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NONINVASIVE MYOGRAPHICAL ASSESSMENTS FOLLOWING
UNACCUSTOMED RESISTANCE EXERCISE

by

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A dissertation submitted in partial fulfillment of the requirements
for the degree of Doctor of Philosophy
in the Department of Learning Sciences and Educational Research
in the College of Community Innovation and Education
at the University of Central Florida
Orlando, Florida

Spring Term
2019

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ABSTRACT

Traditionally, post-exercise muscular alterations have been examined using invasive techniques that lack the ability to single out individual muscle groups. Sonomyography, tensiomyography, and electrical impedance myography allow for noninvasive skeletal muscle assessment. This project aimed to examine changes in muscle contractility and composition that occur in the early stages of recovery following unaccustomed exercise. METHODS: Twenty-one untrained adults (21.9 ± 1.9 y) performed exercise – 10×10 maximal eccentric knee extensions – with their nondominant leg. For each repetition, participants moved through 90° range of motion at $90°•s^{-1}$ with a passive return to the start position. Each set was separated by 60 seconds of rest. Sonomyography, tensiomyography, electrical impedance myography, and maximal isometric contractions of the knee extensors (RF & VL) of both legs were performed before (BL), immediately after (IP), and 24 hours post-exercise (24H). RESULTS: Peak torque and rate of torque development were unaltered in response to the eccentric protocol. Significant limb $\times$ time interactions were noted for reactance, phase angle, and delay time of the exercised VL, and echo intensity of the exercised RF. Compared to the dominant leg, the nondominant leg displayed significantly greater changes ($p < 0.05$) in VL delay time and RF echo intensity at IP. Following exercise, bilateral alterations were identified for reactance, phase angle, maximal displacement, delay time, contraction velocity, cross-sectional area, and thickness of the RF, as well as resistance, echo intensity, cross-sectional area, and thickness of the VL. No between-sex differences were noted in response to exercise. CONCLUSIONS: In the absence of performance decrements, sonomyography,

tensiomyography, and electrical impedance myography successfully detected acute changes in skeletal muscle composition and function following an acute bout of eccentric exercise in untrained men and women. While the exercised leg exhibited specific responses in delay time and echo intensity, bilateral changes are theorized to have occurred due to contralateral stabilization of the non-exercised leg. The current results suggest that different muscle actions (i.e., eccentric and isometric) promote similar consequences to muscle strength, size, echogenicity, contractility, and bioelectrical properties.

This work is dedicated to those who saw and believed in my potential when all I saw

were shortcomings.

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LIST OF ABBREVIATIONS

BIS	Bioelectrical Impedance Spectroscopy
CSA	Cross-Sectional Area
Dm	Maximal Displacement
EI	Echo Intensity
EIM	Electrical Impedance Myography
EIMD	Exercise-Induced Muscle Damage
MT	Muscle Thickness
MVIC	Maximal Voluntary Isometric Contraction
PhA	Phase Angle
PTQ	Peak Torque
R	Resistance
RF	Rectus Femoris
RTD	Rate of Torque Development
SFT	Subcutaneous Fat Thickness
SM	Sonomyography
Tc	Contraction Time
Td	Delay Time
TMG	Tensiomyography
Vc	Contraction Velocity
VL	Vastus Lateralis
Xc	Reactance

CHAPTER ONE: INTRODUCTION

Resistance exercise is understood to elicit temporary muscle damage which commonly manifests as heightened muscle soreness, reduced muscle function, inflammation, and transient muscle edema (Dennis et al., 2004; Fleckenstein, Canby, Parkey, & Peshock, 1988; Howatson et al., 2012). Furthermore, the magnitude of muscle damage and its concomitant side effects is greater when the exercise is unaccustomed (Chen, Chen, Lin, Wu, & Nosaka, 2009). Additionally, the nature of the exercise influences the extent of damage incurred by the muscle fibers where eccentric tasks are understood to produce greater insult (e.g., strength loss and soreness) (Gibala, MacDougall, Tarnopolsky, Stauber, & Elorriaga, 1995). Damage from eccentric exercise is postulated to cause sarcomere disruption and damage to the sarcolemmal membrane which leads to disturbances to the excitation-contraction coupling system and a subsequent reduction in force-generating capabilities (Proske & Morgan, 2001; Proske & Allen, 2005). Though numerous methods exist for the evaluation of acute responses to damaging exercise, many of these techniques are invasive (e.g., tissue biopsy or blood collection) or lack the ability to single out individual muscles (Warren, Lowe, & Armstrong, 1999). Therefore, further examination of alternative, noninvasive assessment tools is warranted to better comprehend the immediate and short-term changes that occur following a single bout of unaccustomed eccentric exercise.

As performance is understood to suffer in the wake of damaging exercise, it is commonplace to assess strength decrements in order to establish the magnitude of

damage incurred. Reductions in maximal voluntary contractile force have served as a primary marker of exercise-induced muscle damage (Hunter et al., 2012; Sayers & Clarkson, 2001), however tensiomyography (TMG) has gained attention in recent years for its ability to noninvasively evaluate passive contractile characteristics of individual skeletal muscles. With the understanding that the transmission of mechanical force is dependent upon integrity of the sarcolemmal membrane, it is reasonable to assume that muscle contractility would be affected following a damaging bout of eccentric exercise.

Changes in TMG parameters have been observed following various resistance exercise protocols (de Paula Simola et al., 2015; García-Manso et al., 2012; Hunter et al., 2012). Impaired muscle contractility has also been reported in response to electrically-induced fatigue (MacGregor, Ditroilo, Smith, Fairweather, & Hunter, 2016). Furthermore, significant correlations have been identified between alterations in TMG contractile properties and decreases in maximal force production after strength training (de Paula Simola et al., 2015; Hunter et al., 2012). TMG has also been regarded as a useful tool for detecting injury risk among athletes through the assessment of muscular asymmetries (Alentorn-Geli et al., 2015; MacGregor, Hunter, Orizio, Fairweather, & Ditroilo, 2018). Therefore, TMG may be considered an effective method to gauge the extent of exercise-induced insult to skeletal muscle.

Bioelectrical impedance is a noninvasive technique that evaluates the electrical properties (i.e., resistance and reactance) of bodily tissues via the transmission of a mild, high-frequency electrical current (Sanchez & Rutkove, 2017b) and can be used to assess the

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presence and extent of soft tissue damage by approximating cell membrane integrity

(reactance) and fluid distribution (resistance) (Lukaski & Moore, 2012; Nescolarde et al.,

2013, 2015). Moreover, the relationship between resistance and reactance – known as

phase angle – provides further information regarding cellular health. While total body

impedance spectroscopy (BIS) provides valuable information regarding body

composition, hydration status, and overall cellular heath, it assumes the body is an

isotropic conductor with uniform length and cross-sectional area (Kushner, 1992).

However, alternate methods and applications of bioelectrical impedance have since

emerged to overcome the previously mentioned limitations and gain a more

comprehensive analysis of individual body segments and specific regions of interest.

Electrical impedance myography (EIM) allows for a more localized impedance analysis

of individual muscles and muscle groups and can provide insight regarding the tissue's

composition and structure (Sanchez & Rutkove, 2017b).

Alterations in muscle composition have been detected by EIM during contraction

(Clemente, Romano, Bifulco, & Cesarelli, 2014; Li, Shin, Li, Li, & Zhou, 2016;

Shiffman, Aaron, & Rutkove, 2003; Zagar & Krizaj, 2008) as well as after exercise

(Alomar et al., 2013; Fu & Freeborn, 2018; Sanchez et al., 2017). EIM has also been

shown to aid in the evaluation of neuromuscular diseases and myopathies by

distinguishing healthy from affected tissue (Sanchez & Rutkove, 2017a). When

monitoring the physiological changes that take place during isometric contraction,

researchers observed increases in local resistance and reactance (Kashuri, Aaron, &

Shiffman, 2007; Shiffman et al., 2003); these parameters reportedly decrease following

insult (Fu & Freeborn, 2018; Li et al., 2016; Sanchez et al., 2017). Additionally, local resistance and reactance have been shown to decrease proportionally with the formation of edema and disruption of tissue structure in an injured state (Nescolarde et al., 2015, 2017). In this sense, EIM assists with the identification and assessment of muscle injury by evaluating local fluid and muscle status changes.

Changes in muscle composition can also be explored by way of localized ultrasound imaging. Skeletal muscle ultrasonography, or sonomyography, is an increasingly popular method used to quickly and painlessly analyze quantitative and qualitative properties of tissue. Through the use of sonomyography it is possible to monitor acute changes in muscle size and echo intensity (EI), which appear to be related to the extent of muscle damage (Chen, Lin, Chen, Lin, & Nosaka, 2011; Chen & Nosaka, 2006; Radaelli, Bottaro, Wilhelm, Wagner, & Pinto, 2012). Following damaging exercise, the affected musculature exhibits a temporary increase in size that is generally attributable to local swelling through the accumulation of water and/or hyperemia (Kristiansen et al., 2014; Matta, Pereira, Radaelli, Pinto, & Oliveira, 2017; Nosaka & Clarkson, 1996; Radaelli et al., 2014). Interpretation of the acute change in EI observed after resistance exercise is less understood, as there are a number of known contributing factors (McGregor, Cameron-Smith, & Poppitt, 2014). While some suggest the temporary increase in EI following exercise is due to glycogen depletion (Hill & San Millan, 2014; Nieman, Shanely, Zwetsloot, Meaney, & Farris, 2015), others relate the change to exercise-induced ultrastructural damage to the muscle (Nosaka & Clarkson, 1996; Radaelli et al., 2012).

Lastly, skeletal muscle is known to exhibit sexual dimorphisms in features such as contractility (Kim, Chai, Kim, Kim, & Bae, 2017; Rodriguez-Ruiz, Rodriguez-Matoso, Quiroga, Sarmiento, & Da Silva-Grigoletto, 2011); muscle stiffness (Martín-san Agustín, Benítez-Martínez, Medina-Mirapeix, & Casaña-Granell, 2018); fatigue resistance (Hicks, Kent-Braun, & Ditor, 2001); and muscle size, thickness, and EI (Arts, Pillen, Schelhaas, Overeem, & Zwarts, 2010). Furthermore, women tend to experience a lesser degree of muscle damage (Hicks, Onambélé, Winwood, & Morse, 2016; Sewright, Hubal, Kearns, Holbrook, & Clarkson, 2008) and inflammation (Stupka et al., 2000) following exercise which may be a function of estrogen's antioxidative and membrane stabilizing properties (Kendall & Eston, 2002; Wiseman & Quinn, 1994). However, it is unclear whether or not men and women experience similar performance decrements following a single bout of unaccustomed exercise. While sex differences in strength loss have been affirmed (Radaelli et al., 2014; Sewright et al., 2008) and contested following resistance exercise (Hicks et al., 2001; Sayers & Clarkson, 2001), the presence of sex differences in muscle contractility following exercise have yet to be elucidated.

Purposes

The aims of this research are to:

1. Determine if the changes in tensiomyography, sonomyography, and electrical impedance myography reflect functional deficits imposed by unaccustomed eccentric exercise, and

2. Explore the presence of sex-based myographical differences in functional deficits imposed by eccentric exercise.

<u>Hypotheses</u>

It is hypothesized that:

- Changes in myographical assessments of skeletal muscle will reflect the functional deficits following unaccustomed eccentric exercise, and

- Men and women will exhibit different responses to a single bout of unaccustomed eccentric exercise as assessed by tensiomyography, sonomyography, and electrical impedance myography.

CHAPTER TWO: REVIEW OF LITERATURE

Insult from Eccentric Exercise

Repeated muscular contractions – such as those performed during a bout of resistance exercise – are known to promote fatigue (Proske & Allen, 2005). This temporary state of reduced force-generating capability is accredited to such factors as metabolic exhaustion, myofilament damage, and excitation-contraction coupling system dysfunction (Allen, Lamb, & Westerblad, 2008; D. L. Morgan & Allen, 1999; Proske & Morgan, 2001). While performance is generally regained within hours of performing concentric and isometric contractions, eccentric contractions induce further damage to the muscle tissue that prompts a series of events known to compromise muscle function (Proske & Allen, 2005).

During eccentric actions, the exercising muscle produces tension as the muscle fibers are being stretched. The aforementioned tension is created through the interaction of myofilaments actin and myosin, where the extent of their overlap determines the amount of tension generated. According to the popping sarcomere hypothesis (Clarkson & Hubal, 2002; Morgan, 1990; Morgan & Proske, 2004), when an eccentrically-active muscle is stretched beyond its optimum length, a portion of its sarcomeres become overstretched to the point where myofilament overlap is no longer possible. In the event of serial eccentric contractions, sarcomeric disruption spreads and, ultimately, results in damage to various elements of the muscle fiber (Evans, Knight, Draper, & Parcell, 2002). Damage to these structures – including the sarcolemma, sarcoplasmic reticulum, and transverse tubules – is postulated to interfere with the excitation-contraction coupling system, and is thereby

responsible for compromising the muscle's ability to generate tension (Morgan & Allen, 1999; Proske & Morgan, 2001; Proske & Allen, 2005). Additionally, eccentric exercise-induced damage causes a rise in muscle stiffness lasting several days (Chleboun, Howell, Conatser, & Giesey, 1998; Cleak & Eston, 1992; Jones, Dewham, & Clarkson, 1987; Whitehead, Weerakkody, Gregory, Morgan, & Proske, 2001) which has been found to coincide with reduced force production during MVIC (MacGregor et al., 2016).

<div align="center">Performance Decrements following Eccentric Exercise</div>

High-force eccentric tasks result in a 50-60% reduction in force-generating capacity immediately after exercise (Clarkson, Nosaka, & Braun, 1992; Clarkson & Hubal, 2002; Sayers & Clarkson, 2001). These reductions in force are typically greatest immediately post-exercise and recover toward baseline levels in the following days (Chen et al., 2011; Hunter et al., 2012). Immediate decreases in peak force/torque following eccentric exercise have been observed among non-resistance trained men (Arroyo et al., 2017; Gibala et al., 1995) and women (Bloomer, 2004; Child, Saxton, & Donnelly, 1998; Gonzalez-Izal, Cadore, & Izquierdo, 2014). Furthermore, these performance decrements persist for days following a bout of eccentric exercise (Byrne, Eston, & Edwards, 2001; Howell, Chleboun, & Conatser, 1993; Radaelli et al., 2012, 2014) where full strength recovery may not be reached until weeks later (Howell et al., 1993; Sayers & Clarkson, 2001). However, with the understanding that exercise-induced muscle damage elicits side effects such as muscle soreness, swelling, and stiffness (Howell et al., 1993; Kazunori Nosaka, Newton, & Sacco, 2002, p.), it is plausible that a portion of the observed reductions in force production are a function of an increased rating of perceived exertion.

<div align="center">8</div>

Accordingly, a less taxing assessment might be more appropriate for the examination of exercise-induced muscle damage so as to avoid further insult (de Paula Simola et al., 2015).

<p style="text-align: center;">Changes in Contractile Properties</p>

Changes in skeletal muscle contractile and mechanical properties associated with exercise-induced muscle damage and fatigue are of particular interest. While performance decrements are commonly evaluated via maximal voluntary force production (i.e., MVIC), the results of said assessment generally provide information regarding an entire muscle group. In addition, MVIC requires voluntary motivation for one to exert maximal force and may induce more damage and/or fatigue upon an already affected muscle. Alternatively, tensiomyography (TMG) is a noninvasive method by which researchers can assess *in vivo* passive contractile properties and neuromuscular function of individual skeletal muscles (de Paula Simola et al., 2015; Hunter et al., 2012; Martin-Rodriguez, Loturco, Hunter, Rodriguez-Ruiz, & Munguia-Izquierdo, 2017). In recent years, TMG has been applied in the contexts of injury, rehabilitation, and performance as it may be used to assess recovery from fatiguing and/or damaging exercise, determine muscle tone and stiffness, estimate muscle fiber composition, and quantify muscle contractility (Dahmane, Djordjevič, Šimunič, & Valenčič, 2005; MacGregor et al., 2018; Pišot et al., 2008; Valenčič & Knez, 1997).

To assess the tensiomyographical properties of a muscle, electrical stimulation is first introduced by way of surface electrodes on the skin superjacent to the muscle belly of

interest. A displacement sensor is then positioned centrally between the electrodes; the sensor ultimately measures the muscle's responses to the electrical stimuli. Due to the near-constant volume nature of skeletal muscle, the transverse diameter changes in relation to muscle length (Baskin & Paolini, 1967; MacGregor et al., 2018). Therefore, the TMG sensor measures radial displacement/deformation of a muscle as it contracts and shortens along its longitudinal axis. In addition to maximal radial displacement (Dm), TMG also provides information regarding the length of time needed to reach Dm (contraction time; Tc) as well as the muscle's responsiveness to the electrical stimulus, portrayed as the delay time of contraction (Td). It should be noted, however, that Tc is influenced by Dm, where a greater Dm requires a longer Tc. Therefore, a measure independent of Dm should also be calculated in order to determine the rate of muscle contraction (contraction velocity, Vc) (MacGregor et al., 2018).

Local fatigue and changes in muscle contractility have been previously detected through the use of TMG following both endurance (García-Manso et al., 2011) and resistance exercise (de Paula Simola et al., 2015; García-Manso et al., 2012; Hunter et al., 2012). As a reflection of muscle stiffness and contractile force (Dahmane et al., 2005; García-Manso et al., 2012), Dm decreases rapidly in response to resistance exercise (de Paula Simola et al., 2015; García-Manso et al., 2012; Hunter et al., 2012) and appears to mimic the pattern of other commonly assessed markers of exercise-induced muscle damage (e.g., peak torque and rate of torque development) (Hunter et al., 2012). Furthermore, these changes in contractility have been reported among trained (de Paula Simola et al., 2015; García-Manso et al., 2012) and untrained adults (Hunter et al., 2012). Reductions

in Vc have also been observed immediately after performing resistance exercise (de Paula

Simola et al., 2015). Conversely, Tc has been shown to increase following eccentric

exercise (Hunter et al., 2012) and high-intensity interval training (Wiewelhove et al.,

2015) which may be demonstrative of impaired EC coupling and/or reduced cell

membrane integrity.

Ultrastructural Muscle Damage

Electrical impedance myography (EIM) is a relatively new technique that has been

implemented as a noninvasive method of skeletal muscle assessment. By applying high-

frequency, low-intensity electrical currents, EIM provides feedback detailing the overall

state of the muscle (Rutkove, 2009). Similar to other forms of bioelectrical impedance

analysis, EIM aims to quantify the passive electrical properties of muscle tissue and is

based on the principle that these tissues act as conductors and insulators. The electrical

impedance properties of a given tissue are influenced by such features as membrane

integrity, cell volume and structure, as well as fluid within the tissue (Fu & Freeborn,

2018). While lean tissue (i.e., skeletal muscle) is relatively high in water content and

electrolytes and is conductive, other tissues such as fat and bone contain a relatively low

amount of water and act as insulators (Kushner, 1992). Furthermore, all cells are

enveloped in a lipid bilayer – an insulating layer between two conductive layers – which

contributes largely to tissues' electrical properties. Briefly, EIM involves sending a

current into a discrete region of muscle tissue and measures the consequent voltage

(Rutkove, 2009). As the electrical current travels through a given tissue, its amplitude is

reduced (i.e., it loses energy) due to the tissue's innate resistance. Therefore, the measured voltage alterations reflect the changes in current as the tissue resists it.

As a relatively convenient tool that requires minimal effort from the subject, EIM has been used for the assessment of neuromuscular disorders (Li, Jafarpoor, Bouxsein, & Rutkove, 2014; Rutkove et al., 2014; Sanchez & Rutkove, 2017a) and traumatic muscle injuries (Nescolarde et al., 2013, 2015, 2017) as it can provide detailed physiological information regarding the structure, composition, and overall health status of a given muscle (Rutkove, 2009; Sanchez & Rutkove, 2017b). Recent research supports that EIM may be valuable for the evaluation and quantification of exercise-induced muscle fatigue (Fu & Freeborn, 2018). Acute changes in muscle status as assessed by EIM have also been detected in healthy and diseased muscles following resistance exercise (Fu & Freeborn, 2018; Sanchez et al., 2017). Specifically, the customary muscle edema observed following damaging exercise has been found to prompt a decrease in local resistance (Nescolarde et al., 2017; Sanchez et al., 2017), while the loss of cell membrane integrity after exercise causes a lesser reactance (Nescolarde et al., 2015). Furthermore, the reductions in resistance and reactance are believed to coincide with the severity of muscle insult (Nescolarde et al., 2015). However, because EIM does not offer a visual interpretation of the muscle tissue, combining it with an imaging technique such as ultrasonography may provide more comprehensive insight (Li et al., 2016; Murphy, Skinner, Martucci, Rutkove, & Halter, 2018; Rutkove et al., 2014).

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Qualitative Muscle Composition

Ultrasound imaging is a convenient technique by which to noninvasively visualize tissues in real-time (Pillen & van Alfen, 2011). When applied to skeletal muscle, sonomyography enables the quantification of such architectural and compositional parameters as muscle thickness (Koo, Wong, & Zheng, 2010; Miyatani, Kanehisa, Ito, Kawakami, & Fukunaga, 2004; Shi, Zheng, Chen, & Huang, 2007), cross-sectional area (Noorkoiv, Nosaka, & Blazevich, 2010; Seymour et al., 2009), fascicle length (Fukunaga, Ichinose, Ito, Kawakami, & Fukashiro, 1997; Kwah, Pinto, Diong, & Herbert, 2013), fiber pennation angle (Fukunaga et al., 1997; Maganaris, Baltzopoulos, & Sargeant, 2002), and echo intensity (EI) (Chen et al., 2009; Radaelli et al., 2012). By analyzing the previously mentioned features, sonomyography allows for the evaluation of fatigue (Shi et al., 2007), swelling/local edema (Damas et al., 2016), and exercise-induced muscle damage (Cadore et al., 2018; Gonzalez-Izal et al., 2014; Wilkinson, Gould, Viana, & Watson, 2019).

While the interpretation of architectural measures of skeletal muscle is straightforward, the evaluation of EI remains abstruse. Cross-sectional examination of EI reflects intramuscular fat and fibrous tissue content (Pillen et al., 2009; Walker, Cartwright, Wiesler, & Caress, 2004; Young, Jenkins, Zhao, & McCully, 2015), where an increased EI value indicates a larger proportion of non-contractile tissue (Fukumoto et al., 2012) and lower muscle density (Sipilä & Suominen, 1993). Furthermore, longitudinal examination has proven its manipulability to a structured resistance training regimen (Cadore et al., 2014; Radaelli et al., 2014; Scanlon et al., 2014; Wilhelm et al., 2014). It

should also be noted that the observed changes (i.e., decreases) in EI are often accompanied by significant improvements in force production/muscle function. Thus, it is theorized that EI provides a qualitative assessment of skeletal muscle (Cadore et al., 2014; Fukumoto et al., 2012).

The concept of muscle quality is broad and remains relatively undefined due to the multitude of potentially influential factors (e.g., muscle architecture, fiber type, intramuscular fat and fibrous tissue content, etc.) (McGregor, Cameron-Smith, & Poppitt, 2014). While some use the term to describe muscle strength or power per unit of muscle mass (Goodpaster et al., 2006; McGregor, Cameron-Smith, & Poppitt, 2014), the quality of skeletal muscle is generally determined according to its composition and functionality (Correa-de-Araujo et al., 2017). Muscle composition in this context has been previously estimated via examination of the extracellular (ECW) to intracellular water (ICW) ratio (Taniguchi et al., 2017; Yamada et al., 2017), bioimpedance-derived phase angle (Bourgeois et al., 2018), and EI values (Bourgeois et al., 2018; Watanabe et al., 2013) as they mirror the ratio of contractile to non-contractile tissue. Unfavorable muscle composition (i.e., high ECW/ICW ratio, elevated EI) is negatively associated with muscle function, particularly among older adults (Cadore et al., 2012; Taniguchi et al., 2017; Watanabe et al., 2013). However, is unclear if poor muscle quality is a function of advanced age or disuse.

Acute changes in EI are less understood. Jajtner and colleagues (Jajtner et al., 2015) observed a significant increase in quadriceps EI among trained men immediately after

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completion of a high-volume resistance exercise protocol. However, this response had

subsided after 24 hours. By contrast, others did not observe an increase in elbow flexor

EI among untrained adults until 24 (Radaelli et al., 2012) or 48 hours (Radaelli et al.,

2014) after exercise. This delayed rise in upper and lower limb EI among untrained men

has been reported elsewhere (Chen et al., 2011). It is unclear whether these discrepant

findings reflect differences in participant training status, the exercise intervention

employed, the muscles examined, or a combination thereof. Interestingly, the pattern of

early exercise-induced alterations in EI does not consistently align with that of other

markers of post-exercise recovery and inflammation (Chen et al., 2011; Fujikake, Hart, &

Nosaka, 2009; Jajtner et al., 2015; Radaelli et al., 2012, 2014). Some relate the short-term

change in EI to post-exercise depletion of glycogen (Hill & San Millan, 2014; Nieman et

al., 2015), while others suggest the change is due to edema (Pillen & van Alfen, 2011)

and ultrastructural damage (Matta et al., 2017; Radaelli et al., 2012). However, the

interpretation of acute alterations in EI as they relate to changes in muscle quality and

function remain unclear.

Sexual Dimorphism in Skeletal Muscle & Performance

Finally, skeletal muscle exhibits sex-based differences in a number of properties. Men

generally demonstrate greater muscle mass and lower EI values (Arts et al., 2010; Miller,

MacDougall, Tarnopolsky, & Sale, 1993), but it is not yet known if this translates to a

superior muscle quality. Men also tend to display greater muscle stiffness, which some

hypothesize is due to greater muscle size (Blackburn, Riemann, Padua, & Guskiewicz,

2004). However, the extent of muscle stiffness is inversely related to its contractility such

that a decrease in stiffness equates to an increase in Dm (Pišot et al., 2008). Therefore, it is understandable that Dm may be lower in men than women. This concept has been observed in some quadriceps muscles among male and female athletes (Martín-san Agustín et al., 2018; Rodriguez-Ruiz et al., 2011), but further investigation is warranted to rule out the influence of sport-specific training adaptations.

Regarding the response to exercise, men tend to experience greater exercise-induced muscle damage and its associated symptoms (Hicks et al., 2016; Sewright et al., 2008). This has been evidenced by higher concentrations of circulating creatine kinase (Hicks et al., 2016; Sewright et al., 2008) and inflammatory cell infiltration (Stupka et al., 2000) in men compared to women following eccentric exercise. Additionally, men have been shown to exhibit lower fatigue resistance than women (Hicks et al., 2001; Russ & Kent-Braun, 2003). It is theorized that these advantageous outcomes observed in women are attributable to estrogen's antioxidative and membrane stabilizing properties which decrease the susceptibility of exercised muscle(s) to damage (Kendall & Eston, 2002; Wiseman & Quinn, 1994). Interestingly, sex differences in fatigability appear to dissipate when a) exercise intensity approaches maximal exertion (Ditor & Hicks, 2000) and 2) exercise is performed under ischemic conditions (Russ & Kent-Braun, 2003). Sex differences in performance decrements following damaging exercise are less consistent. That is, immediate decreases in strength have been observed in men, but not women (Radaelli et al., 2014) and vice versa (Sewright et al., 2008), following an acute bout of resistance exercise while others observed no such difference (Hicks et al., 2001; Sayers & Clarkson, 2001). Moreover, it is currently unknown if sex differences exist in TMG

parameters following an acute bout of unaccustomed eccentric exercise. Further research

is necessary to better understand the functional deficits experienced by men and women

following an acute bout of unaccustomed resistance exercise.

CHAPTER THREE: METHODOLOGY

Participants

Twenty-one healthy adults took part in this research study (Table 1). All participants were non-resistance trained; free of any physical limitations and chronic illnesses; had no recent injuries or surgeries; were not taking any prescription or over-the-counter medications, dietary supplements, performance-enhancing drugs, or anti-inflammatory drugs; and did not regularly lift or carry heavy objects. Following an explanation of all procedures, risks, and benefits, each participant provided his/her informed consent prior to participation in this study. The research protocol was approved by the UCF Institutional Review Board prior to participant enrollment.

Experimental Design

This study utilized a repeated-measures, mixed factorial design to examine the effects of unaccustomed eccentric exercise on changes in myographical parameters and to compare the response between sexes. Participants reported to the Human Performance Laboratory (HPL) on three separate occasions. During the first visit (D0), a researcher explained all procedures, risks, and potential benefits to each participant. Following the eligibility screening, anthropometric measures (i.e., height and weight) were obtained and a familiarization of the exercise protocol was conducted. During the second visit (D1), participants underwent a battery of myographical assessments performed on each leg individually prior to (BL) and immediately after (IP) completing a unilateral lower-body resistance exercise protocol. Participants were then instructed to report back to the HPL 24 hours post-exercise (D2) to undergo one final bout of myographical assessments. For

D1 and D2, participants were asked to refrain from alcohol consumption for 12 hours

prior and to avoid exercise, food, and caffeine consumption for 2 hours prior to arrival to

the HPL.

Eccentric Exercise Protocol

Following the BL assessments on D1, participants completed an exercise protocol

consisting of ten sets of ten repetitions of maximal eccentric knee extensions using the

nondominant leg. Participants performed the exercise in a seated position moving through

a range of motion of 90° ± 10°. Each eccentric action was followed by a passive return to

the start angle. The rest interval between each set was 60 seconds. Participants were

verbally encouraged to give maximal effort during the exercise session.

Anthropometric and Myographical Assessments

Anthropometric Assessment

During D0, participants' body weight and height was recorded. Height and weight were

assessed via a digital physician scale with a height rod attachment (Health o meter®

500KL, Sunbeam Products, Inc., Boca Raton, FL, USA). Height was measured to the

nearest 0.5 cm, while body weight was measured to the nearest 0.1 kg. Body weight was

reassessed at the start of each visit to track daily variations.

Hydration Assessment

As hydration status is known to impact electrical impedance analysis and physical

performance, participants were asked to provide a urine sample upon arrival to the

laboratory on D1 and D2 for analysis of urine-specific gravity (USG) by digital refractometry (Palm Abbe PA202, Misco®, Cleveland, OH, USA) to ensure euhydration (defined as USG ≤ 1.020).

Sonomyographical Assessment

Noninvasive skeletal muscle ultrasound images were obtained from each participant's right and left legs. Prior to image collection, all locations were identified using standardized anatomical landmarks for the RF and VL. For each measurement (except IP), participants were asked to lay supine on an examination table with both legs fully extended for 15 minutes to allow fluid shifts to occur. Images of the RF were captured midway between the anterior inferior iliac crest and proximal patellar border. To obtain RF images, participants were asked to lay flat in a supine position with their legs fully extended and muscles relaxed. Images of the VL were captured on the midline halfway between the greater trochanter and lateral epicondyle. To obtain VL images, participants were asked to relax their leg muscles and maintain the left lateral decubitus position with legs together and a slight (10°) bend in the knees (Bemben, 2002). All ultrasound images were captured and analyzed by the same technician.

All measures were obtained by passing a 12-MHz linear probe scanning head (General Electric LOGIQe, Wauwatosa, WI, USA) coated with water-soluble transmission gel (Aquasonic 100, Parker Laboratories, Inc., Fairfield, NJ, USA) over the surface of the thigh at the predetermined anatomical locations. Images of muscle cross-sectional area (CSA) were captured using a transverse sweep in the extended-field-of-view mode, while

images of muscle thickness (MT) were captured using B-mode. For CSA and MT, three consecutive images were captured and analyzed for each muscle and leg, respectively. The average of the three measurements was used for subsequent statistical analyses. For all images, the probe was positioned on the surface of the skin without depressing the dermal layer; gain was set at 50, image depth was fixed at 7 cm, and dynamic range was set at 72 to optimize spatial resolution.

Further analysis of all ultrasound images was performed using image processing software (ImageJ, National Institutes of Health, Bethesda, MD, version 1.52g) to quantify CSA, MT, and EI. To determine CSA, the perimeter of each muscle was traced using the polygon selection tool taking care to include as much muscle tissue as possible while excluding surrounding fascia. Then, EI was determined as the average gray value of the selected region. To determine MT, a straight line was drawn perpendicular to and spanning the distance between the deep and superficial aponeuroses using the line selection tool. Subcutaneous fat thickness (SFT) values were obtained from the CSA images of each muscle and were calculated as the average thickness of the subcutaneous layer measured laterally, centrally, and medially above the superficial muscle border. These SFT values were then used to calculate corrected EI values using the following equation:

$$\text{Corrected EI} = \text{raw EI} + (\text{SFT} \times 40.5278) \text{ (Young et al., 2015)}$$

ICC values were 0.99 (SEM = 0.36 cm^2), 0.99 (SEM = 0.58 cm^2), 0.96 (SEM = 0.11 cm), 0.98 (SEM = 0.06 cm), 0.99 (SEM = 0.10 cm), 0.99 (SEM = 0.08 cm), 0.90 (SEM = 6.46 au), and 0.78 (SEM = 0.78 au) for RF CSA, VL CSA, RF MT, VL MT, RF SFT, VL SFT, RF EI, and VL EI, respectively.

Bioelectrical Impedance Analysis & Electrical Impedance Myography

For whole-body impedance analysis, a bioelectrical impedance spectrometer was used (ImpediMed SFB7, ImpediMed Inc., Carlsbad, CA, USA). Impedance analysis took place while the participants assumed a supine position on a nonconductive surface. Two pairs of electrodes were placed on the participant's right side. The injecting electrodes were placed on the dorsal surfaces of the hand and foot on the metacarpals and metatarsals, respectively. The sensing electrodes were placed between the medial and lateral malleoli and between the radial and ulnar styloid processes. Injecting and sensing electrodes were placed 5 cm apart. The spectrometer measured whole-body impedance at 256 frequencies between 4 and 1000 kHz to determine tissue resistance and reactance.

The abovementioned spectrometer was also employed for single-muscle analysis. Two pairs of electrodes were positioned over the belly of the muscle being examined. The sensing electrodes were placed 5 cm proximal and distal to the midpoint of the muscle (same site determination as described in previous section), and the injecting electrodes were placed close to the others. This process was replicated for the right and left legs. ICC values were 0.97 (SEM = 4.26 ohms), 0.99 (SEM = 4.12 ohms), 0.97 (SEM = 0.72

ohms), 0.96 (SEM = 1.08 ohms), 0.99 (SEM = 0.67°), and 0.99 (SEM = 0.42°) for RF

resistance, VL resistance, RF reactance, VL reactance, RF phase angle, and VL phase

angle, respectively.

Tensiomyographical Assessment

To assess contractile properties of the RF and VL, TMG measures (TMG Measurement

System, TMG-BMC Ltd., Ljubljana, Slovenia) were recorded. The following variables

were documented for the RF and VL of both legs: delay time (Td), contraction time (Tc),

and maximal radial displacement (Dm). Contraction velocity (Vc) was subsequently

calculated as: [Dm/(Tc + Td)].

For each muscle, the TMG sensor tip was placed at a predetermined location.

Measurements of the RF and VL were performed while participants assumed a supine

position with muscles relaxed and the leg atop a triangular wedge foam cushion to

maintain a fixed knee angle of 120°. A digital displacement transducer, which

incorporates a spring of 0.17 N m^{-1}, was set perpendicular to the muscle belly to acquire

Dm of the selected muscle. The site for measurement of the RF was determined by

placing a transversal mark at the halfway point between the greater trochanter and the

lateral condyle of the proximal and distal ends of the femur, respectively. The participant

was then instructed to contract his/her quadriceps muscle of the leg being examined to

palpate the RF, at which time a line was drawn longitudinally across the transversal line,

thus creating an "X" landmark for the sensor tip placement. The site for measurement of

the VL was located at 30% of the femur length above the patella on the lateral side

(Šimunič et al., 2011). Two square (5 × 5 cm), 2-mm thick self-adhesive electrodes were placed symmetrically ~5 cm distal and proximal to the sensor tip. This procedure was replicated for the right and left legs.

For each electrical stimulation, the pulse duration was 1 ms and the initial current amplitude was set at 30 mA. For each test, current amplitude was progressively increased by 10 mA increments until no further increases in Dm were observed. ICC values were 0.99 (SEM = 0.40 mm), 0.99 (SEM = 0.24 mm), 0.94 (SEM = 1.49 ms), 0.95 (SEM = 0.47 ms), 0.99 (SEM = 0.71 ms), 0.98 (SEM = 0.84 ms), 0.99 (SEM = 0.01 mm/s^{-1}), and 0.99 (SEM = 0.01 mm/s^{-1}) for RF Dm, VL Dm, RF Td, VL Td, RF Tc, VL Tc, RF Vc, and VL Vc, respectively.

Strength Assessment

Isometric force production was assessed using an isokinetic dynamometer (Biodex Medical System, Shirley, NY, USA). After warming up on a cycle ergometer for five minutes at a self-selected cadence, participants were seated and stabilized in the device, and the leg being tested was secured to the lever arm. Participants were then instructed to perform three submaximal, warm-up repetitions at approximately 50% of their maximal perceived effort before performing three maximal voluntary isometric contractions (MVIC) with each leg individually. Each MVIC was three seconds in duration and was performed at 80° knee flexion. A 60-second rest period was provided between each MVIC. Onset of contraction for all MVICs were detected manually by the same investigator as the point at which force increased above baseline amplitude. Eccentric

peak torque was recorded for each repetition during the first and last sets of the exercise protocol. The average eccentric peak torque was calculated for subsequent analysis. The ICC values for peak torque and rate of torque development (0-300 ms) were determined to be 0.85 (SEM = 18.78 N m) and 0.86 (SEM = 70.92 N m/s^{-1}), respectively.

<div align="center">Statistical Analyses</div>

Separate three-way (sex $\times$ limb $\times$ time) repeated-measures analyses of variance (ANOVA) were used to evaluate the influence of sex on changes in performance and myographical assessments among both limbs. Separate two-way repeated-measures ANOVA were used to examine the influence on sex on changes in individual whole-body myographical parameters. Separate one-way repeated-measures ANOVA were used to examine changes in eccentric peak torque, average eccentric peak torque, and total work performed during exercise. Independent-samples *t*-tests were used to determine baseline differences between sexes, while paired-samples *t*-tests were used to determine baseline differences between limbs as well as to evaluate the change in average eccentric peak torque from the first to the last set of eccentric exercise. The effect sizes for the previously mentioned *t*-tests were determined using Cohen's *d*, where values of 0.20, 0.50, 0.80, and 1.30 were interpreted as small, medium, large, and very large effect sizes, respectively (Cohen, 2013). In the event of a significant interaction, Bonferroni *post hoc* tests were used for pairwise comparisons. Pearson product-moment correlations were used to determine relationships between changes in variables recorded on the nondominant leg. Data were analyzed using IBM® SPSS Statistics (version 25.0, IBM Inc. Armonk, NY, USA). Significance was accepted at an alpha level of $p \leq 0.05$.

CHAPTER FOUR: RESULTS

Twenty-one healthy, non-resistance trained men ($n = 9$) and women ($n = 12$) volunteered to participate in this investigation. All participants were euhydrated prior to testing on D1 and D2, and all participants completed the eccentric protocol in its entirety. Furthermore, all female participants completed testing during the mid-follicular phase of their respective menstrual cycles. Participant characteristics are displayed in Table 1, and baseline differences between sexes and limbs are displayed in Tables 2 and 3, respectively.

Table 1: Participants characteristics

	Age (y)	Body fat (%)	Height (m)	Weight (kg)	BMI (kg/m^2)
Total ($n = 21$)	21.9 ± 1.9	33.4 ± 8.4	167.3 ± 9.7	87.2 ± 26.5	31.2 ± 8.7
Men ($n = 9$)	21.6 ± 2.2	31.6 ± 14.3	174.7 ± 5.9	93.3 ± 35.4	30.5 ± 11.1
Women ($n = 12$)	22.1 ± 1.7	36.6 ± 5.2	161.8 ± 8.1	82.7 ± 17.7	31.7 ± 6.9

Values are means $\pm$ SD.

Table 2: Baseline sex differences

	Men ($n = 9$)	Women ($n = 12$)	p-value	Effect size (d)
Dominant				
RF raw EI (au)	98.18 ± 7.33	109.05 ± 13.03	0.047	1.03
RF SFT (cm)	1.15 ± 0.86	1.86 ± 0.43	0.025	1.18
VL SFT (cm)	1.09 ± 0.66	1.85 ± 0.34	0.003	1.64
RF R (Ω)	98.65 ± 28.53	133.52 ± 21.58	0.006	1.50
VL R (Ω)	92.72 ± 26.25	129.15 ± 13.71	0.001	1.96
RF PhA (°)	6.75 ± 4.58	3.64 ± 1.33	0.038	1.08
VL PhA (°)	11.14 ± 3.74	7.60 ± 1.80	0.011	1.37
VL Xc (Ω)	11.16 ± 3.74	7.60 ± 1.80	0.010	1.38
VL Dm (mm)	3.74 ± 1.89	2.02 ± 1.31	0.023	1.14
VL Vc (mm/ms^{-1})	0.09 ± 0.04	0.05 ± 0.03	0.024	1.14
RTD300 (N m/s^{-1})	463.53 ± 95.53	368.96 ± 148.94	0.013	1.27
PTQ (N m)	137.61 ± 12.58	124.15 ± 32.73	0.025	1.13
Nondominant				
VL raw EI (au)	105.61 ± 5.40	112.75 ± 7.96	0.040	1.06
RF SFT (cm)	1.19 ± 0.90	1.86 ± 0.43	0.037	1.09
VL SFT (cm)	1.10 ± 0.67	1.94 ± 0.52	0.006	1.51
RF R (Ω)	102.14 ± 27.39	135.67 ± 20.66	0.007	1.50
VL R (Ω)	93.82 ± 26.96	129.85 ± 17.16	0.002	1.77
RF PhA (°)	6.14 ± 3.99	3.43 ± 1.37	0.050	1.03
VL PhA (°)	8.21 ± 4.66	3.71 ± 1.06	0.004	1.57
VL Xc (Ω)	11.97 ± 3.09	8.34 ± 2.29	0.007	1.45

Values are means ± SD.

Table 3: Baseline differences between the dominant and nondominant legs

	p-value	Effect size (d)
VL raw EI (au)	0.034	0.37
RF PhA (°)	0.005	0.14
VL PhA (°)	< 0.001	1.04
RTD300 (N m/s^{-1})	0.014	0.36
PTQ (N m)	< 0.001	0.77

<u>Performance Measures</u>

Isometric Peak Torque (IsoPTQ)

No significant sex × limb × time ($F = 1.977$; $p = 0.152$; $\eta^2 = 0.094$), sex × time ($F = 0.983$; $p = 0.383$; $\eta^2 = 0.049$), sex × limb ($F = 0.318$; $p = 0.579$; $\eta^2 = 0.016$), or limb × time ($F = 0.669$; $p = 0.518$; $\eta^2 = 0.034$) interactions were noted for IsoPTQ. Similarly, no significant main effect of time was noted ($F = 2.309$; $p = 0.131$; $\eta^2 = 0.108$), but a

significant main effect of limb ($F = 37.070$; $p < 0.001$; $\eta^2 = 0.661$; Figure 1) was

observed where the nondominant leg displayed significantly lower IsoPTQ than the

dominant leg ($p < 0.001$; $d = 0.76$). Figures 2 and 3 display the high variability in the

change in IsoPTQ from BL to IP observed among participants' dominant and

nondominant legs, respectively.

Figure 1: Changes in IsoPTQ of the dominant and nondominant legs following
unaccustomed resistance exercise

Figure 2: Percent change in IsoPTQ of the dominant (non-exercised) leg following unaccustomed resistance exercise

Figure 3: Percent change in IsoPTQ of the nondominent (exercised) leg following unaccustomed resistance exercise

Rate of Torque Development (RTD300)

Figure 4 displays RTD300 values of the dominant and nondominant leg. No significant

sex × limb × time ($F = 2.099$; $p = 0.137$; $\eta^2 = 0.099$), sex × time ($F = 0.804$; $p = 0.455$; η^2

= 0.041), sex $\times$ limb ($F = 0.194$; $p = 0.665$; $\eta^2 = 0.010$), or limb $\times$ time ($F = 1.712$; $p = 0.194$; $\eta^2 = 0.083$) interactions were noted for isometric PTQ. However, significant main effects of limb ($F = 23.762$; $p < 0.001$; $\eta^2 = 0.556$) and time ($F = 3.701$; $p = 0.049$; $\eta^2 = 0.163$) were observed. Follow-up analyses revealed significantly lower RTD300 in the nondominant compared to the dominant leg ($p < 0.001$; $d = 0.55$), but no significant differences were identified between time points. Figures 5 and 6 display the high variability in the change in RTD300 observed from BL to IP among participants' dominant and nondominant legs, respectively.
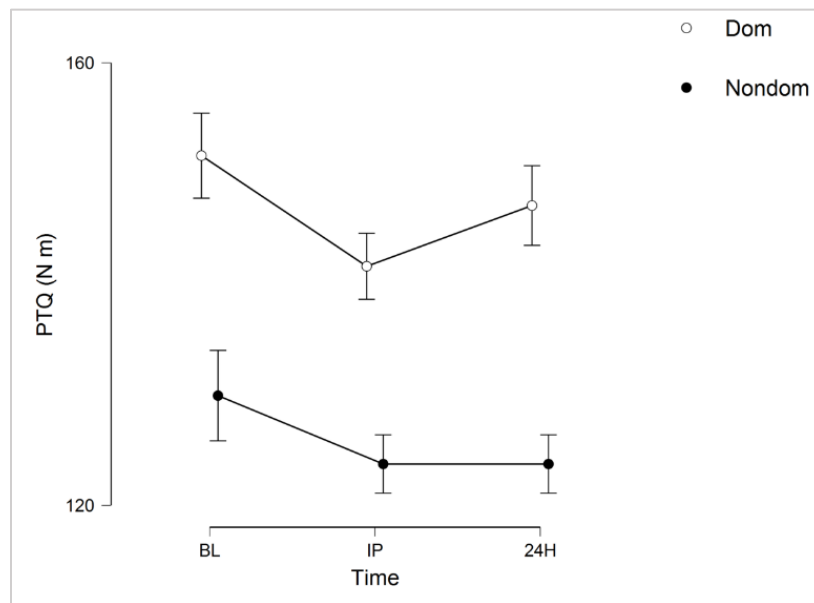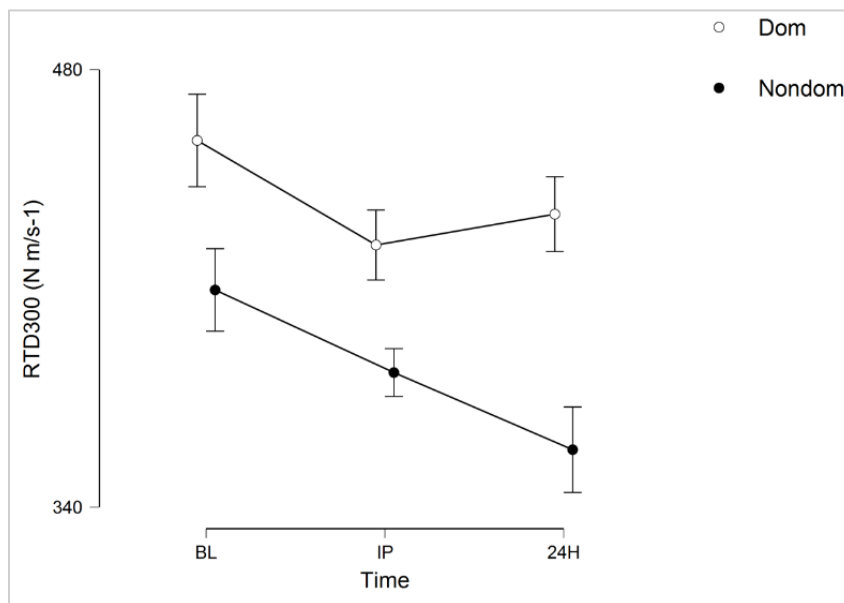

Figure 4: Changes in RTD300 of the dominant and nondominant legs following unaccustomed resistance exercise
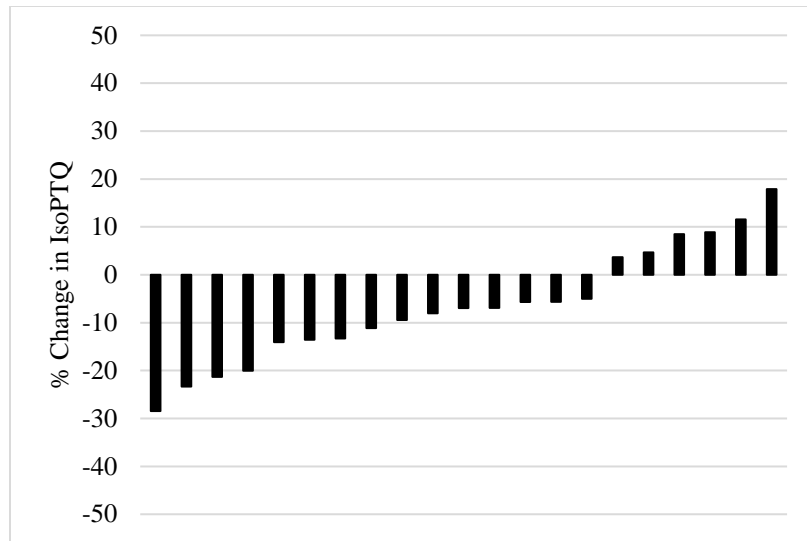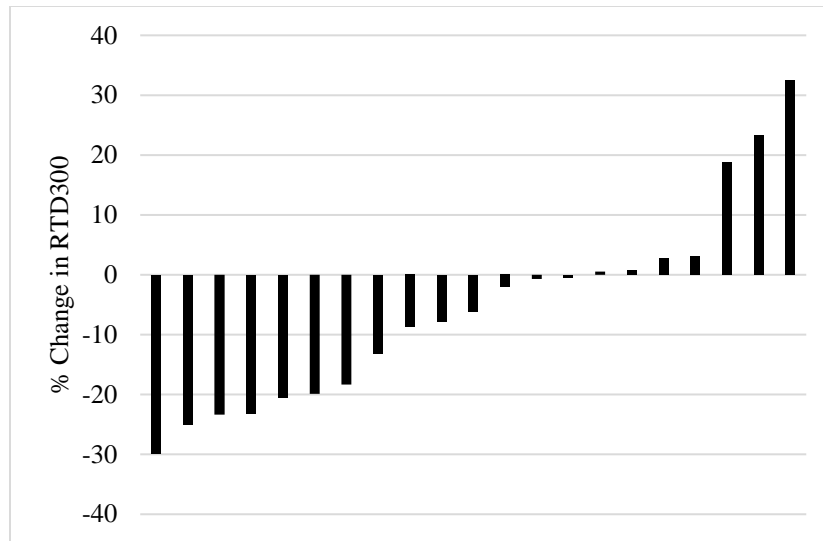
Figure 5: Percent change in RTD300 of the dominant (non-exercised) leg following unaccustomed resistance exercise
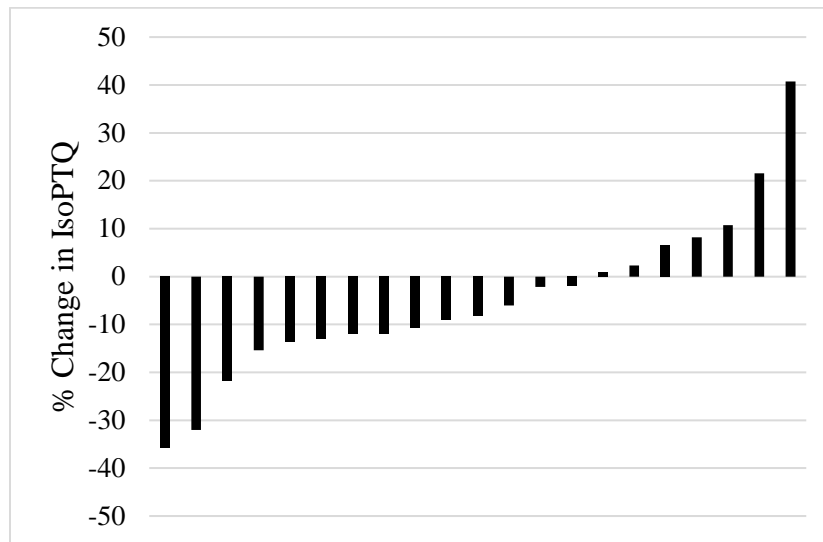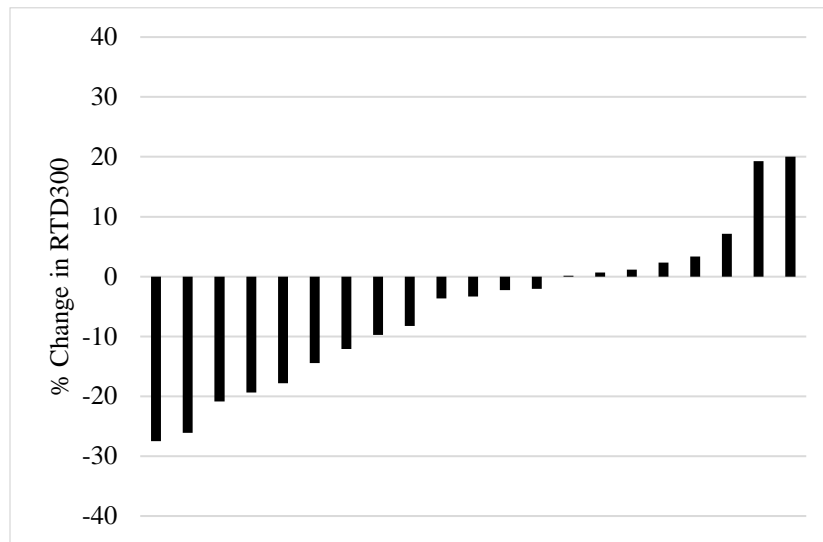

Figure 6: Percent change in RTD300 of the nondominant (exercised) leg following unaccustomed resistance exercise

Average Eccentric Peak Torque (EccPTQ$_{AVG}$)

During the unaccustomed resistance exercise session, a significant sex $\times$ time interaction

was observed for EccPTQ$_{AVG}$ ($F = 2.359$; $p = 0.016$; $\eta^2 = 0.110$), but no significant main

effect of time was noted ($F = 1.595$; $p = 0.177$; $\eta^2 = 0.077$). Further analysis revealed a significant between-sex difference during Set 1, where EccPTQ$_{AVG}$ was significantly lower among females compared to males ($p = 0.029$; $d = 1.09$). No between-sex differences were noted during Sets 2-10. Figure 7 displays the high variability in the change in PTQ$_{AVG}$ from Set 1 to Set 10 observed in the exercised leg.
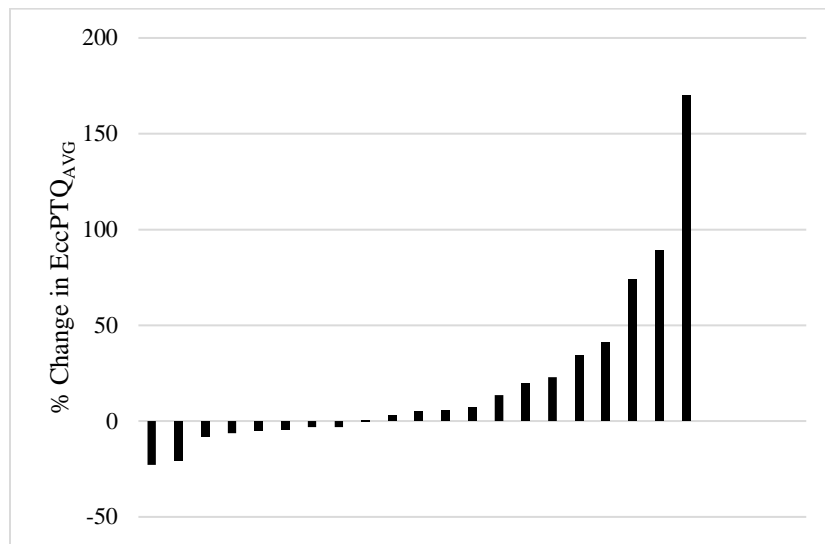
Figure 7: Percent change in PTQ$_{AVG}$ of the nondominant (exercise) leg from Set 1 to Set 10 of eccentric exercise

Eccentric Peak Torque (EccPTQ)

During the unaccustomed resistance exercise session, a significant sex × time interaction was observed for EccPTQ (F = 3.174; $p = 0.001$; $\eta^2 = 0.143$), but no significant main effect of time was noted (F = 0.753; $p = 0.560$; $\eta^2 = 0.038$). Further analysis revealed no significant between-sex differences during the eccentric exercise protocol.

Total Work

During the unaccustomed resistance exercise session, a significant sex × time interaction was observed for total work (F = 2.630; p = 0.007; η^2 = 0.122). Similarly, a significant main effect of time was noted (F = 2.153; p = 0.028; η^2 = 0.102), but no significant differences were identified between sets. Further analysis revealed a significant between-sex difference during Set 1, where total work performed by the females was significantly lower than total work performed by the males (p = 0.007; d = 1.40). No between-sex differences were noted during Sets 2-10.

Electrical Impedance Myography

Resistance (R)

No significant sex × time interaction was noted for whole-body R (F = 0.382; p = 0.686; η^2 = 0.021), but a significant main effect of time was observed (F = 8.969; p = 0.001; η^2 = 0.333). Whole-body R at IP was significantly lower than BL (p = 0.002; d = 0.12) and 24H (p = 0.021; d = 0.13).

Figure 8 displays R values of the dominant and nondominant RF. No significant sex × limb × time (F = 0.268; p = 0.766; η^2 = 0.016), sex × time (F = 0.322; p = 0.727; η^2 = 0.019), sex × limb (F < 0.001; p = 0.989; η^2 < 0.001), or limb × time (F = 0.996; p = 0.380; η^2 = 0.055) interactions were noted for R of the RF. While no significant main effect of limb was observed (F = 0.317; p = 0.581; η^2 = 0.018), a significant main effect of time was noted (F = 3.769; p = 0.050; η^2 = 0.181). However, further analysis revealed no significant difference between time points.

Figure 8: Changes in R of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex $\times$ limb $\times$ time ($F = 1.057$; $p = 0.358$; $\eta^2 = 0.055$), sex $\times$ time ($F = 2.106$; $p = 0.136$; $\eta^2 = 0.105$), sex $\times$ limb ($F = 0.265$; $p = 0.613$; $\eta^2 = 0.015$), or limb $\times$ time ($F = 1.292$; $p = 0.287$; $\eta^2 = 0.067$) interactions were noted for R of the VL. However, a significant main effect of time was noted ($F = 12.569$; $p < 0.001$; $\eta^2 = 0.411$; Figure 9). VL R was significantly reduced at IP compared to BL ($p < 0.001$; $d = 0.31$) and 24H ($p = 0.002$; $d = 0.31$).

Figure 9: Changes in R of the dominant and nondominant VL following unaccustomed resistance exercise

Reactance (Xc)

No significant sex $\times$ time interaction was noted for whole-body Xc ($F = 0.073$; $p = 0.930$; $\eta^2 = 0.004$), but a significant main effect of time was observed ($F = 24.635$; $p < 0.001$; $\eta^2 = 0.578$). Further analysis revealed a significantly lower whole-body Xc at IP compared to BL ($p < 0.001$; $d = 0.35$) and 24H ($p < 0.001$; $d = 0.22$).

Figure 10 displays Xc values of the dominant and nondominant RF. No significant sex $\times$ limb $\times$ time ($F = 0.338$; $p = 0.716$; $\eta^2 = 0.005$), sex $\times$ time ($F = 1.105$; $p = 0.343$; $\eta^2 = 0.061$), sex $\times$ limb ($F = 0.081$; $p = 0.780$; $\eta^2 = 0.015$), or limb $\times$ time ($F = 2.097$; $p = 0.138$; $\eta^2 = 0.110$) interactions were noted for Xc of the RF. However, significant main effects of time ($F = 7.909$; $p = 0.002$; $\eta^2 = 0.318$) and limb ($F = 11.666$; $p = 0.003$; $\eta^2 = 0.407$) were noted with the nondominant leg having lower Xc than the dominant leg ($p =$

0.003; $d = 0.28$), and IP being lower than BL ($p = 0.026$; $d = 0.46$) and 24H ($p = 0.010$, $d = 0.53$).

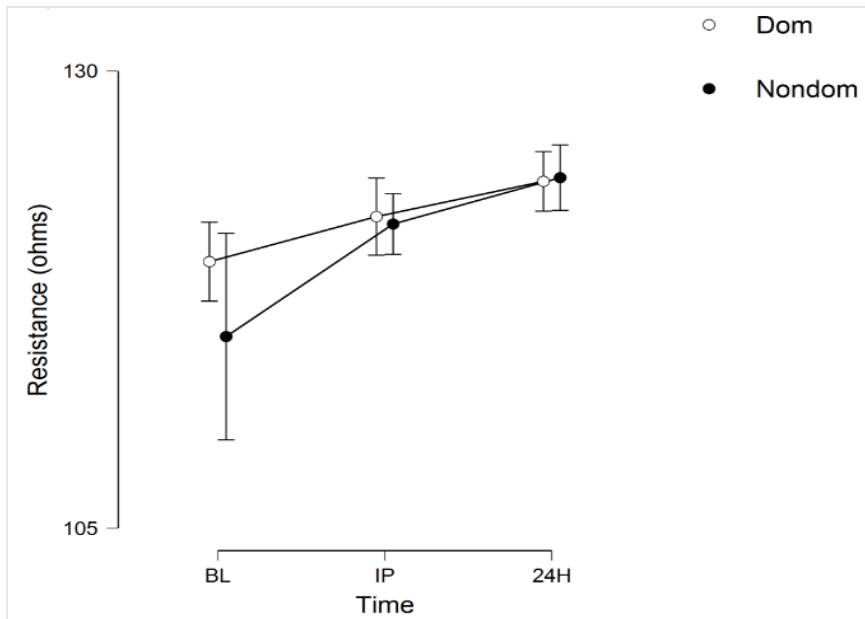Figure 10: Changes in Xc of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex × limb × time ($F = 1.152$; $p = 0.327$; $\eta^2 = 0.060$), sex × time ($F = 0.190$; $p = 0.828$; $\eta^2 = 0.010$), or sex × limb ($F = 0.126$; $p = 0.727$; $\eta^2 = 0.007$) interactions were observed for Xc of the VL. However, a significant limb × time interaction was noted ($F = 3.723$; $p = 0.034$; $\eta^2 = 0.171$; Figure 11). While no significant main effect of limb was observed ($F = 1.258$; $p = 0.277$; $\eta^2 = 0.065$), a significant main effect of time ($F = 43.104$; $p < 0.001$; $\eta^2 = 0.705$) was noted where VL Xc at IP was significantly lower than BL ($p < 0.001$; $d = 0.60$) and 24H ($p < 0.001$; $d = 0.72$).
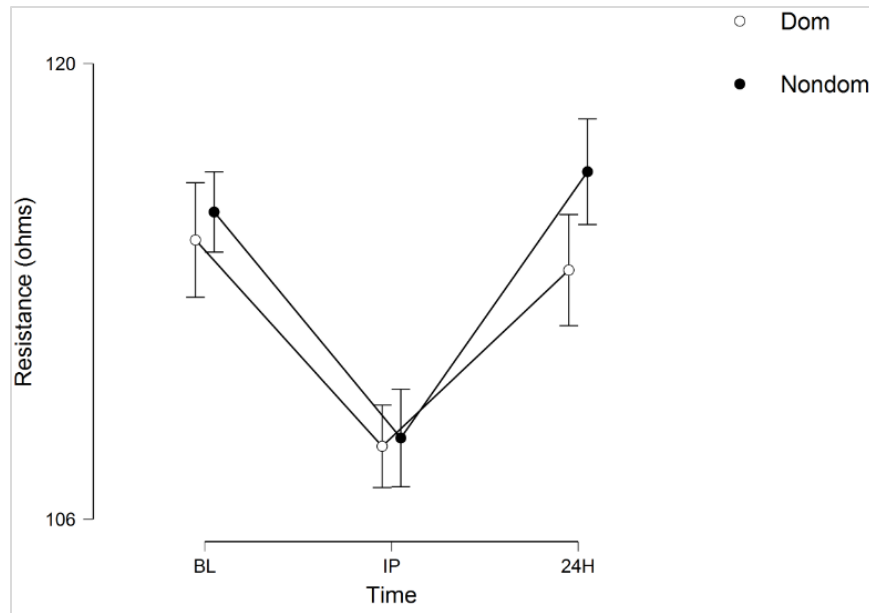
Figure 11: Changes in Xc of the dominant and nondominant VL following unaccustomed resistance exercise

Phase Angle (PhA)

No significant sex $\times$ time interaction was noted for whole-body PhA ($F = 0.078$; $p = 0.925$; $\eta^2 = 0.004$), but a significant main effect of time was observed ($F = 7.329$; $p = 0.002$; $\eta^2 = 0.289$). Further analysis revealed a significantly reduced PhA at IP compared to BL ($p = 0.004$; $d = 0.28$) and 24H ($p = 0.035$; $d = 0.23$).

Changes in PhA of the dominant and nondominant RF are displayed in Figure 12. No significant sex $\times$ limb $\times$ time ($F = 0.260$; $p = 0.772$; $\eta^2 = 0.015$), sex $\times$ time ($F = 0.150$; $p = 0.862$; $\eta^2 = 0.009$), sex $\times$ limb ($F = 1.145$; $p = 0.299$; $\eta^2 = 0.063$), or limb $\times$ time ($F = 2.247$; $p = 0.121$; $\eta^2 = 0.117$) interactions were noted for PhA of the RF. However, significant main effects of time ($F = 9.983$; $p < 0.001$; $\eta^2 = 0.370$) and limb ($F = 12.201$; $p = 0.003$; $\eta^2 = 0.418$) were noted with the nondominant leg having lower PhA than the

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dominant leg ($p = 0.003$; $d = 0.16$), and IP being lower than BL ($p = 0.006$; $d = 0.22$) and 24H ($p = 0.003$; $d = 0.19$).
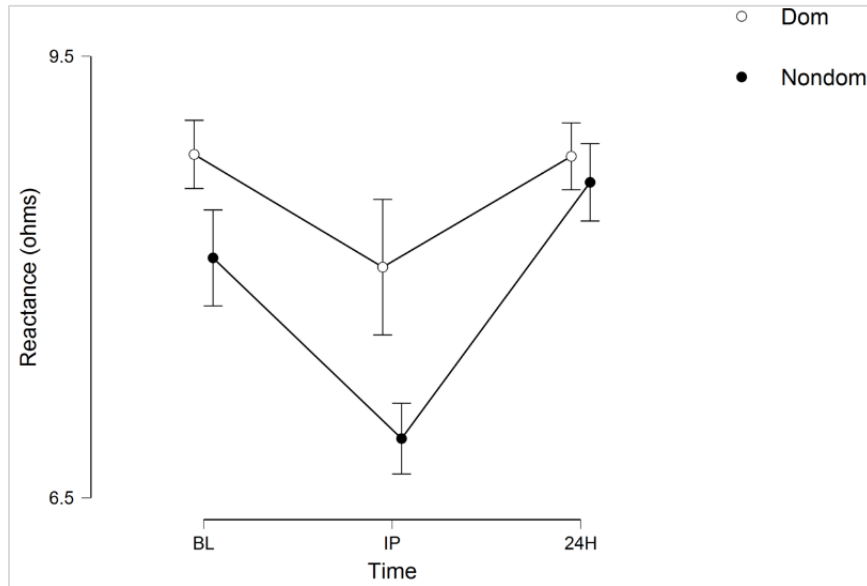

Figure 12: Changes in PhA of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex × limb × time ($F = 2.062$; $p = 0.142$; $\eta^2 = 0.103$), sex × time ($F = 1.773$ $p = 0.184$; $\eta^2 = 0.090$), or sex × limb ($F = 1.308$; $p = 0.268$; $\eta^2 = 0.068$) interactions were observed for PhA of the VL. However, a significant limb × time interaction was noted ($F = 23.676$; $p < 0.001$; $\eta^2 = 0.568$; Figure 13). Additionally, significant main effects of limb ($F = 85.184$; $p < 0.001$; $\eta^2 = 0.826$) and time ($F = 21.984$; $p < 0.001$; $\eta^2 = 0.550$) were noted with the nondominant leg having lower PhA than the dominant leg ($p < 0.001$; $d = 1.29$), and IP being lower than BL ($p < 0.001$; $d = 0.13$) and 24H ($p < 0.001$; $d = 0.15$). While no significant changes were noted in the dominant leg, PhA of the

nondominant VL was significantly lower at IP compared to BL ($p < 0.001$; $d = 0.25$) and

24H ($p < 0.001$; $d = 0.27$).
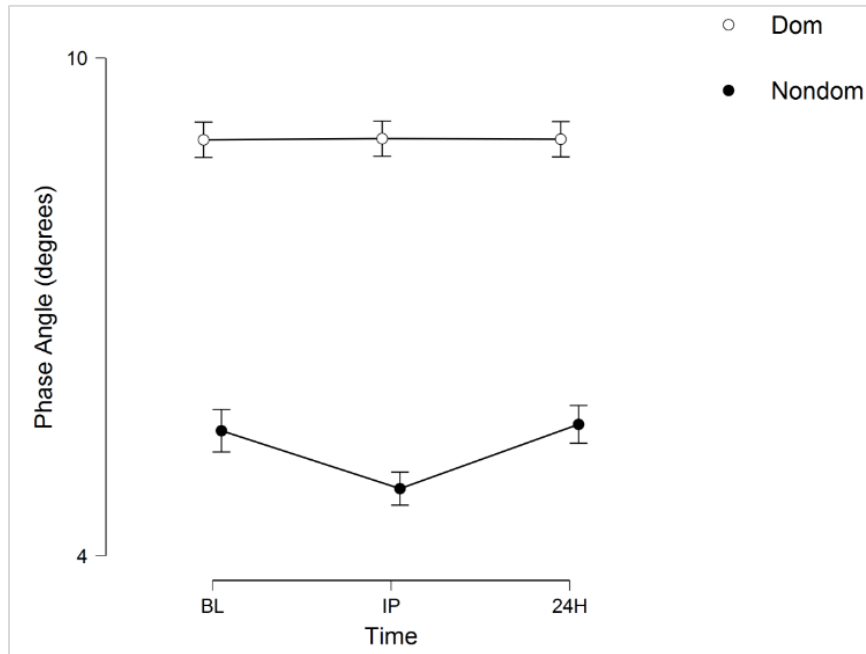
Figure 13: Changes in PhA of the dominant and nondominant VL following
unaccustomed resistance exercise

Tensiomyography

Maximal Radial Displacement (Dm)

No significant sex × limb × time ($F = 1.610$; $p = 0.213$; $\eta^2 = 0.078$), sex × time ($F =$

1.999; $p = 0.149$; $\eta^2 = 0.095$), sex × limb ($F = 1.110$; $p = 0.305$; $\eta^2 = 0.055$), or limb ×

time ($F = 0.910$; $p = 0.411$; $\eta^2 = 0.046$) interactions were observed for Dm of the RF.

While no significant main effect of limb was observed ($F = 0.019$; $p = 0.893$; $\eta^2 = 0.001$),

a significant main effect of time ($F = 6.683$; $p = 0.003$; $\eta^2 = 0.260$) was noted where RF

Dm at IP was significantly lower than BL ($p = 0.005$; $d = 0.41$) but not 24H ($p = 0.141$; d

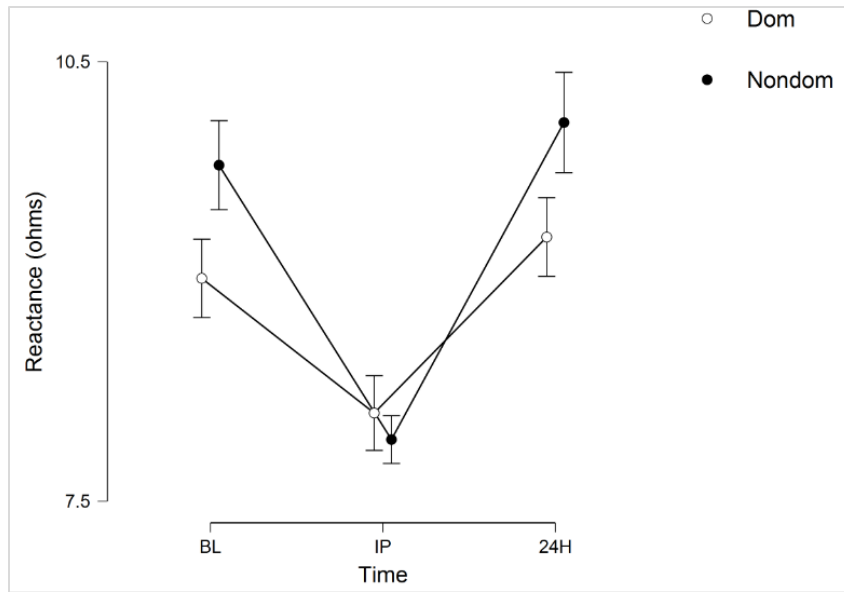$= 0.28$). Changes in Dm of the dominant and nondominant RF are displayed in Figure 14.
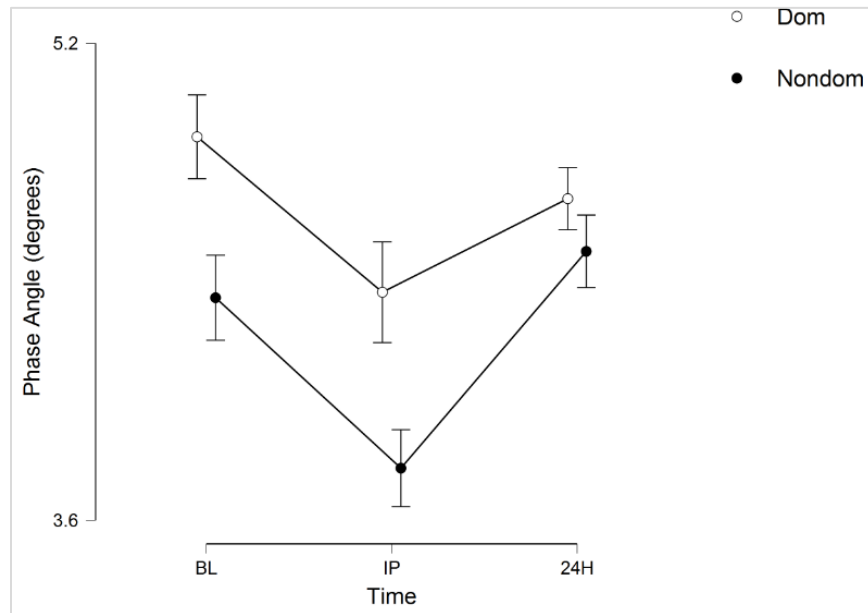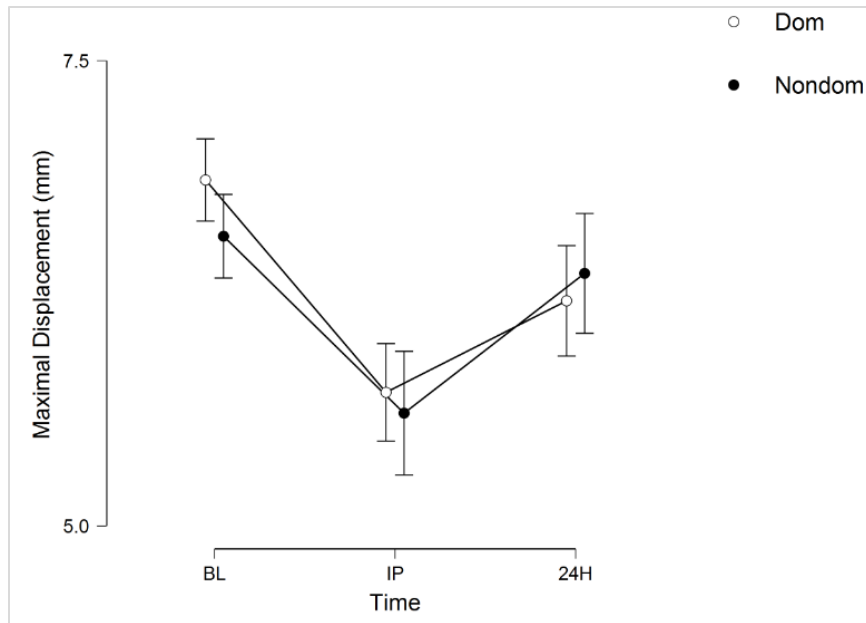
Figure 14: Changes in Dm of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex $\times$ limb $\times$ time ($F = 2.209$; $p = 0.124$; $\eta^2 = 0.104$), sex $\times$ time ($F = 1.941$; $p = 0.157$; $\eta^2 = 0.093$), sex $\times$ limb ($F = 2.552$; $p = 0.127$; $\eta^2 = 0.118$), or limb $\times$ time ($F = 1.583$; $p = 0.219$; $\eta^2 = 0.077$) interactions were observed for Dm of the VL. While no significant main effect of time was observed ($F = 0.971$; $p = 0.388$; $\eta^2 = 0.049$), a significant main effect of limb ($F = 5.165$; $p = 0.035$; $\eta^2 = 0.214$) was noted with the nondominant leg having lower Dm than the dominant leg ($p = 0.035$; $d = 0.41$). Changes in Dm of the dominant and nondominant VL are displayed in Figure 15.

Figure 15: Changes in Dm of the dominant and nondominant VL following
unaccustomed resistance exercise

Delay Time (Td)

No significant sex × limb × time ($F = 1.386$; $p = 0.263$; $\eta^2 = 0.068$), sex × time ($F = 1.251$; $p = 0.298$; $\eta^2 = 0.062$), sex × limb ($F = 0.032$; $p = 0.859$; $\eta^2 = 0.002$), or limb × time ($F = 0.438$; $p = 0.648$; $\eta^2 = 0.023$) interactions were observed for Td of the RF. While no significant main effect of limb was observed ($F = 0.265$; $p = 0.613$; $\eta^2 = 0.014$), a significant main effect of time ($F = 6.192$; $p = 0.013$; $\eta^2 = 0.246$) was noted where RF Td at IP was significantly lower than BL ($p = 0.022$; $d = 0.35$) but not 24H ($p = 0.059$; $d = 0.42$). Changes in Td of the dominant and nondominant RF are displayed in Figure 16.

Figure 16: Changes in Td of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex × limb × time ($F = 1.834$; $p = 0.174$; $\eta^2 = 0.092$), sex × time ($F = 1.793$; $p = 0.181$; $\eta^2 = 0.091$), or sex × limb ($F = 0.002$; $p = 0.966$; $\eta^2 < 0.001$) interactions were observed for Td of the VL. However, a significant limb × time interaction was noted ($F = 6.633$; $p = 0.004$; $\eta^2 = 0.269$; Figure 17). Additionally, significant main effects of limb ($F = 4.796$; $p = 0.042$; $\eta^2 = 0.210$) and time ($F = 21.629$; $p < 0.001$; $\eta^2 = 0.546$) were noted with the nondominant leg having lower Td than the dominant leg ($p = 0.042$; $d = 0.43$), and IP being lower than BL ($p = 0.001$; $d = 0.40$) and 24H ($p < 0.001$; $d = 0.54$). Td of the nondominant VL was significantly lower at IP compared to BL ($p < 0.001$; $d = 0.53$) and 24H ($p < 0.001$; $d = 0.59$), while Td of the dominant VL was significantly higher at 24H compared to IP ($p = 0.017$; $d = 0.38$) but not BL ($p = 0.101$; $d = 0.30$). Compared to the dominant VL, Td of the nondominant VL

was significantly lower at IP ($p = 0.001$; $d = 0.75$) but not BL ($p = 0.502$; $d = 0.16$) or

24H ($p = 0.066$; $d = 0.36$).
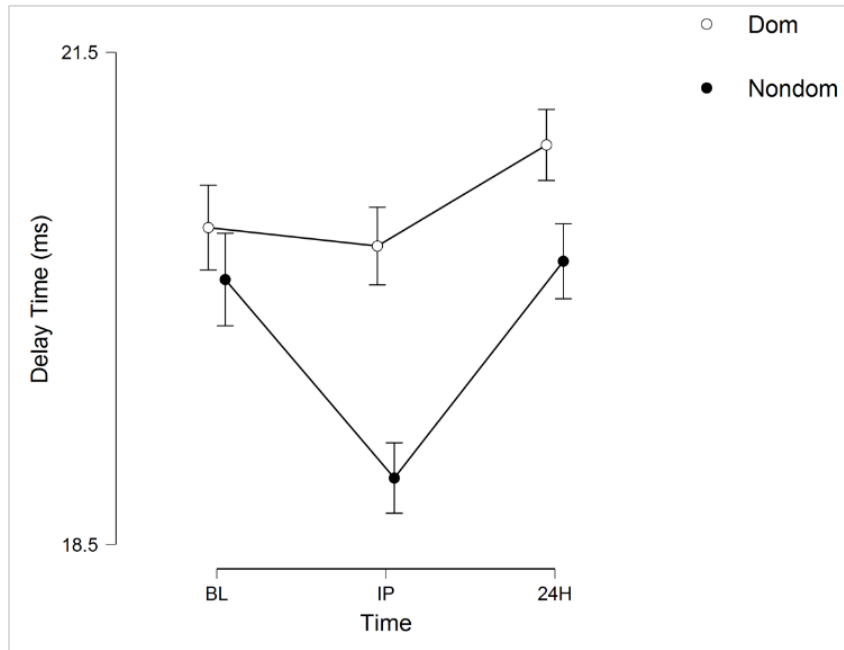
Figure 17: Changes in Td of the dominant and nondominant VL following unaccustomed
resistance exercise

Contraction Time (Tc)

No significant sex × limb × time ($F = 0.005$; $p = 0.995$; $\eta^2 < 0.001$), sex × time ($F =$

1.602; $p = 0.215$; $\eta^2 = 0.078$), sex × limb ($F = 0.224$; $p = 0.642$; $\eta^2 = 0.012$), or limb ×

time ($F = 0.418$; $p = 0.661$; $\eta^2 = 0.022$) interactions were observed for Tc of the RF.

Similarly, no significant main effects of limb ($F = 0.257$; $p = 0.618$; $\eta^2 = 0.013$) or time

($F = 0.998$; $p = 0.359$; $\eta^2 = 0.050$) were noted.

No significant sex × limb × time ($F = 2.573$; $p = 0.090$; $\eta^2 = 0.119$), sex × time ($F =$

0.765; $p = 0.473$; $\eta^2 = 0.039$), sex × limb ($F = 0.278$; $p = 0.604$; $\eta^2 = 0.014$), or limb ×

time ($F = 0.786$; $p = 0.463$; $\eta^2 = 0.040$) interactions were observed for Tc of the VL.

Similarly, no significant main effects of limb ($F = 1.398$; $p = 0.252$; $\eta^2 = 0.069$) or time ($F = 2.034$; $p = 0.145$; $\eta^2 = 0.097$) were noted.

Contraction Velocity (Vc)

Changes in Vc of the dominant and nondominant RF are displayed in Figure 18. No significant sex × limb × time ($F = 2.187$; $p = 0.126$; $\eta^2 = 0.103$), sex × time ($F = 0.698$; $p = 0.504$; $\eta^2 = 0.035$), sex × limb ($F = 2.440$; $p = 0.135$; $\eta^2 = 0.114$), or limb × time ($F = 0.825$; $p = 0.446$; $\eta^2 = 0.042$) interactions were observed for Vc of the RF. While no significant main effect of limb was observed ($F = 0.216$; $p = 0.648$; $\eta^2 = 0.011$), a significant main effect of time ($F = 7.155$; $p = 0.002$; $\eta^2 = 0.274$) was noted where RF Vc at IP was significantly lower than BL ($p = 0.004$; $d = 0.40$) but not 24H ($p = 0.174$; $d = 0.23$).
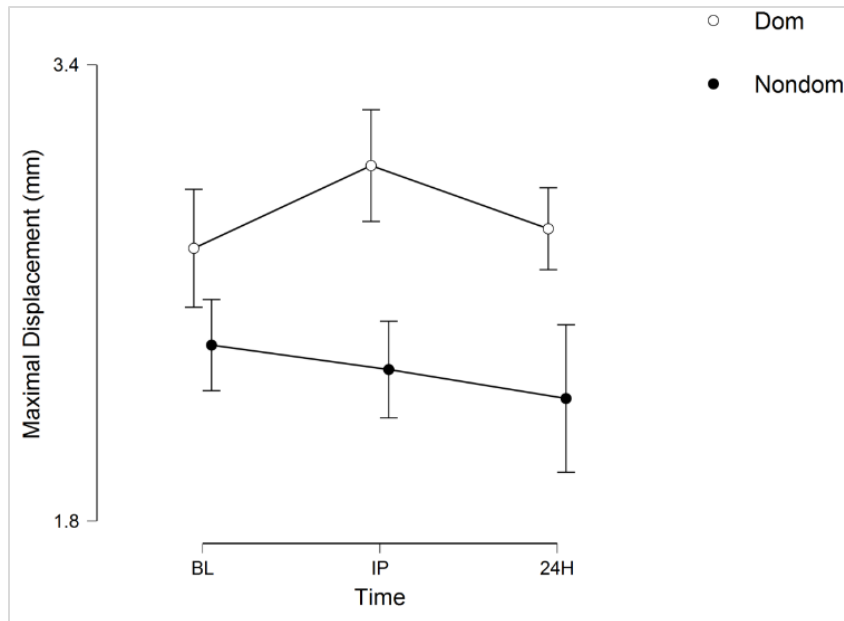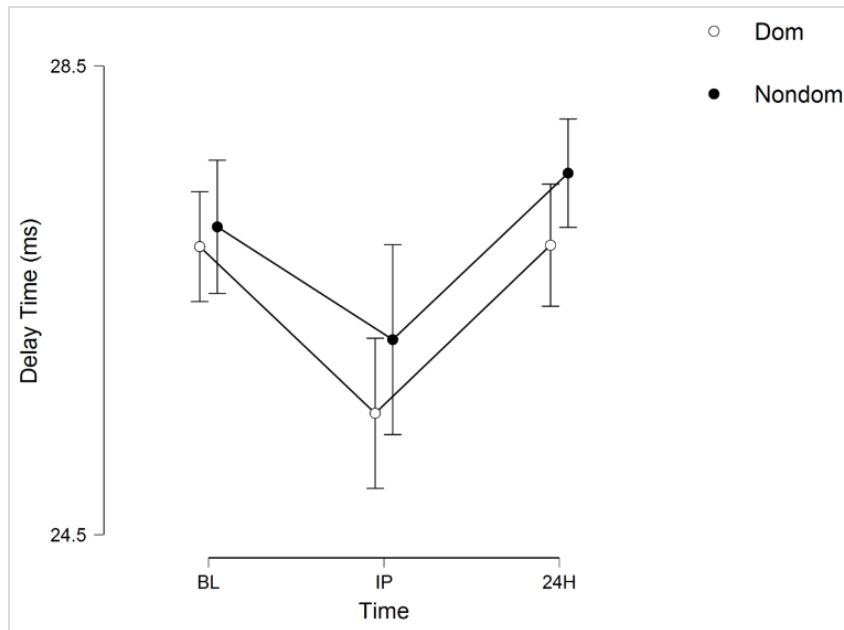

Figure 18: Changes in Vc of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex $\times$ limb $\times$ time ($F = 1.846$; $p = 0.172$; $\eta^2 = 0.089$), sex $\times$ time ($F = 2.207$; $p = 0.124$; $\eta^2 = 0.104$), sex $\times$ limb ($F = 2.303$; $p = 0.146$; $\eta^2 = 0.108$), or limb $\times$ time ($F = 1.846$; $p = 0.172$; $\eta^2 = 0.089$) interactions were observed for Vc of the VL. While no significant main effect of time was observed ($F = 1.397$; $p = 0.260$; $\eta^2 = 0.068$), a significant main effect of limb ($F = 6.716$; $p = 0.018$; $\eta^2 = 0.261$) was noted with the nondominant leg having lower Vc than the dominant leg ($p = 0.006$; $d = 0.42$). Changes in Vc of the dominant and nondominant VL are displayed in Figure 19.
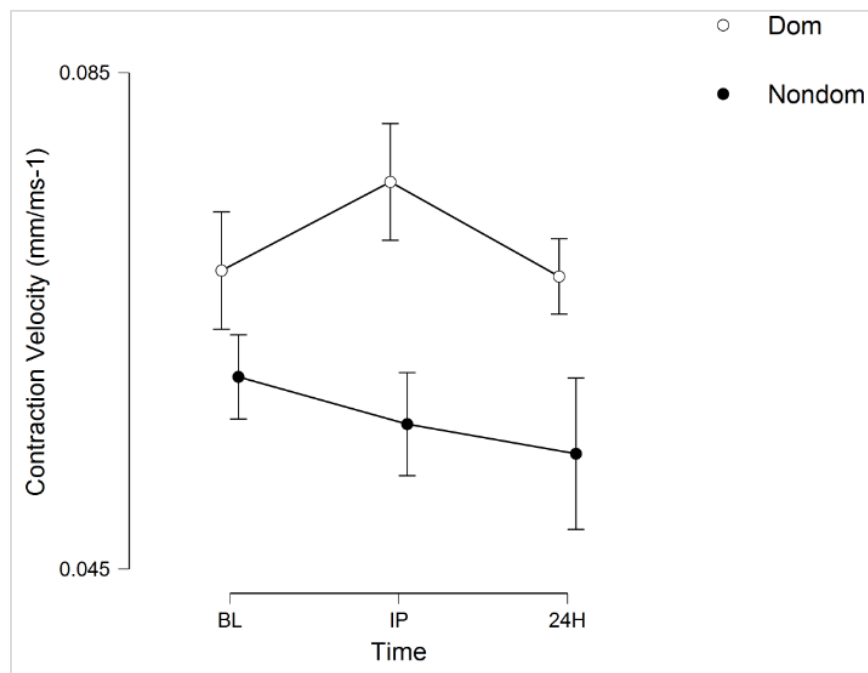
Figure 19: Changes in Vc of the dominant and nondominant VL following unaccustomed resistance exercise

<u>Sonomyography</u>

Echo Intensity (EI)

No significant sex $\times$ limb $\times$ time ($F = 0.436$; $p = 0.650$; $\eta^2 = 0.024$), sex $\times$ time ($F = 0.362$; $p = 0.699$; $\eta^2 = 0.020$), or sex $\times$ limb ($F = 1.614$; $p = 0.220$; $\eta^2 = 0.082$)

interactions were observed for raw EI of the RF. However, a significant limb $\times$ time interaction was noted ($F = 5.943$; $p = 0.006$; $\eta^2 = 0.248$). While no main effect of limb was observed ($F = 2.751$; $p = 0.115$; $\eta^2 = 0.133$), a significant main effect of time ($F = 8.406$; $p = 0.005$; $\eta^2 = 0.318$) was noted where RF raw EI at IP was significantly higher than BL ($p < 0.001$; $d = 0.44$) and 24H ($p = 0.014$; $d = 0.59$). Raw EI of the nondominant RF was significantly higher at IP compared to BL ($p < 0.001$; $d = 0.64$) and 24H ($p = 0.003$; $d = 0.83$), while EI of the dominant RF was significantly higher at IP compared to BL ($p = 0.035$; $d = 0.20$) but not 24H ($p = 0.200$; $d = 0.32$). Compared to the dominant RF, raw EI of the nondominant RF was significantly higher at IP ($p = 0.022$; $d = 0.45$) but not BL ($p = 0.488$; $d = 0.10$) or 24H ($p = 0.696$; $d = 0.05$).

After correcting for SFT, no significant sex $\times$ limb $\times$ time ($F = 0.744$; $p = 0.482$; $\eta^2 = 0.040$), sex $\times$ time ($F = 0.086$; $p = 0.918$; $\eta^2 = 0.005$), or sex $\times$ limb ($F = 1.408$; $p = 0.251$; $\eta^2 = 0.073$) interactions were observed for EI of the RF. However, a significant limb $\times$ time interaction was noted ($F = 5.943$; $p = 0.006$; $\eta^2 = 0.248$). While no main effect of limb was observed ($F = 2.647$; $p = 0.121$; $\eta^2 = 0.128$), a significant main effect of time ($F = 8.332$; $p = 0.005$; $\eta^2 = 0.316$) was noted where RF EI at IP was significantly higher than BL ($p < 0.001$; $d = 0.23$) and 24H ($p = 0.013$; $d = 0.32$). Corrected EI of the nondominant RF was significantly higher at IP compared BL ($p < 0.001$; $d = 0.33$) and 24H ($p = 0.002$; $d = 0.44$), while corrected EI of the dominant RF was significantly higher at IP compared to BL ($p = 0.043$; $d = 0.11$) but not 24H ($p = 0.043$; $d = 0.18$). Compared to the dominant RF, corrected EI of the nondominant RF was significantly higher at IP ($p = 0.020$; $d = 0.27$) but not BL ($p = 0.423$; $d = 0.07$) or 24H ($p = 0.853$; $d =$

0.02). Changes in corrected EI of the dominant and nondominant RF are displayed in
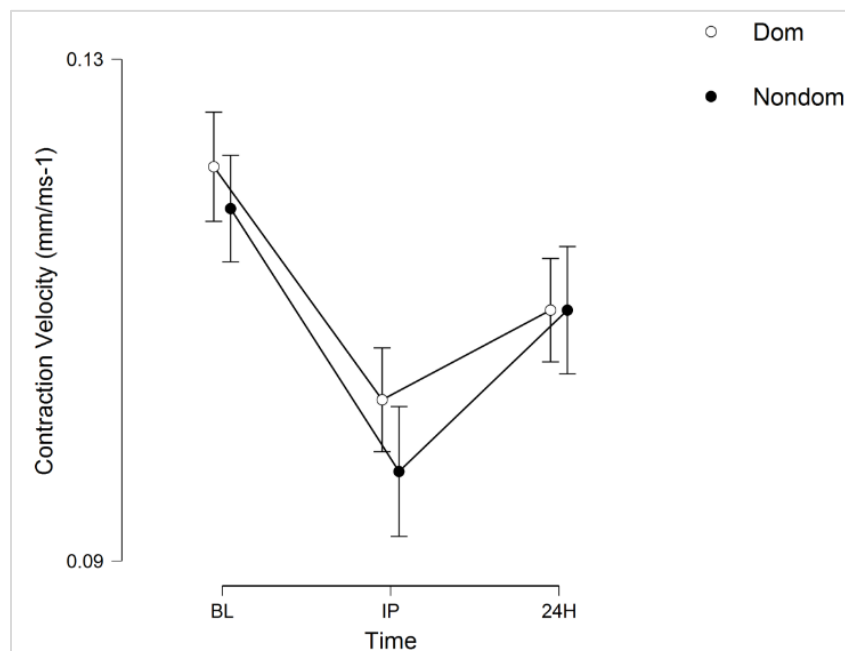
Figure 20.
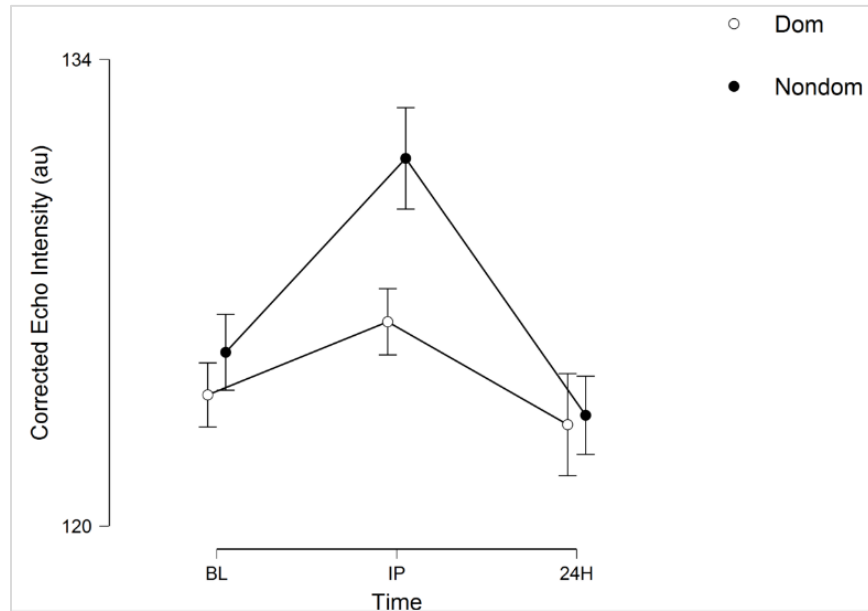
Figure 20: Changes in corrected EI of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex $\times$ limb $\times$ time ($F = 1.569$; $p = 0.222$; $\eta^2 = 0.080$), sex $\times$ time ($F = 0.455$; $p = 0.638$; $\eta^2 = 0.025$), sex $\times$ limb ($F = 2.246$; $p = 0.151$; $\eta^2 = 0.151$), or limb $\times$ time ($F = 0.180$; $p = 0.836$; $\eta^2 = 0.010$) interactions were observed for raw EI of the VL. However, significant main effects of limb ($F = 4.921$; $p = 0.040$; $\eta^2 = 0.215$) and time ($F = 9.545$; $p = 0.002$; $\eta^2 = 0.347$) were noted with the nondominant leg having lower raw EI than the dominant leg ($p = 0.040$; $d = 0.36$), and IP being higher than BL ($p < 0.001$; $d = 0.72$) and 24H ($p = 0.005$; $d = 0.63$).

After correcting for SFT, no significant sex $\times$ limb $\times$ time ($F = 1.220$; $p = 0.307$; $\eta^2 = 0.063$), sex $\times$ time ($F = 0.465$; $p = 0.632$; $\eta^2 = 0.025$), sex $\times$ limb ($F = 2.156$; $p = 0.159$;

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$\eta^2 = 0.107$), or limb × time ($F = 0.081$; $p = 0.922$; $\eta^2 = 0.004$) interactions were observed for EI of the VL. While no main effect of limb was observed ($F = 3.278$; $p = 0.087$; $\eta^2 = 0.154$), a significant main effect of time ($F = 9.505$; $p = 0.002$; $\eta^2 = 0.346$) was noted where VL EI at IP was significantly higher than BL ($p < 0.001$; $d = 0.28$) and 24H ($p = 0.006$; $d = 0.30$). Changes in corrected EI of the dominant and nondominant VL are displayed in Figure 21.
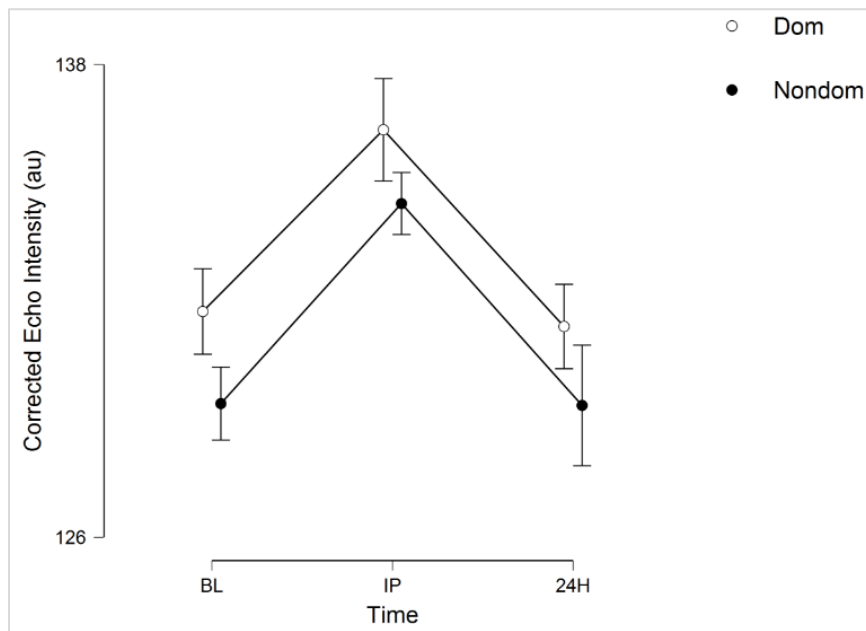
Figure 21: Changes in corrected EI of the dominant and nondominant VL following unaccustomed resistance exercise

Cross-Sectional Area (CSA)

No significant sex × limb × time ($F = 0.863$; $p = 0.431$; $\eta^2 = 0.046$), sex × time ($F = 2.661$; $p = 0.084$; $\eta^2 = 0.129$), or limb × time ($F = 2.532$; $p = 0.094$; $\eta^2 = 0.123$) interactions were observed for CSA of the RF. However, a significant sex × limb ($F = 10.691$; $p = 0.004$; $\eta^2 = 0.373$) interaction was noted. While no main effect of limb was

observed ($F = 0.505$; $p = 0.487$; $\eta^2 = 0.027$), a significant main effect of time ($F = 28.643$; $p < 0.001$; $\eta^2 = 0.614$) was noted where RF CSA at IP was significantly higher than BL ($p < 0.001$; $d = 0.27$) and 24H ($p < 0.001$; $d = 0.13$). For males, nondominant RF CSA was significantly lower than dominant RF CSA at BL ($p = 0.045$; $d = 0.81$) but not IP ($p = 0.263$; $d = 0.34$) or 24H ($p = 0.117$; $d = 0.44$). For females, nondominant RF CSA was significantly greater than dominant RF CSA at IP ($p = 0.016$; $d = 0.25$) but not BL ($p = 0.061$; $d = 0.18$) or 24H ($p = 0.055$; $d = 0.18$). However, no significant between-sex differences were noted at any time point. Changes in RF CSA of the dominant and nondominant legs are displayed in Figure 22.
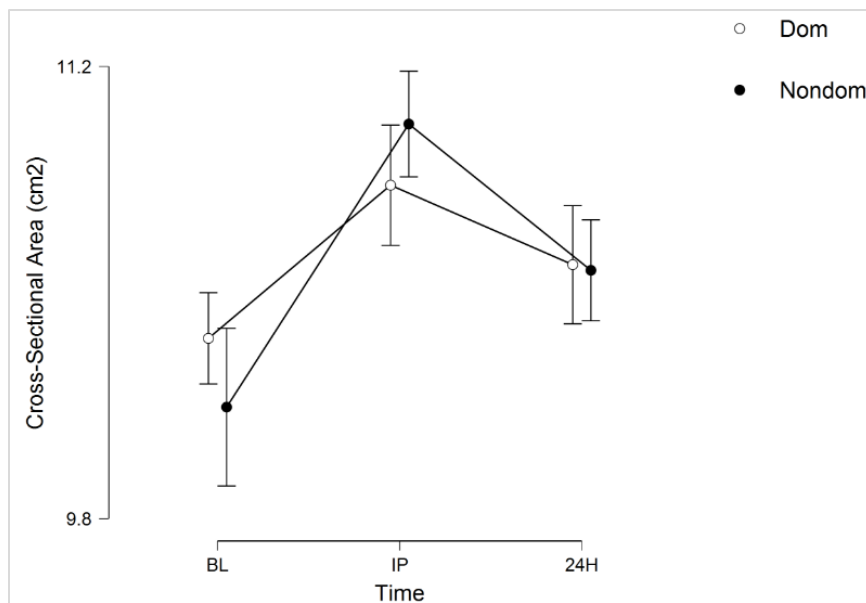
Figure 22: Changes in CSA of the dominant and nondominant RF following unaccustomed resistance exercise

No significant sex $\times$ limb $\times$ time ($F = 1.011$; $p = 0.374$; $\eta^2 = 0.053$), sex $\times$ time ($F = 2.949$; $p = 0.065$; $\eta^2 = 0.141$), sex $\times$ limb ($F = 1.145$; $p = 0.299$; $\eta^2 = 0.060$), or limb $\times$ time ($F = 0.438$; $p = 0.649$; $\eta^2 = 0.024$) interactions were observed for CSA of the VL.

While no main effect of limb was observed ($F = 2.364$; $p = 0.142$; $\eta^2 = 0.116$), a significant main effect of time ($F = 6.187$; $p = 0.005$; $\eta^2 = 0.256$) was noted where VL CSA at IP was significantly higher than BL ($p = 0.019$; $d = 0.12$) and 24H ($p = 0.020$; $d = 0.09$). Changes in VL CSA of the dominant and nondominant legs are displayed in Figure 23.
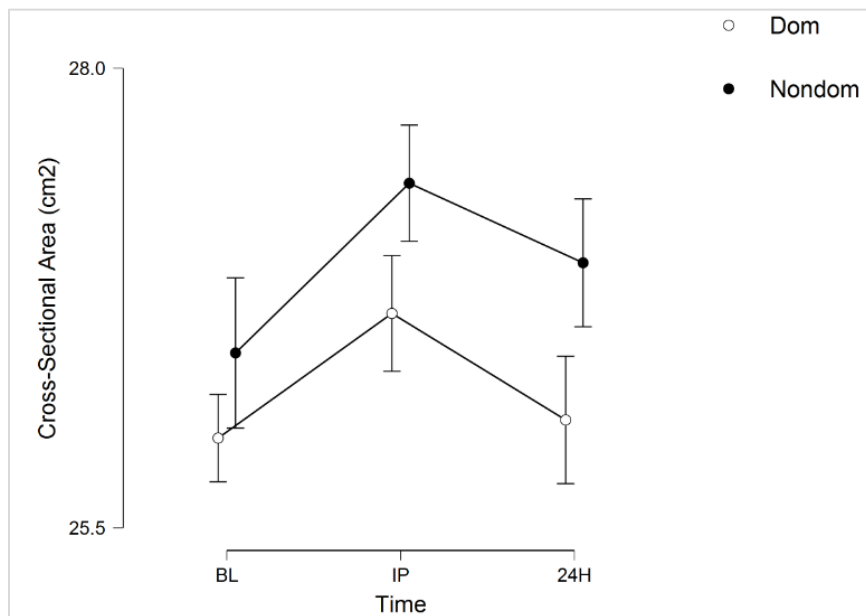
Figure 23: Changes in CSA of the dominant and nondominant VL following unaccustomed resistance exercise

Muscle Thickness (MT)

No significant sex × limb × time ($F = 0.487$; $p = 0.618$; $\eta^2 = 0.026$), sex × time ($F = 0.143$; $p = 0.867$; $\eta^2 = 0.008$), sex × limb ($F = 0.321$; $p = 0.578$; $\eta^2 = 0.017$), or limb × time ($F = 0.714$; $p = 0.496$; $\eta^2 = 0.038$) interactions were observed for MT of the RF. While no main effect of limb was observed ($F = 1.671$; $p = 0.212$; $\eta^2 = 0.085$), a significant main effect of time ($F = 4.742$; $p = 0.015$; $\eta^2 = 0.209$) was noted where RF

MT at IP was significantly higher than BL ($p = 0.012$; $d = 0.14$) but not 24H ($p = 0.170$; $d = 0.09$).

No significant sex $\times$ limb $\times$ time ($F = 0.775$; $p = 0.468$; $\eta^2 = 0.041$), sex $\times$ time ($F = 2.735$; $p = 0.078$; $\eta^2 = 0.132$), sex $\times$ limb ($F = 1.229$; $p = 0.282$; $\eta^2 = 0.064$), or limb $\times$ time ($F = 1.614$; $p = 0.213$; $\eta^2 = 0.082$) interactions were observed for MT of the VL. While no main effect of limb was observed ($F = 0.686$; $p = 0.419$; $\eta^2 = 0.037$), a significant main effect of time ($F = 4.488$; $p = 0.018$; $\eta^2 = 0.200$) was noted where VL MT at IP was significantly higher than BL ($p = 0.029$; $d = 0.17$) but not 24H ($p = 0.380$; $d = 0.10$). Changes in RF and VL MT for the dominant and nondominant legs are displayed in Figures 24 and 25, respectively.
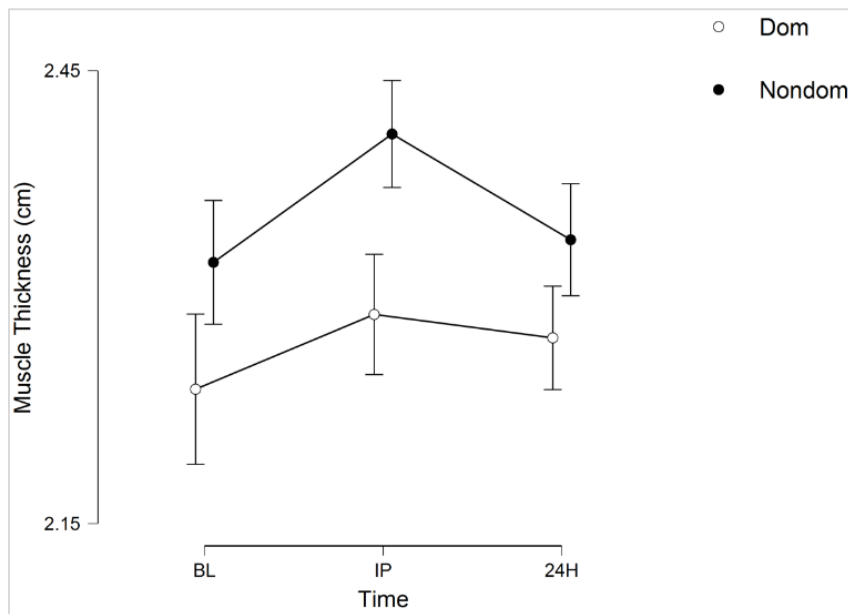
Figure 24: Changes in MT of the dominant and nondominant RF following unaccustomed resistance exercise
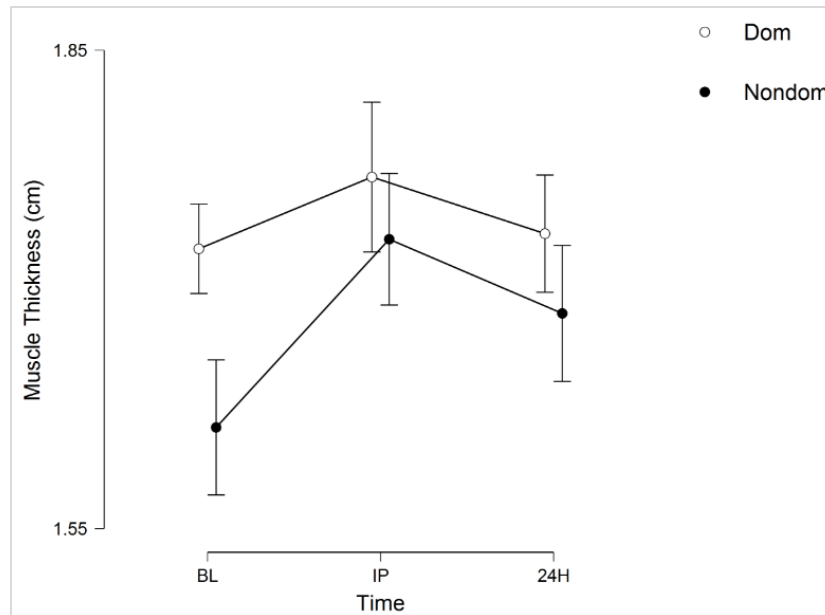
Figure 25: Changes in MT of the dominant and nondominant VL following unaccustomed resistance exercise

Correlational Analyses

Tables 4 and 5 list significant correlations identified among changes from BL to IP in strength and myographical parameters of the nondominant RF and VL, respectively.

Table 4: Significant correlations identified in the nondominant RF

		Pearson's *r*	*p*-value
% Δ BL – IP			
CSA	PTQ	0.462	0.040
CSA	R	0.494	0.031
CSA	PhA	0.484	0.036
PTQ	R	0.635	0.003
PTQ	PhA	0.596	0.007

Table 5: Significant correlations identified in the nondominant VL

		Pearson's *r*	***p*-value**
% Δ BL – IP			
Td	R	0.543	0.020
Td	Xc	0.518	0.028
Td	MT	0.496	0.031
Dm	R	0.466	0.044
Vc	RTD300	0.466	0.033
Vc	PTQ	0.652	0.001
Vc	PhA	0.466	0.038
Xc	CSA	-0.506	0.027
PhA	CSA	-0.543	0.013

CHAPTER FIVE: DISCUSSION

Results of this study demonstrate that a single bout of unilateral eccentric exercise temporarily alters various properties of skeletal muscle (i.e., echogenicity, morphology, cell membrane integrity, and contractility). Contrary to the initial hypotheses, changes in the myographical assessments observed occurred despite evidence of specific functional deficits in isometric strength and rate of torque development following unaccustomed resistance exercise. Additionally, no sex-based differences in the myographical response to the selected exercise regimen were identified as was anticipated.

Changes in Performance

Peak torque and rate of torque development were significantly lower in the nondominant (exercised) limb compared to the dominant (non-exercised) limb at all time points throughout this study. This finding is in agreement with previous research identifying greater isokinetic torque production of the dominant versus nondominant knee extensors (Wyatt & Edwards, 1981). However, it is unclear why the eccentric exercise protocol employed in this study did not impair peak torque or rate of torque development as this protocol has previously been shown to elicit significant reductions (~28%) in isometric peak torque among untrained adults (Byrne et al., 2001). In the current study, peak torque of the exercised leg was reduced by 4.9 ± 17.1% while rate of torque development decreased by 5.4 ± 12.9% immediately after exercise. It should be noted that Byrne and colleagues had participants perform exercise in the prone position through a range of motion of approximately 100°, while the exercise protocol employed in the current study was carried out in the supine position with the knee extending through a range of motion

of approximately 90° which may be the cause of these discrepant findings. Although the exercise protocol used in the current study was selected with the aim of replicating muscle damage as previously reported (Byrne et al., 2001), the lack of apparent strength deficits suggests that participants in this study did not experience the anticipated degree of muscle insult. It should be noted that all participants completed the exercise protocol in its entirety, therefore it is speculated that these unexpected outcomes are a function of submaximal and/or inconsistent effort during exercise and/or the MVIC assessments. Additionally, it is possible that participants tempered their exertion throughout the protocol to avoid unwanted symptoms of exercise-induced muscle damage (e.g., soreness) or acute discomfort. Lastly, given the high degree of variability in strength changes during in response to the exercise protocol (see Figures 1-7), it is plausible that participants were simply unaccustomed to the demands of such a novel stimulus. Nevertheless, the performance of 100 submaximal repetitions likely caused temporary metabolic perturbations rather than explicit tissue damage which might explain the detection of compromised strength. Consequences of metabolic fatigue such as substrate depletion (e.g., glycogen) and by-product accumulation (e.g., H^+ ions) have been shown to affect muscle contractility (Allen, Lannergren, & Westerblad, 1995; Ørtenblad, Westerblad, & Nielsen, 2013) which may partially explain the current findings with respect to the myographical measures.

Traditionally, resistance exercise is understood to promote fatigue, with the general assumption that its effects are limited to the local musculature recruited during contraction. Interestingly, results of the current study indicated that both legs experienced

comparable changes in peak torque (dominant: -6.6 ± 12.2%; nondominant: -4.9 ± 17.1%) and rate of torque development (dominant: -5.6 ± 16.4%; nondominant: -5.4 ± 12.9%) suggesting a similar level of fatigue between limbs. This phenomenon of temporary performance decrements in non-exercised muscles following a fatiguing exercise protocol has been reported elsewhere and is believed to occur through centrally-mediated mechanisms (Rattey, Martin, Kay, Cannon, & Marino, 2006; Ye, Beck, Wages, & Carr, 2018). In light of this, it is reasonable to assume the cross-over effects may have contributed to the other myographical changes observed in the non-exercised leg.

Contralateral Contribution

Bilateral reductions in myographical assessments were observed for a number of measures recorded during this study. Although the eccentric exercise protocol was performed with the nondominant knee extensors, properties of the dominant knee extensors displayed similar disturbances. This interlimb interaction is believed to be the result of concurrent contralateral activation from isometric muscle actions during exercise and has been shown to occur especially in lower extremities (Kang, Na, Moon, Chun, & Yoon, 1997). In support of this, bilateral reductions in VL resistance and reactance were observed immediately after exercise. With the understanding that changes in resistance and reactance are affected independent of the level of exercise intensity (Fu & Freeborn, 2018), these results suggest that bioelectrical impedance properties of the knee extensors are equally affected following isometric (i.e., contralateral stabilization) and eccentric contractions. Additionally, both legs displayed similar changes in cross-sectional area of the RF and VL, as well as echo intensity and thickness of the VL in response to the

unaccustomed resistance training protocol. Acute changes in muscle size are commonly reported following resistance exercise (Jajtner et al., 2015; Radaelli et al., 2014) and are related to swelling and fluid accumulation (Nosaka & Clarkson, 1996; Radaelli et al., 2014). Muscle echogenicity has also been shown to increase immediately after resistance exercise (Jajtner et al., 2015). As with the changes in BIA parameters, the comparable bilateral changes observed in contractile properties (i.e., Dm, Td, and Vc) of the RF suggest similar effects of eccentric and isometric muscle actions.

<u>Changes in Bioelectrical Properties</u>

The exercise protocol employed in this study caused acute reductions in whole-body resistance, reactance, and phase angle. With consideration for the amount of muscle mass involved during exercise and contralateral stabilization, and an understanding of the interrelationship between phase angle, resistance, and reactance, it is plausible that the abovementioned changes in whole-body BIA parameters were indicative of the systemic exercise response and collectively influenced by the localized reductions in resistance in the VL, reactance in the VL and RF, and phase angle in the RF observed in both legs. The reduced reactance – representative of compromised cell membrane integrity (Nescolarde et al., 2013) – was accompanied by a decrease in VL phase angle of the exercised leg only with no change in the non-exercised VL. This suggests a greater involvement of the VL than the RF during the eccentric protocol focused on the knee extensors and may be attributed to its monoarticular nature. Given the hypothesized contralateral stabilization performed with the non-exercised leg, it is likely that this isometric knee flexion resulted in co-activation of the RF as a biarticular stabilizing

muscle. Conversely, the dynamic nature of the work performed on the exercised leg and attempts to isolate movement about the knee likely warranted greater involvement of the VL which is evidenced by the localized ipsilateral perturbation (i.e., reduced phase angle) of the exercised VL with no such change in the non-exercised VL.

Local resistance and reactance, as assessed by EIM, have been shown to decrease following isometric (Li et al., 2016), isotonic (Fu & Freeborn, 2018), and eccentric contractions (Sanchez et al., 2017). Furthermore, these changes are associated with the accumulation of muscle fatigue after task failure (Fu & Freeborn, 2018; Li et al., 2016) which supports the current findings of altered BIA parameters following serial contractions. However, acute changes in phase angle have yet to be explored. Though interlimb differences in bioelectrical impedance properties are undefined in the literature, it seems reasonable that the dominant musculature would display a greater phase angle based on the premise that it is improved through chronic stimulation (e.g., exercise) (Sardinha, 2018; Tomeleri et al., 2018). Reductions in local reactance and phase angle tend to occur after injury, fatigue, and exercise-induced muscle damage and are purportedly linked to disruption of the muscle structure (Fu & Freeborn, 2018; Nescolarde et al., 2015). Furthermore, these changes are believed to be a function of the disruptive nature of repeated contractions on the cell membrane and appear to decrease proportionally with respect to the severity of tissue disruption that takes place in response to insult (Fu & Freeborn, 2018; Gibala et al., 1995; Nescolarde et al., 2015, 2017). The current results suggest that these changes occur before or potentially without any detectable deficits in strength. Because the transmission of mechanical force is dependent

upon the cell membrane integrity, changes in bioelectrical properties likely precede alterations in muscle contractility. In support of this, the change in peak isometric torque of the exercised leg from BL to IP was correlated with the changes in RF reactance and phase angle of the exercised leg suggesting RF contribution during isometric force production. However, to further support the theorized preferential contribution of the VL during eccentric exercise, significant correlations were identified between the change in maximal displacement from BL to IP and VL resistance of the exercised leg, the change in delay time from BL to IP and the changes in VL resistance and reactance of the exercised leg, as well as between the change in contraction velocity of the exercised VL from BL to IP and the change in VL phase angle of the exercised leg.

<u>Changes in Tensiomyographical Parameters</u>

Results of this study demonstrate significant reductions in maximal displacement and contraction velocity of the RF in both legs immediately after exercise which are collectively suggestive of fatigue. This is in accordance with previous research demonstrating rapid impairments in muscle contractility (e.g., maximal displacement and contraction velocity) following various resistance exercise protocols (de Paula Simola et al., 2015; García-Manso et al., 2012; Hunter et al., 2012) and in response to electrically-induced fatigue (MacGregor et al., 2016). The current findings also indicate no change in contraction time which is consistent with previous research reporting unaltered Tc immediately after eccentric exercise using the biceps brachii of the dominant arm in untrained men (Harmsen et al., 2018). Conversely, others have noted a significant rise in contraction time 24 hours after eccentric exercise using the elbow flexors of the

nondominant arm among untrained men (Hunter et al., 2012). While the discrepancy in previous findings may be attributed to limb dominance, it is plausible that the lack of change in contraction time of the exercised leg is indicative of the absence of altered maximal isometric strength. Furthermore, delay time of the exercised VL and the RF of both legs was significantly reduced at IP with a greater potential reduction for the exercised leg. It is possible that the observed reduction in delay time from BL to IP was a function of lowered anticipatory muscle inhibition as participants became more familiarized with the electrical stimulus. Alternatively, as delay time provides a measure of muscle responsiveness (MacGregor et al., 2018), this finding appears to indicate a potentiating effect of the exercise protocol employed during this study. Reduced delay time has also been noted following a repeated sprint ability test (7×30 m) (Sánchez-Sánchez et al., 2018), where the authors speculated that the improved responsiveness implied a potentiating effect in the absence of local muscular fatigue. Considering the inconsistent effort given by our participants, in conjunction with a lack of performance decrements (i.e., peak torque, rate of torque development, and contraction time), it is possible that the decreased delay time reflects a similar potentiating effect of the exercise protocol used in this study.

Significant correlations have been identified between alterations in TMG contractile properties and decreases in maximal force production after strength training (de Paula Simola et al., 2015; Hunter et al., 2012). In fact, maximal displacement is regarded as an indicator of muscular fatigue (García-Manso et al., 2012), as it has been suggested that the decrease in maximal displacement mimics the pattern of other commonly assessed

markers of exercise-induced muscle damage (e.g., peak torque and rate of torque development) (Hunter et al., 2012). While no correlations were identified between the change in maximal displacement and any of the strength measures, the change in VL contraction velocity from BL to IP was shown to correlate strongly with changes in peak torque and rate of torque development which further supports the notion that the VL contributes largely to rapid force generation.

<div align="center">Changes in Sonomyographical Measures</div>

Bilateral increases in size (i.e., MT and CSA) of the RF and VL were observed in response to the unaccustomed exercise protocol. These observed changes in the ipsilateral leg are consistent with previous research reporting a temporary rise in muscle size following resistance exercise in an untrained population (Nosaka & Sakamoto, 2001; Radaelli et al., 2012, 2014). While the increase in size is often attributed to exercise-induced swelling, edema, and/or hyperemia (Kristiansen et al., 2014; Matta et al., 2017; Nosaka & Clarkson, 1996; Radaelli et al., 2014), it has also been linked to the development of muscle fatigue (Shi et al., 2007). Furthermore, while contralateral increases in muscle size from an acute bout of resistance exercise are not typical, the changes in CSA and MT observed in the non-exercised RF and VL support our theory that contralateral stabilization, though unanticipated, was a sufficient stimulus to elicit muscle fatigue and its associated swelling.

Following exercise, significant increases in echo intensity (EI) of the exercised RF were observed which were greater than those noted for the non-exercised RF. These acute

changes have been previously associated with the incurrence of muscle damage (Chen et al., 2011; Chen & Nosaka, 2006; Radaelli et al., 2012). A number of potential explanations have been proposed with respect to acute changes in EI observed. While some attribute the change to post-exercise glycogen depletion (Hill & San Millan, 2014; Nieman et al., 2015) or edema (Pillen & van Alfen, 2011), others suggest ultrastructural damage may govern this alteration such that altered EI represents sarcomeric disruption (Matta et al., 2017; Radaelli et al., 2012). Nevertheless, the incidence of a short-term elevation in EI is inconsistent. Jajtner and colleagues (2015) observed a significant increase in quadriceps EI immediately after completing a high-volume resistance exercise protocol, while others did not detect any rise in EI until 24-48 hours post-exercise (Radaelli et al., 2012, 2014). Furthermore, the pattern of early exercise-induced alterations in EI does not consistently align with that of other markers of post-exercise indices of recovery (Chen et al., 2011; Fujikake et al., 2009; Jajtner et al., 2015; Radaelli et al., 2012, 2014) and its implications on muscle function remain unclear. Accordingly, no significant correlations were identified between the change in corrected EI from BL to IP and any other changes in myographical or strength parameters.

<div align="center">Conclusions</div>

Changes in myographical parameters in the absence of impaired torque production support the use of electrical impedance myography, sonomyography, and tensiomyography as effective, noninvasive tools for the assessment of muscular fatigue following unaccustomed exercise. Furthermore, the detection of such changes without any apparent muscle damage highlights the sensitivity of these assessments.

Although many of the changes occurred bilaterally, VL delay time was significantly altered at IP in the exercised leg only suggesting greater involvement during dynamic movements involving the knee extensors. Compared to the eccentrically-exercised leg, the contralateral stabilization carried out with the non-exercised leg resulted in a number of comparable, if not equivocal, myographical changes. Results of the current study demonstrate that different muscle actions (i.e., eccentric and isometric) performed at submaximal intensities promote similar consequences to muscle strength, size, echogenicity, contractility, and bioelectrical properties.

APPENDIX A: UCF IRB LETTER

University of Central Florida Institutional Review Board
Office of Research & Commercialization
12201 Research Parkway, Suite 501
Orlando, Florida 32826-3246
Telephone: 407-823-2901 or 407-882-2276
www.research.ucf.edu/compliance/irb.html

Approval of Human Research

From: **UCF Institutional Review Board #1**
FWA00000351, IRB00001138

To: **Carleigh H. Boone, MS** and Co-Pls: **Ariel D. Boffey, Chad H. Herring, David Fukuda, Erica Goldstein, Tristan Michael Starling-Smith**

Date: **November 15, 2018**

Dear Researcher:

On 11/15/2018 the IRB approved the following human participant research until 11/14/2019 inclusive:

Type of Review:	UCF Initial Review Submission Form
	Expedited Review
Project Title:	Noninvasive myographical assessments following unaccustomed resistance exercise
Investigator:	Carleigh H Boone, MS
IRB Number:	BIO-18-14518
Funding Agency:	
Grant Title:	
Research ID:	N/A

The scientific merit of the research was considered during the IRB review. The Continuing Review Application must be submitted 30 days prior to the expiration date for studies that were previously expedited, and 60 days prior to the expiration date for research that was previously reviewed at a convened meeting. Do not make changes to the study (i.e., protocol, methodology, consent form, personnel, site, etc.) before obtaining IRB approval. A Modification Form **cannot** be used to extend the approval period of a study. All forms may be completed and submitted online at https://iris.research.ucf.edu .

If continuing review approval is not granted before the expiration date of 11/14/2019, approval of this research expires on that date. When you have completed your research, please submit a Study Closure request in iRIS so that IRB records will be accurate.

Use of the approved, stamped consent document(s) is required. The new form supersedes all previous versions, which are now invalid for further use. Only approved investigators (or other approved key study personnel) may solicit consent for research participation. Participants or their representatives must receive a signed and dated copy of the consent form(s).

All data, including signed consent forms if applicable, must be retained and secured per protocol for a minimum of five years (six if HIPAA applies) past the completion of this research. Any links to the identification of participants should be maintained and secured per protocol. Additional requirements may be imposed by your funding agency, your department, or other entities. Access to data is limited to authorized individuals listed as key study personnel.

In the conduct of this research, you are responsible to follow the requirements of the Investigator Manual.

This letter is signed by:

Signature applied by Adrienne Showman on 11/15/2018 01:34:24 PM EST

Designated Reviewer

APPENDIX B: CHANGES IN TMG PARAMETERS

Table 6: Changes in TMG parameters of the dominant and nondominant legs following unaccustomed resistance exercise

	BL	IP	24H
Dominant			
RF Dm (mm)	6.86 ± 3.06	5.72 ± 2.48	6.21 ± 3.15
RF Td (ms)	26.96 ± 3.43	25.54 ± 3.26	26.97 ± 3.99
RF Tc (ms)	32.16 ± 8.09	32.52 ± 8.65	31.93 ± 7.87
RF Vc (mm/ms^{-1})	0.12 ± 0.06	0.10 ± 0.05	0.11 ± 0.06
VL Dm (mm)	2.76 ± 1.77	3.05 ± 1.82	2.82 ± 1.74
VL Td (ms)	20.34 ± 1.46	20.16 ± 1.74	20.81 ± 1.82
VL Tc (ms)	17.84 ± 3.88	18.69 ± 3.63	18.61 ± 3.49
VL Vc (mm/ms^{-1})	0.07 ± 0.04	0.08 ± 0.04	0.07 ± 0.04
Nondominant			
RF Dm (mm)	6.56 ± 2.99	5.61 ± 2.66	6.36 ± 3.08
RF Td (ms)	27.13 ± 4.89	26.16 ± 5.25	27.58 ± 4.18
RF Tc (ms)	30.27 ± 5.33	32.41 ± 4.52	31.86 ± 7.02
RF Vc (mm/ms^{-1})	0.12 ± 0.06	0.10 ± 0.05	0.11 ± 0.06
VL Dm (mm)	2.42 ± 1.78	2.33 ± 1.58	2.23 ± 1.59
VL Td (ms)	20.12 ± 2.53	18.91 ± 2.19	20.23 ± 2.20
VL Tc (ms)	16.93 ± 4.32	17.15 ± 4.86	18.09 ± 4.51
VL Vc (mm/ms^{-1})	0.06 ± 0.04	0.06 ± 0.04	0.05 ± 0.03

Values are means ± SD

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