effect for muscle group (p=0.001) and time (p=0.029). Muscle quality increased with training for all muscle groups combined, independent of whey protein or placebo.
Table 5.3. Muscle quality (1-RM/cm) for the elbow and knee flexor and extensor muscle groups in postmenopausal women before and after 10 weeks of supplementation and resistance training.

<table>
<thead>
<tr>
<th>Muscle Group</th>
<th>Protein</th>
<th></th>
<th>%</th>
<th>Placebo</th>
<th></th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre</td>
<td>Post</td>
<td></td>
<td>Pre</td>
<td>Post</td>
<td></td>
</tr>
<tr>
<td>Elbow Flexors</td>
<td>7.8±1.4</td>
<td>10.0±2.1**</td>
<td>28.2</td>
<td>8.3±2.1</td>
<td>8.9±1.7*</td>
<td>7.2</td>
</tr>
<tr>
<td>Elbow Extensors</td>
<td>3.9±0.8</td>
<td>6.2±1.8*</td>
<td>58.9</td>
<td>3.7±0.6</td>
<td>5.9±1.2*</td>
<td>59.4</td>
</tr>
<tr>
<td>Knee Flexors</td>
<td>14.2±2.5</td>
<td>13.8±2.6</td>
<td>-2.8</td>
<td>13.7±2.6</td>
<td>13.7±2.2</td>
<td>0.0</td>
</tr>
<tr>
<td>Knee Extensors</td>
<td>20.0±4.4</td>
<td>20.3±4.6</td>
<td>1.5</td>
<td>20.2±5.9</td>
<td>22.0±5.9</td>
<td>8.9</td>
</tr>
</tbody>
</table>

Values are mean ± standard deviation. %= percent change over time.
*Significantly different after training (p<0.05).
**Significantly greater than placebo (p<0.05).
5.5 Lean Tissue

There was a significant main effect of muscle region (p=0.000) for lean tissue mass (n=11), where leg lean tissue mass was greater than arm lean tissue mass. Changes in lean tissue mass were similar (p>0.05) between protein and placebo for the arm (Protein: Pre 1847.3 ± 419.0 g, Post 1920.5 ± 419.4 g; Placebo: Pre 1808.9 ± 341.9 g, Post 1853.0 ± 337.9 g) and leg (Protein: Pre 6067.7 ± 975.8 g, Post 6228.9 ± 1082.1 g; Placebo: Pre 6162.7 ± 923.5 g, Post 6186.4 ± 1080.3 g).

5.6 Training Volume and Diet

There were no differences in total training volume between protein and placebo for biceps curl (Protein: 36099.1 ± 6858.2 kg; Placebo 37353.3 ± 10218.8 kg), leg extension (Protein: 47453.5 ± 16653.4 kg; Placebo: 49056.8 ± 17025.4 kg), triceps extension (Protein: 13677.2 ± 5324.1 kg; Placebo: 14458.4 ± 3701.6 kg), and leg curl (Protein: 56299.1 ± 13833.2 kg; Placebo: 51997.5 ± 15836.7 kg).

The average number of repetitions performed per set over the 10 weeks of training were similar between protein and placebo (Biceps curl: Protein 33.7 ± 7.8 reps; Placebo 33.4 ± 7.8 reps; Leg extension: Protein 13.4 ± 2.5 reps; Placebo 13.3 ± 2.6 reps; Triceps extension: Protein 20.5 ± 8.0 reps; Placebo 21.6 ± 5.3 reps; Leg curl: Protein 15.8 ± 4.8 reps; Placebo 14.6 ± 4.5).

Independent of protein or placebo, postmenopausal women performed more repetitions for the upper body exercises (Biceps curl: 32.2 ± 6.0 reps; Triceps extension: 20.2 ± 5.8 reps; p=0.000-0.016) compared to the lower body exercises (Leg extension: 13.3 ± 2.5 reps; Leg Curl: 15.2 ± 4.3 reps; p<0.05).
There was a significant increase in carbohydrate intake over time (Pre 150.0 ± 42.3g, Post 185.3 ± 29.4g; p=0.015), with no other differences (Kcal: Pre 1423.4 ± 270.9, Post 1627.2 ± 258.1; Fat: Pre 65.0 ± 43.9, Post 60.4 ± 19.9g; Protein: Pre 73.1 ± 19.4, Post 79.0 ± 18.3g).

CHAPTER SIX: Discussion

6.1 Discussion

The primary purpose of this study was to examine the effects of whey protein and unilateral HVRT on muscle accretion and strength in postmenopausal women. Based on previous findings showing that (1) whey protein increases the rates of MPS (Yang et al., 2012), (2) post-exercise protein ingestion is important for muscle accretion in older adults (Esmarck et al., 2001), and (3) high-volume resistance training (3-4 sets at 30% 1-RM to volitional fatigue) increases the rates of MPS leading to muscle hypertrophy and strength (Burd et al., 2010; Mitchell et al., 2012), it was hypothesized that whey protein and HVRT would lead to greater physiological adaptations compared to a placebo in postmenopausal women. However, results showed that whey protein had no greater effect on lean tissue mass, muscle thickness, strength or muscle quality compared to placebo. These results support previous findings of no substantial beneficial effect from whey protein following resistance training in older adults (Arnarson et al., 2013; Chale et al., 2013; Candow et al., 2006). Postmenopausal women and older men who ingested ≤ 40 grams of whey protein following resistance training for 12-24 weeks experienced similar gains in lean body mass and strength compared to older adults who consumed a carbohydrate placebo following training (Chale et al., 2013; Arnarson et al., 2013).
Furthermore, older men who supplemented with whey protein (26 grams) immediately following resistance training sessions for 12 weeks experienced no greater muscle accretion, except for the knee extensors, compared to placebo (Candow et al., 2006). On the other hand, in examining the effects of protein (~30g/day; 10 days) supplementation on body composition and whole-body protein kinetics in 17 malnourished elderly subjects, Bos et al. (2000) found a significant increase in MPS and fat-free mass from supplementation. It is important to note that malnourished older individuals may have higher protein turnover rates and a greater loss of muscle protein than healthy older individuals as a result of a hyper-catabolic state (Beaumont et al., 1989). It has been suggested that a main contributing factor of ‘anabolic resistance’ with aging is the blunted response to exogenous amino acids and that aging adults may require additional dietary protein following resistance training to achieve an anabolic environment for muscle growth (Breen and Phillips, 2013). We considered this theory and prescribed the highest dosage of whey protein (40 grams) shown to maximize the rates of MPS in aging adults (Yang et al., 2012) using a unique supplementation strategy (i.e. 10 grams of protein following each exercise) when combined with a safe and practical resistance training program. However, our results suggest that 40 grams of whey protein during short-term, high-volume resistance training does not lead to greater muscle accretion and strength compared to placebo in postmenopausal women. Furthermore, whey protein had no effect on the ability to perform more repetitions to volitional fatigue, which supports our previous findings (Candow et al., 2006; Weisgarber et al., 2012).

Despite the lack of benefit from whey protein, a unique and very important finding of this study is that HVRT to volitional fatigue increases muscle strength (i.e.
elbow flexors and extensors, knee extensors) and some measures of muscle thickness (i.e. elbow flexors and extensors) in postmenopausal women. From a health promotion and knowledge translational perspective, these results are important as the reduction in muscle strength with aging decreases the ability to perform activities of daily living (Hunter, McCarthy & Bamman, 2004; Roubenoff, 2003) and improvements in muscle size may lead to greater functional benefits (Chale et al., 2013). Furthermore, this study provides evidence that HVRT is a safe (no adverse effects were reported) and an alternative intervention to traditional resistance training recommendations (i.e. 2-3 sets at 70-80% 1-RM) to increase muscle mass and strength. These results indirectly support the findings of Mitchell et al., (2012) who showed that young healthy men who performed 3 sets of leg extension exercise at 30% 1-RM to volitional fatigue experienced a significant increase in muscle accretion and strength. Previous work from the same laboratory showed that resistance training performed at 30% 1-RM to volitional fatigue prolonged the muscle protein synthetic response compared to the same exercises performed at 90% 1-RM (Burd et al., 2010). In a more recent study, Ogasawara, Loenneke, Thiebaud & Abe (2013) showed that non-resistance trained young men experienced muscle hypertrophy when performing chest press exercise for 6 weeks at 30% 1-RM to volitional fatigue. Although it is difficult to compare results across studies which use different methodology, resistance exercise performed at 30% 1-RM to volitional fatigue appears to be an effective intervention to increase indices of muscle accretion and strength in young adults and postmenopausal women. However, as shown in Table 1, traditional resistance training programs (12-52 wks in duration, > 30% 1-RM) increases muscle mass by approximately 15.2% and muscle strength by 39.4% in older women compared to
increases of 1.9% for muscle mass and 12.9% for muscle strength from high-volume resistance training in the present study.

While the mechanistic actions explaining the positive effects of HVRT remain to be elucidated, muscle fiber recruitment patterns may be involved (Burd et al., 2009). Hennemann’s ‘size principle’ of neuromuscular adaptation to resistance training indicates a hierarchy of muscle fiber recruitment, with Type I fibers being recruited first followed by Type IIa and then Type IIx (Henneman, 1957). Type II muscle fibers contribute to strength and muscle hypertrophy to a much greater degree than Type I fibers (Thorstensson, Hulten, von Dobeln, & Karlsson, 1976). Speculation exists that the lighter loads performed to volitional fatigue, independent of training volume, may pre-fatigue Type I muscle fibers leading to earlier recruitment of Type II fibers. As such, there should be earlier recruitment of Type II muscle fibers with HVRT compared to traditional low-volume (high load) resistance training (Mitchell et al., 2012). This theory is supported by the findings of Mitchell et al. (2012) who observed an equal hypertrophic response of Type I and Type II muscle fibers from high-volume resistance training (30% 1-RM) in young adults; suggesting that earlier recruitment of Type II may contribute to the greater muscle protein synthetic response compared to low-volume training (Burd et al., 2010; Mitchell et al., 2012).

Postmenopausal women performed more repetitions per set over 10 weeks of training for upper body muscle groups (bicep curl, triceps extension) compared to lower body muscle groups (leg extension, leg curl) which may help explain the significant increase in upper body muscle hypertrophy and strength. It is unclear why upper body muscle groups responded more favorably to resistance training than lower body muscle
groups. However, there is evidence that lower body muscle groups may be more negatively affected with aging than upper body muscle groups. It has been previously shown that muscle thickness and strength of the knee flexors and extensors are reduced more with aging than the elbow flexors and extensors in healthy older men (Candow & Chilibeck, 2005). Furthermore, Lynch et al. (1999) found that arm and leg muscle mass and strength decreased with age but the decrease was significantly greater in the leg muscles of postmenopausal women, possibly because of alterations in muscle contractile properties, connective tissue, or architectural components. Potentially, the greater decline in muscle physiology of lower body muscle groups may help explain the blunted response to resistance training compared to upper body muscle groups that was observed in postmenopausal women. Therefore, increasing the duration (i.e. > 10 weeks) and intensity of resistance training (i.e. > 30% 1RM) may be required to produce significant muscle hypertrophy in lower body muscle groups in postmenopausal women.

6.2 Limitations

There were several limitations to the study, which may have influenced the results. First, to achieve 80% statistical power, 17 postmenopausal women were required. Unfortunately, 12 completed the study. This reduction in statistical power could have resulted in Type II error; the inability to detect a statistically significant treatment effect. Second, we chose to use a 10-week resistance-training program as Mitchell et al., (2012) found a significant increase in muscle accretion and strength when 3 sets of exercise were performed at 30% 1-RM to volitional fatigue for 10 weeks in young males. Results of the present study indicated that 10 weeks of HVRT increased muscle strength of the elbow flexors, elbow extensors and knee extensors and muscle size
of the elbow flexors and elbow extensors but had no effect on knee flexor strength or muscle size of the knee flexors or knee extensors. Therefore, a longer training period (>10 weeks) may have been needed to increase muscle size and strength in all muscle groups assessed. Third, methodological issues may have decreased our ability to detect small changes in muscle accretion over the 10 weeks of training. For example, limb lean tissue mass was measured using DXA. DXA measures lean tissue mass in the entire upper and lower limb. Exercises performed in the resistance training program primarily targeted the muscles proximal to the elbow and knee. Muscle groups distal to the elbow and knee would be minimally recruited and therefore could have diluted any small increase in muscle accretion of the proximal muscle groups as measured by DXA.

Fourth, all postmenopausal women consumed whey protein which limits our ability to make conclusions about the independent effects of whey protein on muscle mass and strength. For example, Yang et al. (2012) showed that whey protein (40 grams) immediately following an acute bout of unilateral leg resistance training increased the rates of muscle protein synthesis in the non-exercised leg in older adults. Therefore, it is possible that whey protein during training of one side of the body influenced the rates of muscle protein synthesis in the opposite side (placebo) of the body in our postmenopausal women. Furthermore, the muscle protein synthetic response to resistance training is elevated for 24 hours post-exercise when dietary protein is consumed in young adults (Burd et al., 2011). Potentially, muscle groups trained on the placebo days may be influenced by whey protein consumption on opposite training days. However, it is unknown whether this 24 hour anabolic window to dietary protein occurs in postmenopausal women. Fifth, habitual dietary protein intake may have masked the
effects of whey protein supplementation. Postmenopausal women were consuming, on
average, 78 grams of protein per day or approximately 0.95/kg/day of dietary protein
during this study. The current Dietary Recommended Intake for adults > 50 years of age
is 0.8g/kg/day. Potentially, women in the current study may have been consuming
sufficient protein, independent of the whey protein supplement, which could have
influenced our results. Sixth, our unilateral training design potentially involved cross-
education, where the training of one muscle group can lead to neurally mediated strength
gains in the untrained contralateral muscle group (Farthing, 2009). However, since both
sides of the body performed 20 training sessions over 10 weeks using the same exercises,
and the side of the body was randomly allocated, the potential inter-limb effects of cross-
education should be minimal. Furthermore, recent data using a unilateral design found
no correlation between strength gains in the left vs. right leg (r=0.33) when performing
the same amount of resistance training at different training intensities (i.e. 30% vs. 80%
1-RM to volitional fatigue). Seventh, baseline 1RM strength testing was performed prior
to the familiarization period which could have underestimated 1RM strength and the 30%
1RM training loads during the study. Finally, no measure of muscle protein kinetics was
made in this study. Future research should assess transcriptional and translational
efficiency through isotopic tracer or muscle biopsy technique to help understand the
mechanism of an anabolic response from amino acids.

6.3 Conclusions

Whey protein supplementation following HVRT has no greater effect on muscle
mass or strength compared to placebo in postmenopausal women. However, low
intensity, high-volume resistance training appears to be a safe and effective intervention
to increase some measures of muscle size and strength in postmenopausal women, particularly for the upper limbs. Future research should examine the safety and effectiveness of long-term, low intensity, high-volume resistance training on aging muscle and bone biology and tasks of functionality in males and females, and alternative strategies to enhance training induced responses for lower limb muscles in older adults.
References


Henneman, E. (1957). Relation between size of neurons and their susceptibility to


*Current Topics in Developmental Biology*, 68, 123-142.


Kumar, V., Selby, A., Rankin, D., Patel, R., Atherton, P., Hildebrandt, W., Williams, J.,
differences in the dose–response relationship of muscle protein synthesis to
211-217.

Lenk, K., Schuler, G. & Adams, V. (2010). Skeletal muscle wasting in cachexia and
sarcopenia: molecular pathophysiology and impact of exercise training. *Journal of
Cachexia Sarcopenia and Muscle*, 1, 9-21.


Loenneke, J. & Pujol, T. (2011). Sarcopenia: an emphasis on occlusion training and

(1999). Muscle quality: I. Age-associated differences between arm and leg muscle


Appendix A

Participant Information and Consent Form

Title of the study: Effect of whey protein following light-weight, high repetition weight training on muscle mass and strength in postmenopausal women

Researchers: Darren G. Candow, Ph.D., Associate Professor, Faculty of Kinesiology and Health Studies, University of Regina, phone: 306-585-4906, email: Darren.Candow@uregina.ca

Krissy Weisgarber, MSc student, Faculty of Kinesiology and Health Studies, email: kdw924@yahoo.com

24-hour emergency telephone contact: 306-209-0280

Disclosure: Dr. Darren Candow, the principal investigator of this study, has been affiliated with a company that develops and markets dietary protein supplement products. He held the appointment of Chief Scientific Advisor to RIVAL-US Sport Nutrition. Although that commercial entity is not a sponsor of this study, it could benefit from the results of this study. This information is provided to potential participants so they can decide whether or not to participate in this study.

Funded by: University of Regina

Introduction: You are being invited to participate in this research study because we want to determine the effects of whey protein immediately after weight training on body composition, muscle size, and muscle strength.

Before you decide to participate, it is important that you understand what the research involves. This consent form will inform you about the study, why the research is being performed, what will happen to you during the study, and the possible benefits, risks, and discomforts.

If you wish to participate, you will be asked to sign this form. Your participation is entirely voluntary, so it is up to you to decide whether or not to participate in this study. If you decide to take part in this study, you are free to withdraw at any time without giving any reasons for your decision and your refusal to participate will not affect your relationship with any of the researchers or institutions conducting the research. Please take time to read the following information carefully and to discuss it with your family, friends, and doctor or health professional before you decide.
**Purpose of the study:** To examine the effects of whey protein following light-weight, high repetition weight training on muscle mass and strength.

**Possible benefits of the study:** You might increase your muscle mass and strength by participating in this study. These benefits are not guaranteed.

**Procedures:** If you agree to participate in this study, the following will occur:

You will initially be given a questionnaire (Physical Activity Readiness Questionnaire; PAR-Q) which assesses whether you are at a health risk for participating in weight training. If you indicate a possible health risk, you will be given a Physical Activity Readiness Medical Examination (PARMED-X) form to be filled out by your family physician before being permitted to participate in this study.

Prior to the start of the study, you will be randomized to perform 4 weight training exercises (2 for the legs and 2 for the arms) using one side of the body and consume 10 grams of a protein or placebo beverage after each exercise for 3 sets to muscle fatigue using a pre-determined light weight, 2 days per week (Monday, Thursday). On 2 different days of the week (Tuesday, Friday), you will perform the same exercises listed above but use the opposite side of the body and consume the opposite drink for 12 weeks. The whey protein and placebo will be identical in taste, texture and appearance. The whey protein and placebo will be mixed in water. Neither you nor the researchers will know which beverage you are consuming each day until the end of the study, but we can find out which beverage you are consuming if there is an emergency (for example, an adverse reaction to the protein or placebo). Placebo is a compound which should provide no beneficial effect to muscle mass and strength. By having a placebo, we can determine whether whey protein increases muscle mass and strength over weight training alone.

You will participate in 12 weeks of weight training (i.e. 48 training sessions). Supplementation with whey protein and placebo will begin on the first day of the study and occur only on training days for 12 weeks. Supervised weight training will be performed 4 days per week (i.e., Monday, Tuesday, Thursday, Friday) and each training session will last approximately 45 minutes.

The following measurements will be performed at baseline and after 12 weeks:

- Lean tissue mass will be assessed using dual energy x-ray absorptiometry. This procedure requires you lying down on a table while your body is scanned by an x-ray beam. This measurement takes approximately 15 minutes.

- Muscle thickness will be measured using ultrasound by placing gel over your skin and applying a probe to your skin surface. Muscle thickness will be measured at the front and back of your upper arm, thigh, and lower leg. This procedure will take approximately 20 minutes.
- Your muscular strength will be measured by four different exercises (leg extension, leg curl, biceps curl, triceps extension). These procedures will take approximately 20 minutes.

- You will be required to write down all the foods you eat for 3 consecutive days and fill out a physical activity and muscle soreness questionnaire.

**Foreseeable risks, side effects or discomfort:** The weight training and strength testing may result in muscle pulls and strains. You will be given a proper warm-up prior to exercising and this will minimize the risk. Adequate rest will be given between training and testing sessions to ensure that your muscles are recovered by the next training session.

There is a small amount of radiation exposure from the dual energy X-ray scans. The amount of radiation from the dual energy X-ray scans is about 1/20th of the amount of radiation you would receive from taking a trans-Atlantic flight from North American to Europe.

**Alternatives to the study:** You do not have to participate in this study to have your body composition assessed. You could have your muscle mass determined through an appointment at the Faculty of Kinesiology and Health Studies, University of Regina and this can be performed by a number of different techniques (i.e. skin folds). You do not have to participate in this study to increase muscular strength. You can perform alternative exercises (i.e. free-body exercises such as push-ups or chin ups and wall squats instead of the weight training program in this study).

**Research-related injury:** There will be no cost to you for participation in this study. You will not be charged for any research procedures. In the event you become ill or injured as a result of participating in this study, necessary medical treatment will be made available at no additional costs to you. By signing this document you do not waive any of your legal rights.

**Confidentiality:** While absolute confidentiality cannot be guaranteed, every effort will be made to ensure that the information you provide for this study is kept entirely confidential. Your name will not be attached to any information, nor mentioned in any study report, nor be made available to anyone except the scientific team. It is the intention of the research team to publish results of this research in scientific journals and to present the findings at related conferences and workshops, but your identity will not be revealed. All data collected will be kept confidential in a locked storage cabinet and in password-protected computer files only the researchers can access.

**Voluntary withdrawal:** Your participation in this research is entirely voluntary. You may withdraw from this study at any time. If you decide to enter the study and to withdraw at any time in the future, there will be no penalty or loss of benefits to which you are otherwise entitled. If you choose to enter the study and then decide to withdraw at a later
time, all data collected about you during the enrolment in the study will be retained for analysis.

**Who to Contact for Questions or Concerns:** If you have questions concerning the study you can contact Dr. Darren Candow at 306-585-4906, or 306-209-0280 (24 hour cell).

This project has been approved on ethical grounds by the UofR Research Ethics Board on November 29, 2012. Any questions regarding your rights as a participant may be addressed to the committee at (585-4775 or research.ethics@uregina.ca). Out of town participants may call collect.

We will advise you of any new information that will have a bearing on your decision to continue in the study.

By signing below, I confirm the following:

- I have read or have had this form read to me and understand the research subject information and consent form.
- I have had sufficient time to consider the information provided and to ask for advice if necessary.
- I have had the opportunity to ask questions and have had satisfactory response to my questions.
- I understand that all the information collected will be kept confidential and that the results will only be used for scientific objectives.
- I understand that my participation in this study is voluntary and that I am completely free to refuse to participate or to withdraw from this study at any time without changing in any way the quality of training that I receive or my relationship with members of the research team of participating institutions.
- I understand that there is no guarantee that this study will provide any benefits to me.
- I have read and understand this form and I freely consent to participate in this study.
- I have been told that I will receive a dated and signed copy of this form.
- I agree that my family physician can be contacted about my participation in this study:

  _____ Yes    _____ No OR I do not have a family physician

Participant’s Name (printed): __________________________

Participant’s Signature: ___________________________ Date: ______________________

Name of Individual conducting the consent process (printed): __________________________

Signature of Individual conducting the consent process: __________________________

Date: ______________________
Appendix B

PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you are planning to become much more physically active than you are now, start by answering the seven questions in the box below. If you are between the ages of 15 and 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly: check YES or NO.

<table>
<thead>
<tr>
<th>YES</th>
<th>NO</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Has your doctor ever said that you have a heart condition and that you should only do physical activity recommended by a doctor?</td>
<td></td>
</tr>
<tr>
<td>2. Do you feel pain in your chest when you do physical activity?</td>
<td></td>
</tr>
<tr>
<td>3. In the past month, have you had chest pain when you were not doing physical activity?</td>
<td></td>
</tr>
<tr>
<td>4. Do you lose your balance because of dizziness or do you ever lose consciousness?</td>
<td></td>
</tr>
<tr>
<td>5. Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?</td>
<td></td>
</tr>
<tr>
<td>6. Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?</td>
<td></td>
</tr>
<tr>
<td>7. Do you know of any other reason why you should not do physical activity?</td>
<td></td>
</tr>
</tbody>
</table>

If you answered YES to one or more questions

Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.

- You may be able to do any activity you want — as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kinds of activities you wish to participate in and follow his advice.
- Find out which community programs are safe and helpful for you.

If you answered NO to all questions

If you answered NO honestly to all PAR-Q questions, you can be reasonably sure that you can:
- Start becoming much more physically active — begin slowly and build up gradually. This is the safest and easiest way to go.
- Take part in a fitness appraisal — this is an excellent way to determine your basic fitness so that you can plan the best way for you to live actively. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 144/94, talk with your doctor before you start becoming much more physically active.

PLEASE NOTE: If your health changes so that you then answer YES to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.

Note: This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.
Appendix C

**PARmed-X**

**PHYSICAL ACTIVITY READINESS MEDICAL EXAMINATION**

The PARmed-X is a physical activity-specific checklist to be used by a physician with patients who have had positive responses to the Physical Activity Readiness Questionnaire (PAR-Q). In addition, the Conveyance/Referral Form in the PARmed-X can be used to convey clearance for physical activity participation, or to make a referral to a medically-supervised exercise program.

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. The PAR-Q by itself provides adequate screening for the majority of people. However, some individuals may require a medical evaluation and specific advice (exercise prescription) due to one or more positive responses to the PAR-Q.

Following the participant’s evaluation by a physician, a physical activity plan should be devised in consultation with a physical activity professional (CSEP Certified Exercise Physiologist®). To assist in this, the following instructions are provided:

**PAGE 1:**
- Sections A, B, C, and D should be completed by the participant BEFORE the examination by the physician. The bottom section is to be completed by the examining physician.

**PAGES 2 & 3:**
- A checklist of medical conditions requiring special consideration and management.

**PAGE 4:**
- Physical Activity & Lifestyle Advice for people who do not require specific instructions or prescribed exercise.
- Physical Activity Readiness Conveyance/Referral Form - an optional tear-off tab for the physician to convey clearance for physical activity participation, or to make a referral to a medically-supervised exercise program.

### A PERSONAL INFORMATION:

<table>
<thead>
<tr>
<th>NAME</th>
<th>ADDRESS</th>
<th>TELEPHONE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>BIRTHDATE</th>
<th>GENDER</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>MEDICAL No.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
</tbody>
</table>

### B PAR-Q: Please indicate the PAR-Q questions to which you answered YES

- Q 1 Heart condition
- Q 2 Chest pain during activity
- Q 3 Chest pain at rest
- Q 4 Loss of balance, dizziness
- Q 5 Bone or joint problem
- Q 6 Blood pressure or heart drugs
- Q 7 Other reason:

### C RISK FACTORS FOR CARDIOVASCULAR DISEASE:

- Less than 30 minutes of moderate physical activity most days of the week.
- Currently smoker (tobacco smoking 1 or more times per week).
- High blood pressure reported by physician after repeated measurements.
- High cholesterol level reported by physician.
- Excessive accumulation of fat around waist.
- Family history of heart disease.

### D PHYSICAL ACTIVITY INTENTIONS:

What physical activity do you intend to do?

#### Physical Exam:

<table>
<thead>
<tr>
<th>Physical Exam:</th>
<th>Physical Activity Readiness Conveyance/Referral:</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>HT</strong></td>
<td>Bu</td>
</tr>
<tr>
<td><strong>Wt</strong></td>
<td>BP</td>
</tr>
<tr>
<td><strong>BP</strong></td>
<td>/</td>
</tr>
</tbody>
</table>

**Conditions limiting physical activity:**

- Cardiovascular
- Respiratory
- Musculoskeletal
- Abdominal
- Other

**Tests required:**

- ECG
- Exercise Test
- X-Ray
- Blood
- Urinalysis
- Other
Physical Activity Readiness
Medical Examination
(revised 2002)

ng, J., Shephard, R.J. (1992). Revision of the Physical
4-338-345.

2
Appendix D

3 Day Food Log Instruction

1) The purpose of this diary is to record all the food (including drinks) which you eat for a three day period. The three day period should include 2 weekdays and 1 weekend day.

2) Two pages are provided for each day of the three day period.

3) After each meal or snack that you eat, please write down in detail each separate food item you consumed – including the type of food (e.g. processed cheese) and the amount of food in household measures (e.g. 1 cup of cooked spaghetti). A meal will have to be listed by its separate parts (e.g. fried steak – 8 oz, French fries – 1 cup, coleslaw – 3 tbsp).

4) The best way to record the information is by carrying this diary around with you wherever you go. Before going to sleep you should look over the diary to check that you have not missed anything. Remember to include snacks!

5) If you eat fast food, you can just type the type of food you ate (e.g. 1 Big Mac, 1 large fries, 1 chocolate milkshake).

6) The following pages explain the use of household measures and the description of the food. A sample days diet sheet is given. Please take the time to read these pages as it will help to make your diet record more accurate.