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ACUTE PHYSIOLOGICAL AND PERCEPTUAL RESPONSES TO RESISTANCE
EXERCISE WITH BLOOD FLOW RESTRICTION IN INDIVIDUALS WITH
MULTIPLE SCLEROSIS

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A DISSERTATION APPROVED FOR THE
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“Eu sozinho em meu caminho

Sou eu, sou todos, sou tudo

E isso sem nunca ter contudo

Jamais ficado sozinho.”

(Paulo César Pinheiro)

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Abstract

OBJECTIVE: This study aimed to compare the acute physiological and perceptual effects of low-load blood flow restriction (BFR) resistance exercise (LLBFR+RE) and high-load resistance exercise without blood flow restriction (HL-RE) in people living with multiple sclerosis (MS). Fifteen individuals (4 males and 11 females) with a physician-confirmed diagnosis of relapsing remitting MS and a disability score ≤ 6.5 volunteered to participate. **METHODS:** Participants completed a total of five visits to the laboratory. Visit 1 consisted of consenting and filling out standardized forms and questionnaires. During visit 2, participants completed measurements of several cardiovascular parameters, total arterial occlusion pressure for each leg, and completed the one-repetition maximum (1-RM) test for the leg press and knee extension exercises. Visit 3 included measurements of total body and regional body composition and bone mineral density using dual energy X-ray absorptiometry, then the 1-RM test for the same exercises was repeated. Visits 4 and 5 consisted of randomly completing the following experimental conditions: LLBFR+RE, consisting of 30+15+15+15 repetitions of leg press and knee extension at 20% of 1-RM, combined with 50% of BFR; and HL-RE, which included 4 sets of 10 repetitions of the same leg press and knee extension exercises at 75% of 1-RM, without BFR. Venous blood samples were collected and used to measure the plasma concentrations of whole-blood lactate, cortisol, interleukin-6 (IL-6), myostatin, and the mammalian target of rapamycin (mTOR), at baseline, 5 minutes post-, and 60 minutes post-exercise. The same blood samples were also used to measure hematocrit concentration and plasma volume changes at the same time points.

Additionally, muscle swelling was estimated through muscle thickness and thigh circumference measures, taken at baseline and at 30 minutes and 60 minutes post-exercise. Myoelectric activity of the vastus medialis and vastus lateralis muscles of the right and left leg was measured using surface electromyography (sEMG) during each experimental exercise condition. The perceptual responses consisted of ratings of perceived exertion (RPE), measured immediately after completion of each set of exercise; ratings of pain, measure immediately before and immediately after each set; and levels of soreness, measured before exercise and 5 minutes, 30 minutes, 60 minutes, and 24 hours post-exercise. All perceptual variables were measured using validated visual numeric scales. All physiological data were analyzed using parametric statistics; thus, two-way (condition \times time) repeated measures analyses of variance were used to test all main effects and interactions. In the case of significant interactions, pairwise *t* tests were used to test the simple effects. Familywise error rate was controlled using the Bonferroni procedure. The perceptual data were analyzed using non-parametric statistics; therefore, the Wilcoxon test was used to compare the two experimental conditions within specific time points. The Friedman's nonparametric test was used to test for significant differences in the median rank scores across the time points. If a significant difference was detected, pairwise Wilcoxon nonparametric tests with Bonferroni procedure were used to locate the differences. **RESULTS:** Whole-blood lactate levels significantly ($p < 0.05$) increased 5 min post-exercise compared to pre-exercise values, with HL-RE displaying significantly ($p < 0.05$) greater increases than LLBFR+RE. No significant ($p > 0.05$) condition or time effects were observed for plasma concentrations of myostatin, IL-6, and mTOR. Although a significant ($p < 0.05$) condition effect was also not detected for

cortisol, a significant ($p < 0.05$) decrease from baseline was observed for both conditions 1 hour post-exercise. There were also no significant ($p > 0.05$) time or condition effects for changes in hematocrit concentration and plasma volume. Muscle thickness and thigh circumference significantly ($p < 0.05$) increased from baseline immediately post-exercise following both experimental trials, with no significant ($p > 0.05$) differences between conditions. The HL-RE condition elicited significantly ($p < 0.05$) greater myoelectric activity than the $LLBFR+RE$ trial for the vastus medialis and vastus lateralis muscles and during the leg press and knee extension exercises. Regarding the perceptual responses, HL-RE resulted in significantly ($p < 0.05$) greater RPE than $LLBFR+RE$ during leg press and knee extension. Similar ($p > 0.05$) ratings of pain were observed during both experimental exercise conditions immediately after each set, however, for the ratings of pain measured immediately before each set, $LLBFR+RE$ induced significantly ($p < 0.05$) greater pain than HL-RE. Finally, no significant ($p > 0.05$) increases in muscle soreness were observed up to 24 hours post-exercise following both trials. **CONCLUSIONS:** This study demonstrated that people living with MS are capable of tolerating and performing $LLBFR+RE$ without any major adverse effects. This study also demonstrated that $LLBFR+RE$ is capable of acutely increasing many of the physiological parameters related to the hypertrophic response commonly observed following traditional resistance exercise without BFR, indicating that it may potentially serve as a training alternative to HL-RE for MS patients unable or unwilling to lift heavy loads. The perceptual data also demonstrated that $LLBFR+RE$ requires less muscular exertion compared to HL-RE, and does not cause exaggerated pain during exercise or elevated delayed-onset muscle soreness up to 24 h post-exercise.

Chapter I: Introduction

Multiple sclerosis (MS) [from the Greek *skleros*, meaning hard] is an inflammatory, auto-immune disease (Weiner, 2004) of the central nervous system, characterized by neuron axonal demyelination (Love, 2006) at the level of the brain and the spinal cord. MS is thought to be caused by an interaction of several environmental (Pugliatti et al., 2008) and genetic (Compston, 1999) factors, with some studies suggesting that viral infections may also be responsible for triggering the disease (Alvarez-Lafuente et al., 2004). In addition to the fact that the direct contribution of viruses to the development of MS remains unclear (Owens et al., 2011), the etiology of the disease also remains elusive. It has been estimated that approximately 2.3 million people around the world are living with MS (National Multiple Sclerosis Society, 2018). The expensive healthcare costs of treating the MS symptoms ultimately lead to significant social and economic burdens.

A wide range of symptoms may be present in people living with MS. Most of these symptoms involve problems related to physical function, which oftentimes include fatigue, imbalance, weakness, increased muscle tension, and even paralysis (Bakshi, 2003). Nevertheless, symptoms unrelated to physical function may also be present, such as pain, bowel, visual, and communicating problems, and even depression (Bakshi, 2003). Thus, researchers from all over the world have sought possible interventions to decrease the healthcare costs and to attenuate some of these symptoms, subsequently improving the quality of life of individuals affected by this disease.

The impaired physical function related to MS is primarily due to skeletal muscle atrophy and decreased muscular strength, which is commonly observed in these

individuals (Wens et al., 2014). Therefore, exercise interventions that are capable of maintaining skeletal muscle mass or promoting muscle hypertrophy and increasing muscular strength would be extremely beneficial to this clinical population. Resistance exercise training is commonly used to induce these positive adaptations in healthy individuals (Burd et al., 2010). The observed positive outcomes associated with resistance training in healthy individuals has led to the investigation of its use as a potential intervention for improving physical capacities in individuals with MS. White et al. (2004) conducted a study in which people living with MS performed a twice-weekly resistance training program (8 to 10 repetitions at 50% of one-repetition maximum (1-RM)) for the lower-body over the course of 8 weeks on measures of muscle size and strength. The authors observed that the significant increases in muscle cross-sectional area and strength were also accompanied by a decrease in self-reported fatigue. Similar results were reported by Souza-Teixeira et al. (2009), who identified significant improvements in muscle hypertrophy, power, strength, and muscular endurance following 8 weeks of progressive moderate resistance exercise. In both studies, the increases in muscular performance were observed as early as in 2 weeks of training. Several other studies have confirmed the benefits of resistance exercise in MS participants in terms of improving muscle size (Dalgas et al., 2010), strength (Manca et al., 2017), physical function (Kjølhede et al., 2015), and even potential neuroprotective effects (González Torre et al., 2017).

Although previous research has confirmed the effectiveness of resistance training to induce significant muscle hypertrophy (Shepstone et al., 2005) and strength gains (Munn et al., 2005) in healthy individuals, this training modality imposes a challenge to

clinical populations, especially to people living with MS. This is attributed to the fact that it is traditionally recommend that resistance exercise should be performed using training loads that are superior to 65% of 1-RM (ACSM, 2009). Accordingly, such high training loads require great effort from participants and often induces oxidative stress (Çakır-Atabek et al., 2015; McBride et al., 1998), muscle damage (Kanda et al., 2013), delayed-onset muscle soreness (Cleak & Eston, 1992), and inflammation (Heavens et al., 2014), in addition to the risk of causing musculoarticular injuries, making the implementation of this method of training unfeasible, or at least risky, for this clinical population. Therefore, there is a critical need to develop exercise modalities that are capable of increasing muscle size and strength in people living with MS without incurring into the adverse effects typically observed with high-load resistance exercise. Low-load resistance exercise would serve as a safer training modality as it has been shown to induce positive adaptations similar to those often observed with high-load resistance exercise (Ogasawara et al., 2013). However, when training using lower loads, repetitions need to be completed until volitional failure in order for the exercise to induce physiological responses that are similar to those from resistance exercise at higher loads, which means that much greater training times and exercise volumes are needed for these adaptations to be achieved, oftentimes making this type of training impractical. Nonetheless, in the past few decades, a new low-load, low-volume resistance exercise training program was developed, which is capable of inducing positive neuromuscular adaptations without causing the adverse effects typically observed with high-load resistance exercise. This training approach was first developed in Japan and is most commonly known as blood flow restriction (BFR) training or KAATSU training. This technique consists of wrapping standardized

inflatable cuffs around the most proximal portion of arms or legs during exercise, which are used to reduce arterial blood flow and occlude venous return.

The mechanisms through which BFR exercise elicits its positive adaptations remains unclear, although a myriad of factors seem to be involved (Pearson & Hussain, 2014). For instance, the venous occlusion induced by BFR exercise leads to an accumulation of metabolites in the intramuscular environment such as lactate ion (La^-), hydrogen ion (H^+), adenosine diphosphate (ADP), inorganic phosphate (P_i), and dihydrogen phosphate (H_2PO_4) (Suga et al., 2009, 2010; Sugaya et al., 2011; Yasuda et al., 2010). The buildup of these metabolic byproducts induce the release of anabolic hormones such as growth hormone (GH) (Takarada, Nakamura, et al., 2000) and insulin-like growth factor 1 (IGF-1) (Abe et al., 2005), possibly by the triggering of metaboreceptors and activation of the type III and IV afferent fibers. Due to decreased enzymatic activity and reduced oxygen availability, it has been hypothesized that BFR exercise may induce early fatigue of the aerobic type I muscle fibers and an early recruitment of the type II anaerobic muscle fibers. Type II muscle fibers are more responsive to training and often display a greater hypertrophic response to resistance training (Andersen & Aagaard, 2010). Moreover, the production and accumulation of metabolites inside the cell increases osmolarity and forces water to move from the interstitial space to inside of muscle fibers through osmosis, ultimately resulting in muscle swelling. This acute muscle swelling commonly observed post-exercise (Freitas et al., 2017) has also been proposed as one of the factors contributing to the BFR exercise hypertrophic response (Loenneke, Fahs, Rossow, Abe, & Bembien, 2012; Takarada, Takazawa, & Ishii, 2000). Finally, several other mechanisms are thought to be involved

with the physiological adaptations to BFR exercise, including the up and downregulation of several biomolecular pathways that regulate muscle protein synthesis (Fry et al., 2010; Fujita et al., 2007; Sudo et al., 2015) and degradation (Holliss et al., 2013; Laurentino et al., 2012).

Even though BFR exercise elicits acute physiological responses and chronic adaptations that are similar to those observed with traditional high-load resistance exercise (Laurentino et al., 2012; Lixandrão, Ugrinowitsch, et al., 2018b), low-load resistance exercise combined with BFR ($LLRE+BFR$) has been demonstrated to be relatively safe (Clark et al., 2011). Previous studies have reported that $LLBFR-RE$ does not induce muscle damage, delayed muscle soreness, oxidative stress, or inflammation (Goldfarb et al., 2008; Sudo et al., 2015; Thiebaud et al., 2014), as often observed with traditional resistance exercise. This makes this method of training applicable for those who cannot tolerate the high training loads commonly used with traditional resistance exercise, such as elderly, individuals recovering from surgeries, and clinical populations. To illustrate, Segal et al. (2015) reported significant increases in strength levels following 4 weeks of $LLRE+BFR$ performed 3 times a week in women with risk factors for symptomatic knee osteoarthritis. In another study, strength significantly increased following 12 weeks of $LLBFR-RE$ and was accompanied by increased muscle cross-sectional area and improved physical function in older individuals (Yasuda et al., 2014).

Therefore, as previously observed in other clinical populations, $LLRE+BFR$ may serve as a potential non-pharmacological method to attenuate skeletal muscle atrophy and weakness, commonly observed in those living with MS as the disease progresses, and it may also serve as an effective training method to increase muscle size, strength, and

physical function in these individuals. Finally, no research has yet investigated the potential benefits of LLBFR-RE in MS patients; however, the first step in this line of research in people living with MS is to document the psychophysiological acute response of LLBFR-RE compared to traditional high-load resistance exercise without BFR.

Purpose

The purpose of this study was to compare the acute physiological and perceptual responses of individuals living with MS to a single bout of low-load resistance exercise combined with BFR (LLRE+BFR) and high-load (70% 1RM) resistance exercise without BFR (HL-RE).

Research Questions

1. Does LLRE+BFR induce the same metabolic response (whole-blood lactate) as traditional HL-RE?
2. Are changes in electromyography amplitude similar between LLRE+BFR and HL-RE?
3. Is there a difference in the acute exercise-induced muscle swelling response (muscle thickness and thigh circumference) between LLRE+BFR and HL-RE?
4. Is the hormonal stress response (cortisol) similar between LLRE+BFR and HL-RE?
5. Do biomolecular markers of muscle anabolism (mTOR) and catabolism (myostatin) display similar responses to LLRE+BFR -RE and HL-RE?
6. Is the exercise-induced inflammatory response (interleukin-6) similar between LLRE+BFR and HL-RE?

7. Are the post-exercise changes in plasma volume and hematocrit levels similar between LLRE+BFR and HL-RE?
8. Do LLRE+BFR and HL-RE elicit similar ratings of perceived exertion?
9. Are pain levels perceived during LLRE+BFR similar to those perceived during HL-RE?
10. Is the 24-h post-exercise delayed-onset muscle soreness response similar between LLRE+BFR and HL-RE.

Research Subquestions

1. Were individuals living with MS able to complete the pre-determined standard BFR protocol (i.e., 4 sets of 30+15+15+15 repetitions at 20% of 1-RM)?
2. Were these participants able to complete the pre-determined high-load resistance exercise protocol (4 sets of 10 repetitions)?
3. Is there any difference in exercise volume between leg press and knee extension exercises within the same exercise protocol?
4. Were individuals with MS able to tolerate the application of BFR during exercise?
5. Was there any difference in electromyography amplitude when comparing muscles of the right and left legs?
6. Was 1-RM testing a reliable method to measure maximum dynamic strength in MS patients?

Hypotheses

1. Considering the literature suggesting the exercise-induced metabolic response as one of the potential mechanisms contributing to muscle hypertrophy following

LLRE+BFR and the several studies reporting similar hypertrophy gains following both LLRE+BFR and HL-RE, it was hypothesized that a similar metabolic response (whole-blood lactate) would be observed between the LLRE+BFR and HL-RE protocols.

2. Myoelectric activity during exercise would be greater during HL-RE in comparison to LLRE+BFR. This hypothesis was based on multiple studies demonstrating smaller myoelectric activity during LLRE+BFR compared to HL-RE.
3. There are also several studies demonstrating that LLRE+BFR and HL-RE may induce similar post-exercise responses. Thus, it was hypothesized that the exercise-induced muscle swelling response (muscle thickness and thigh circumference) would be similar between LLRE+BFR and HL-RE.
4. Although only a few studies have directly compared the hormonal stress response following LLRE+BFR and HL-RE, considering the higher mechanical stress involved with HL-RE, it was hypothesized that a greater hormonal stress (cortisol) response would be observed following HL-RE compared to LLRE+BFR.
5. As the regulation of biomolecular pathways has also been suggested as potential mechanisms through which both LLRE+BFR and HL-RE elicit their positive adaptations, it was hypothesized that similar levels of biomolecular markers of muscle

anabolism (mTOR) and catabolism (myostatin) would be observed following $LLRE+BFR$ and $HL-RE$.

6. The higher mechanical loads used during $HL-RE$ have been well documented to induce muscle damage after an exercise bout, whereas the current literature is yet to demonstrate that $LLRE+BFR$ induces any muscle damage. Considering the common inflammatory response taking place following damaging exercise, it was hypothesized greater inflammation (interleukin-6) would be observed following $HL-RE$ compared to $LLRE+BFR$.
7. There would be no difference in changes in plasma volume and hematocrit levels between $LLRE+BFR$ and $HL-RE$. This hypothesis was based on previous literature demonstrating minimal to no changes in plasma volume and hematocrit levels.
8. Considering the higher mechanical loads used during $HL-RE$, it was hypothesized that $HL-RE$ would result in greater ratings of perceived exertion (RPE) compared to $LLRE+BFR$.
9. Although one would naturally expect $LLRE+BFR$ to result in lower ratings of pain compared to $HL-RE$ due to the use of lower loads, it should also be considered that the restriction of blood flow may, on the other hand, contribute to increase the ratings of pain during exercise. Therefore, it was hypothesized that $LLRE+BFR$ would result in similar ratings of pain when compared to $HL-RE$.

10. As HL-RE is expected to result in greater muscle damage than LLRE+BFR, it was hypothesized that HL-RE would also result in greater ratings of delayed-onset muscle soreness 24 h post-exercise, while LLRE+BFR will not induce any delayed-onset muscle soreness.

Subhypotheses

1. Based on fact that participants are resistance untrained, not familiar with LLRE+BFR, and that individuals with MS fatigue more quickly compared to healthy individuals, it was hypothesized that most participants would not be able to complete all the repetitions for the 4 sets of the BFR exercise protocol.
2. Also considering the fact that participants are resistance untrained and have a compromised ability to perform high-load resistance exercise for prolonged periods of time, it was hypothesized that most participants would not be able to complete the 10 repetitions of the last 2 sets of the high-load resistance exercise protocol.
3. It was hypothesized that greater exercise volume would be observed with the leg press compared to knee extension exercise, as participants may experience greater fatigue during knee extension, which will be performed after the leg press.

4. Although unpleasant, considering the lower levels of BFR applied, it was hypothesized that most participants would be able to tolerate the application of BFR during exercise.
5. Taking into consideration the studies demonstrating limb asymmetry in people suffering from MS, it was hypothesized that left and right legs would display differences in sEMG amplitude.
6. Considering limb asymmetry and the fact that participants were not familiar with the technique of resistance training, it was hypothesized that 1-RM would not be a reliable testing method to assess maximum dynamic strength in people living with MS.

Significance of the Study

It is extremely important for people suffering from MS to be able to at least maintain adequate muscle size and strength levels in order to maintain adequate activities of daily living, since muscle atrophy and strength loss are common in these individuals. Additionally, it is critical to increase muscle size and strength in order to compensate for the normal decrements often observed over time as the disease progresses. Low-load resistance exercise combined with BFR has been shown to be effective at improving skeletal muscle mass and strength across a variety of populations, without the issues usually observed with traditional high-load resistance exercise, such as exercise-induced muscle damage, delayed-onset muscle soreness, oxidative stress, and inflammation, which make this type of exercise challenging for this population. Surprisingly, no study

has investigated the effects of resistance exercise with BFR and the physiological responses related to muscle hypertrophy in people living with MS. Therefore, this study will offer physicians and physiotherapists a possible non-pharmacological intervention that may be used to enhance muscle size and strength in individuals with MS, consequently translating into improved physical function and enhanced quality of life.

Assumptions

1. Participants were correctly diagnosed having MS by their physicians.
2. Participants were correctly quantified having a disability score ≤ 6.5 .
3. Participants responded to all questionnaires truthfully.
4. Participants carefully followed all instructions given by the investigator such as avoiding caffeine and strenuous exercise before each testing visit.
5. Muscle thickness measured with ultrasound, hematocrit levels, and plasma volume change are valid estimators of muscle swelling.
6. The ELISA kits used are valid and reliable methods to quantify myostatin expression and mTOR activity.

Delimitations

1. The findings from the current study are limited to individuals living with MS, and, thus, cannot be extended to other clinical populations.
2. These results can only be inferred to individuals with relapsing-remitting MS and with a disability score (EDSS) equal or below 6.5.

Limitations

1. Muscle swelling and plasma volume changes do not provide a direct measure of muscle cell swelling, caused by the influx of water into the intracellular space.
2. Electromyography provides only a gross estimation of muscle activation as it does not give any information related to motor unit recruitment or rate coding.
3. Acute changes in the variables related to muscle hypertrophy do not necessarily result in chronic skeletal muscle hypertrophy.

Abbreviations

1. 1-RM – One-repetition maximum.
2. ABI – Ankle-brachial index.
3. ADP – Adenosine diphosphate.
4. ATP – Adenosine triphosphate
5. BFR – Blood flow restriction.
6. EDSS – Expanded disability status scale.
7. GH – Growth hormone.
8. H⁺ – Hydrogen ion.
9. H₂PO₄ – Dihydrogen phosphate.
10. HCT – Hematocrit.
11. HL-RE – High-load resistance exercise.
12. IGF-1 – Insulin-like growth factor 1.
13. IL-6 – Interleukin 6.
14. La⁻ – Lactate.
15. LLBFR-RE – Low-load blood flow restriction resistance exercise.

16. MS – Multiple Sclerosis.
17. MSTN – Myostatin.
18. mTOR – Mammalian target of rapamycin.
19. Pi – Inorganic phosphate.
20. RPE – Ratings of perceived exertion.
21. sEMG – Surface electromyography.
22. WBL – Whole-blood lactate.

Operational Definitions

1. Adenosine diphosphate (ADP) – A phosphate group bound to an adenosine. It is of the metabolic products from the hydrolysis of adenosine triphosphate.
2. Blood flow restriction (BFR) resistance exercise – Resistance exercise performed while the blood flow to the working muscles is restricted by standardized restrictive cuffs.
3. Cortisol – A catabolic steroid hormone related to protein breakdown.
4. Electromyography – Technique used to indirectly measure muscle electrical activity.
5. Expanded disability status scale – A scale used to quantify the disability level in individuals with MS.
6. Hematocrit – A test that measures the proportion of red blood cells in the blood.
7. Hydrogen ion [H^+] – A hydrogen proton dissociated from a weak acid.
8. Inorganic phosphate (Pi) – One of the metabolic products from the hydrolysis of adenosine triphosphate.

9. Lactate [La^-] – Product of the reduction of pyruvate by pyruvate dehydrogenase in the lactic fermentation reaction.
10. Mammalian target of rapamycin complex 1 (mTORC1) – a regulator of protein synthesis.
11. Multiple sclerosis (MS) – A neurological disease characterized by the axonal demyelination.
12. Muscle thickness (MT) – The distance measured from the adipose tissue-muscle interface to the muscle-bone interface using ultrasound. Used as an estimator of muscle swelling.
13. Myostatin (MSTN) – An inhibitor of muscle growth.
14. One-repetition maximum (1-RM) – The maximum amount of weight that can be lifted with a single concentric and eccentric contraction.
15. pKa – Negative base-10 logarithm of the acid dissociation constant of a solution.
16. Plasma volume change – The change in plasma volume in the blood over a certain period of time.
17. OMNI-RES scale – A scale used to quantify the amount of pain or discomfort felt post a set or a bout of resistance exercise in a scale from 0 to 11.
18. Ratings of perceived exertion (RPE) scale – A scaled used to quantify how strenuous and heavy a set or a bout of resistance exercise feels in a scale from 0 to 10.
19. Thigh circumference – The circumference of the thigh measured at the 50% distance from the great trochanter to the lateral condyle of the femur.
20. Whole-blood lactate (WBL) – The blood lactate concentration in mmol/L. It represents the overall net lactate production and removal at the whole-body level.

Chapter II: Literature Review

Mechanisms and Physiology of Blood Flow Restriction Exercise

Although resistance training combined with BFR has been shown to effectively enhance muscle size and strength across a wide variety of populations, the mechanisms through which this model of training exerts its positive adaptations remain unclear. A myriad of possible factors has been used in an attempt to explain these adaptations. The following sections will explore in detail the main factors claimed to be involved with the benefits of low-load resistance exercise with BFR (LLRE+BFR).

Metabolic Response

Traditional high-intensity resistance training has been proven to significantly increase muscle size and strength. Although the specific mechanisms responsible for these adaptations are not fully understood, this exercise training is thought to elicit its benefits through the activation of molecular pathways (i.e., mTORC1 and MAPK) that ultimately increase protein synthesis. The activation of these pathways is believed to be triggered through mechanotransduction, in which the mechanical stress placed on the muscle is converted into a chemical signal used to initiate a cascade of events within the muscle cells. However, since low-load resistance training with BFR has also been shown to induce increases in muscle size and strength that are similar to those observed with traditional high-load resistance training, but at much lower mechanical stresses (~70% of 1RM vs ~30% of 1RM), additional mechanisms have been considered. Even though the extent of the mechanical stress applied during each training method (BFR [20 – 30% of 1RM] vs traditional [65 – 80% 1 of RM] resistance exercise) is considerably different,

previous studies have reported that the metabolic stress evoked by both exercise models are similar. Therefore, metabolic stress has emerged as a possible factor that may play a crucial role in the positive adaptations observed with low-intensity resistance training with BFR. Supporting the metabolic stress hypothesis, Takarada et al. (2012) reported increases in levels of muscle mass and strength increases that were proportional to the increases in the exercise-induced metabolic stress.

The exercise-induced metabolic changes observed with LLRE+BFR includes accumulation of lactate (La^-), dihydrogen phosphate (H_2PO_4), inorganic phosphate (Pi), and concomitant decreases in pH due to the accumulation of hydrogen ions (H^+) (Suga et al., 2012; Sugaya et al., 2011; Yasuda et al., 2010). Suga et al. (2010) demonstrated that the intramuscular changes observed immediately post one single bout of LLRE+BFR performed at 30% of 1RM were similar to those from high-load resistance exercise (HL-RE) at 65% of 1RM. Previous studies have also demonstrated that this increased post-exercise metabolic response is accompanied by increased plasma levels of anabolic hormones such as growth hormone and insulin-like growth factor-1 (IGF-1) (Abe et al., 2005; Madarame et al., 2010; Manini et al., 2012; Pierce et al., 2005; Takano et al., 2005; Takarada et al., 2000). Manini et al. reported a positive correlation between lactate and growth hormone concentrations (2012). However, there is an intense debate in the literature regarding whether exercise-induced increased levels of endogenous anabolic hormones may or may not have any additive effect in the hypertrophic responses to resistance training (Morton et al., 2016; Schroeder et al., 2013; West et al., 2009, 2010; Wilkinson et al., 2006).

The greatest contribution of the metabolic responses to the adaptations to BFR resistance exercise lie in the fact that these metabolites may help anticipate the onset of muscle fatigue and increased motor unit recruitment. Low pH and Pi accumulation are known for inducing fatigue by inhibiting cross-bridge cycling (Debold, 2012) and by causing impairment of calcium kinetics, due to altered activity of the sarco/endoplasmic reticulum calcium ATPase enzyme (Allen & Westerblad, 2001). Therefore, as muscle fatigue onsets, additional motor units are recruited in order for the activity to be maintained. Additionally, accumulation of H⁺ may also impair intracellular metabolism and inhibit phosphofructokinase 1 activity, the enzyme responsible for the commitment step in glycolysis, therefore, limiting carbohydrate utilization and ATP synthesis, especially in the anaerobic type II glycolic muscle fibers.

To date, no study has been performed that has documented the direct contribution of metabolites to the hypertrophic responses of both traditional high-intensity and low-intensity resistance exercise with BFR. However, there is a body of evidence suggesting a potential indirect contribution of the buildup of metabolites to the positive adaptations observed with both training methods.

Hormonal Responses

Low-load resistance exercise combined with BFR has been shown to be capable of acutely affecting the production and secretion of certain anabolic (GH and IGF-I) and catabolic (cortisol) hormones (Abe et al., 2005; Manini et al., 2012; Takarada, Nakamura, et al., 2000), while others, such as testosterone, do not seem to be affected (Reeves et al., 2005). Although an intense debate among exercise physiologists persists regarding

whether anabolic hormones can actually contribute significantly to the hypertrophic response to resistance exercise (Morton et al., 2016; Schroeder et al., 2013; West et al., 2009, 2010; Wilkinson et al., 2006), the plasma concentration of anabolic hormones has also been considered as one of the possible contributors to the chronic adaptations to $LLRE+BFR$.

An early study by Takarada et al. (2000) demonstrated the potential of $LLRE+BFR$ to induce significant increases in plasma levels of anabolic hormones post-exercise. These findings were later corroborated by many other research groups (Abe et al., 2005; Karabulut et al., 2013; Madarame et al., 2010; Manini et al., 2012; Reeves et al., 2005; Takano et al., 2005; Takarada et al., 2014). Although it has been shown that $LLRE+BFR$ is a potent stimulus for the secretion of anabolic hormones, particularly GH and IGF-1, the mechanisms underlying this exercise-induced endocrine response is not completely understood; although, it has been speculated that this increased release of GH and IGF-I may be linked to the acute metabolic stress experienced during $LLRE+BFR$ (Goto et al., 2005; Viru et al., 1998).

It is possible that metaboreceptors may induce the secretion of GH through the afferent-pituitary axis, by sympathetic stimulation via muscle afferent fibers. The muscle afferent fibers are divided into types I, II, III, and IV. The groups III and IV muscle afferent fibers are sensitive to both mechanical stimuli and to metabolic byproducts during ischemic contractions (Kaufman & Rybicki, 1987). Kaufman and Rybicki (1987) were one of the first to indicate the sensitivity of these afferent fibers to metabolic stimuli, but the authors were unable to determine what specific metabolites were responsible for inducing such stimulation. However, in a later experiment, Rotto and Kaufman (1988)

infused the triceps surae of cats with metabolites known for accumulating within the muscle during exercise (phosphate, La^- , lactic acid, adenosine, and arachidonic acid) and observed that only lactic acid and arachidonic acid were able to activate both type III and IV afferent fibers. These findings are of great relevance considering that lactic acid but not La^- was able to activate these fibers. Due to its pKa of 3.86, at physiological pH ($\approx 7.35 - 7.45$), lactic acid dissociates into a lactate ion, the conjugate base, and a hydrogen ion (i.e., $\text{La} = \text{La}^- + \text{H}^+$), which consequently leads to a decrease in pH. For this reason, the authors then hypothesized that this decrease in pH was probably responsible for the activation of these two afferent fibers. Later work by Sinoway et al. (1993) confirmed the contribution of lactic acid to the activation of the type III afferent muscle fibers.

To study the impact of the metabolic stress on the endocrine response to resistance exercise, Goto et al. (2005) investigated the changes in GH, epinephrine, norepinephrine, and La^- following an acute bout of two different bouts of resistance exercise at the same intensity (75% 1-RM) and volume (3 – 5 sets with 10 repetitions each): 1) no rest regimen (NR) and 2) with rest regimen (WR). The only difference between these two protocols was that an intraset period of 30 seconds between the fifth and the sixth repetition in addition to the 1-min rest period between sets was allowed for the WR but not for the NR trial. The authors observed a greater hormonal concentration as well as a higher metabolic response for the exercise protocol without an intraset period (NR). In the same study, the authors also observed greater increases in muscular size, strength, and endurance following 12 weeks of training for the NR. Regarding LLRE+BFR , other studies have also observed a greater hormonal response accompanied by significant metabolic stress (Madarame et al., 2010; Manini et al., 2012; Takarada, Nakamura, et al., 2000).

Although these studies reinforce the hypothesis of an exercise-induced metabolite response contributing to the positive adaptations to $LLRE+BFR$, they are limited by their inability to control for other possible factors. For example, greater metabolic responses are generally followed by increased muscle activation. In this regard, Gosselink et al. (1998) observed that exercise using nerve electrical stimulation was also capable of exciting type I and type II afferent muscle fibers and that these afferent fibers were able to modulate the secretion of GH either stimulating or inhibiting hormone release through a muscle fiber type fashion model (K. Gosselink & Grindeland, 2000; K. L. Gosselink et al., 1998). Therefore, muscle activation may also play an important role in the exercise-induced hormonal response, however, current studies have presented limited methodological designs in an attempt to test the sole contribution of the metabolic accumulation on the endocrine response.

Muscle Activation and Fatigue

Muscle activation has been considered as one of the most important variables driving the positive chronic adaptations to $LLRE+BFR$. Before delving into the details regarding the effects of this mode of exercise, it is important to highlight that two main features distinguishing $LLRE+BFR$ from traditional $HL-RE$ without BFR : 1) the reduction of blood flow to the active muscle and 2) the use of lower relative exercise loads, usually within 20 to 30% of 1-RM. Although being performed at significantly lower loads, the reduced blood flow induces an early fatigue of type I muscle fibers, in terms of lower force production capacity, and an early activation of the higher threshold type II muscle

fibers in order to maintain muscular work. Therefore, as seen with traditional HL-RE, low-load resistance exercise is also capable of activating the type II muscle fibers.

In fact, similar levels of muscle activation have been reported between HL-RE and LLRE+BFR. Takarada et al. (2000) reported similar levels of muscle activation between elbow flexion at 40% of 1-RM with BFR and 80% of 1-RM without BFR. Moreover, Suga et al. (2012) investigated muscle fiber recruitment by splitting of Pi using splitting Pi peak and observed muscle fiber recruitment during to that of HL-RE without BFR.

Biomolecular Signaling Pathways

In addition to the mechanisms detailed above, the modulation of molecular signaling pathways seem to play a critical role regarding the adaptations to BFR resistance training. Previous studies have reported that this exercise method is capable of upregulating and downregulating several cellular pathways involved with protein synthesis and degradation and muscular hypertrophy in young and even in older individuals (Fry et al., 2010; Fujita et al., 2007; Laurentino et al., 2012; Nakajima et al., 2016; Sudo et al., 2015).

One of the main signaling pathways that is thought to be involved with muscle growth through an increase in protein synthesis is the mammalian target of rapamycin (mTOR). mTOR is a multidomain protein kinase that phosphorylates serine and threonine residues and ultimately activate downstream pathways that result in increased protein synthesis. Activation of the mTOR signaling pathway may occur through nutritional, chemical, as well as mechanical factors. Therefore, exercises that are capable of

activating greater muscle mass and higher threshold type II muscle fiber such as HL-RE have been shown to activate this pathway. Since LLRE+BFR has been shown to activate high-threshold type II muscle fibers to levels similar to those from HL-RE, the activation of the mTOR pathway has been considered as an important factor for the hypertrophic response observed with BFR training. Fujita et al. (2007) demonstrated that an acute bout of LLRE+BFR (30+15+15+15 sets of bilateral leg extension at 20% of 1-RM) was capable of stimulating ribosomal S6 kinase 1 phosphorylation, one of the downstream targets of mTOR, and protein synthesis in human skeletal muscle. Similar results were observed by Fry (2010) in older males. In both studies, significant mTOR stimulation was observed following an acute bout of LLRE+BFR but not after the same exercise protocol performed at the same relative workload and intensity without BFR.

While mTOR works as a crucial positive regulator of muscle hypertrophy, myostatin – or growth differentiation factor-8 – plays a role as a negative regulator of muscle mass. Increased expression of myostatin commonly leads to reduced muscle mass, accompanied by reduced fiber size. Moreover, in animal models, knockout of myostatin leads to exaggerated muscle mass. Therefore, myostatin has been considered another crucial regulator of muscle hypertrophy. In this sense, only one study has compared the mRNA expression of genes related to myostatin signaling post 8 weeks of both HL-RE (80% of 1RM) and LLRE+BFR (20% of 1RM) (Laurentino et al., 2012). The authors reported, that both training modalities induced significant gains in muscle size and strength, which were accompanied by significant diminished myostatin gene expression.

Although mTOR activation seems to be essential for muscle growth, several mechanisms seem to induce its activation. These include mechanical deformation of the

fiber via the mechanical stress imposed by resistance exercise as well as the action of anabolic hormones, especially GH and IGF-1 (either liver or muscle-derived forms). However, it should be highlighted that additional pathways independent of mTOR activation may also contribute to increase protein synthesis and consequent skeletal muscle hypertrophy. These pathways include testosterone-mediated gene expression and satellite cell activation. Satellite cell activation is another mechanism that may induce muscle growth. These consist of resident skeletal muscle stem cells that become active primarily following muscle damage. Once activated, satellite cells migrate into the muscle fiber and differentiates into a cell nucleus, thus increasing the nuclei pool within the muscle fiber. As gene expression and protein synthesis occurs within the cell nucleus, increased satellite cell density within muscle fibers increases the muscle fibers' gene expression capacity.

Therefore, there is strong evidence reported throughout the literature regarding the potential of BFR resistance exercise to activate biomolecular signaling pathways within the muscle that favor protein synthesis and ultimately contributes to the gains in muscle size and strength extensively.

Muscle Swelling

Although LLRE+BFR has been shown to significantly improve muscle size, strength, and power, the application of BFR in the absence of exercise has also been shown to positively affect muscle size by attenuating muscle atrophy in post-operative patients.

Takarada et al. (2000) was one of the first to report the benefits of the application of BFR in the absence of exercise following surgery of the anterior cruciate ligament. Participants were submitted to an occlusive stimulus twice a day from the 3rd to the 14th day post-surgery (total of 10 days). The stimulus was applied using pneumatic cuffs and consisted of 5 bouts of 5 min of BFR (180 to 238 mm Hg) and a 3 min break period between bouts. The authors observed that the application of BFR in the absence of exercise diminished muscle atrophy, normally observed following this medical procedure, as participants in the experimental group displayed lower muscle wasting compared to participants in the control group. Although no other measures that could potentially explain how the occlusive stimulus was involved in this response, this study provided the first evidence that the application of BFR in the absence of exercise may positively affect muscle physiology. In this study, the authors assumed that BFR in the absence of exercise may affect some of the factors that are thought to be involved in the adaptive response to BFR exercise such as hormone production and metabolite accumulation, although such measures were not taken. However, in a subsequent study, Kubota et al. (2008) investigated the effects of the applying BFR without exercise in muscle size changes as well as muscle function and GH plasma concentration. Muscle weakness was induced by cast immobilization of the left ankle for a period of 2 weeks. Participants were allocated into a control, an experimental BFR, or an exercise group without BFR. Participants in the BFR group were exposed to sessions of BFR over the course of 2 weeks, twice a day as performed in the study by Takarada et al. (2000). Participants in the exercise group performed isometric contraction twice a day for the same period of time; and participants in the control group were not exposed to any

intervention. The authors reported that after 2 weeks of cast immobilization, the application of BFR without exercise significantly prevented muscle weakness and atrophy compared to the control group that displayed significant decreases in muscle size and strength. These results displayed in the BFR group were even greater than those from the isometric exercise group for some of the measures of muscle strength. Additionally, these protective effects of BFR application occurred without any change in blood levels of GH. These findings from Kubota et al. (2008) reinforce those from Takarada et al. (2000) in which BFR application in the absence of exercise has an antiatrophic effect and also seems to help to preserve muscle function. However, even though these findings are relevant, these studies did not provide any evidence of how the application of BFR by without exercise was able to induce these responses. Loenneke et al. (2012) hypothesized that muscle swelling was most likely the driving factor responsible for these observations by triggering the activation of molecular pathways that ultimately result in gene expression and protein synthesis. This hypothesis was strengthened when the authors replicated the study design from Takarada et al. (2000) and Kubota et al. (2008) and observed significant changes in muscle thickness accompanied by changes in plasma volume with no changes in La^- concentration or electromyography amplitude (Loenneke et al., 2012). It is also important to highlight that the muscle swelling observed during LLRE+BFR is similar to that from HL-RE without BFR (Freitas et al., 2017). Moreover, similar changes in muscle size and strength have also been reported using both training methods. Although there is strong evidence suggesting the effects of muscle swelling on muscle physiology and on the positive adaptations to LLRE+BFR , more research is needed in order to determine the underlying mechanisms.

Perceptual Responses

Although LLRE+BFR appears to be a possible training alternative to traditional HL-RE because of the lower mechanical stress imposed to the musculature and joints, less attention has been given to the perceptual responses to this training modality. The perceptual responses to an exercise intervention may affect participant motivation and adherence to any training program (Van Roie et al., 2015). Therefore, the short- and long-term effects of LLRE+BFR on the perceptual responses of practitioners to the BFR stimuli is of great importance. The perceptual responses have been generally considered in terms of levels of pain and ratings of perceived exertion.

The results from studies comparing the perceptual responses to LLRE+BFR and HL-RE are conflicting. For instance, Loenneke et al. (2015) reported greater levels of pain during LLRE+BFR in comparison to HL-RE; whereas, Lixandrão et al. (2018) reported lower levels of pain and perceived exertion during LLRE+BFR. The discrepancy in these results may be due to the differences in the designs of these studies. Loenneke et al. (2015) had participants exercising to failure while Lixandrão et al. (2018) used 4 fixed sets of 15 repetitions. A recent study by Sieljacks et al. (2018) investigated the perceptual responses (ratings of effort and pain) and neuromuscular adaptations (muscle size and strength) over the course of 8 weeks of LLRE+BFR (25% of 1-RM), performed to failure or non-failure. Significant increases in both muscle size and strength were observed; however, greater perceptual responses were observed with LLRE+BFR performed to failure compared to the non-failure exercise condition. Other factors such as the amount pressure applied during exercise may influence the perceptual responses to LLRE+BFR. In another study, Mattocks et al. (2017) observed that LLRE+BFR tended to elicit greater

ratings of pain and effort at higher restrictive pressure, despite lower total volumes of exercise being completed.

Therefore, there is a need to better design a LLRE+BFR protocol that can be tolerated and still able to produce physiological benefits to participants, especially those with physical limitations such as elderly, injured, and participants with clinical conditions such as MS. In this regard, Korakakis, Whiteley, and Giakas (2018) demonstrated that physical therapy combined with BFR was able significantly to reduce anterior knee pain to a greater extent than physical therapy alone.

Implications and Safety of Resistance Exercise combined with Combined for People Living with MS

No study has yet investigated the physiological responses of individuals living with MS to LLRE+BFR. Therefore, the precise risks and benefits of this training modality for clinical population are unknown. However, based on the results from previous studies, it is possible to speculate some of the possible effects of LLRE+BFR on MS patients.

Muscle Damage

Muscle soreness and damage are typical responses commonly observed over days following a single bout of traditional resistance exercise. Elevated exercise-induced muscle damage may be prejudicial, specially to those living with MS, because it requires more recovery time between sessions, which may limit training frequency. Often, exercise-induced muscle damage is accompanied by muscle soreness and the initiation of

an anti-inflammatory response within the muscle, which may be contra-indicated for those suffering from MS.

Although there is not a consensus in the literature whether LLRE+BFR causes muscle damage, most of the studies indicate that LLRE+BFR does not lead to significant muscle damage. For instance, Loenneke et al. (2013) observed that low-intensity resistance exercise with or without BFR did not result in significant decrements in torque up to 24 hours post-exercise. Although significant decreases in torque values were observed at 1 hour post-exercise, it was most likely due to fatigue rather than actual muscle damage. It is important to highlight that the exercise-induced muscle damage commonly reported with high-intensity exercise is predominantly due to the eccentric phase of the contraction. With this in mind, Thiebaud et al. (2014) submitted participants to LLRE+BFR at 30% of 1RM with only eccentric actions being performed. The results from this study were similar to those from Loenneke et al. (2013) in which torque decrements were observed only 1 h post-exercise. Additionally, there were no significant changes in muscle soreness, muscle thickness, limb circumference, or range of motion up to 4 days following exercise. These results were further corroborated by another study by the same research group (Thiebaud et al., 2013). In contrast, Sieljacks et al. (2015) observed significant muscle damage, measured as decreased torque, increased soreness, water retention, and plasma concentrations of muscle proteins, after a single bout of LLRE+BFR at 30% of 1-RM to failure. The discrepancies observed across these and other studies may be due to differences in their methodological designs. For example, Sieljacks et al. (2015) had participants exercising to failure, not commonly incorporated in BFR

exercise protocols, while Thiebaud et al. (2014) and Loenneke et al. (2013) used fixed sets of 30+15+15+15 repetitions, which is a protocol most often reported in the literature.

Finally, no study has yet compared the amount of muscle damage resulting from a single bout of $_{LL}RE+BFR$ to a bout of traditional resistance exercise. Therefore, even if $_{LL}RE+BFR$ does result in muscle damage, it probably occurs at a much lower extent compared to traditional resistance exercise. Hence, BFR resistance exercise stands out as a safer and more tolerable training method capable of increasing muscle size and strength in individuals with MS, without the concerns normally associated with high-load training programs.

Inflammation

Local inflammation is another often observed physiological response to traditional resistance exercise. Inflammation occurs as a result of muscle damage as the body works to repair the damaged evoked by the exercise. MS sclerosis is characterized as a chronic inflammatory disorder that causes demyelination of the neurons inside the central nervous system. Since MS is characterized by this increased chronic inflammation, exercise interventions that may potentially cause more inflammation should be avoided.

Regarding the potential of $_{LL}RE+BFR$ to induce inflammation, Karabulut et al. (2013) compared the effects of 6 weeks of $_{LL}RE+BFR$ (20% of 1-RM) to traditional high-load (80% of 1-RM) resistance training, performed 3 times a week with interleukin-6 being used as a marker of inflammation in older men. The authors observed that neither training regimen elicited significant increases in plasma levels of interleukin-6. Similarly,

in a recent study, Bugera et al. (2018) investigated the acute effects of performed (30+15+15+15 repetitions at 30% of 1-RM) and traditional high-load resistance exercise (4 sets of 7 repetitions at 80% of 1-RM) on interleukin-6 and interleukin-15 immediately post-, 1 h post-, and 24 h post-exercise. No significant changes were observed in any of the inflammation markers after any of the tested exercise interventions. Clark et al. (2011) also did not observe any changes in C-reactive protein, used as an inflammation marker, either immediately after a single bout of exercise or after 4 weeks of both high-intensity (3 sets of 10 repetitions at 80% of 1RM) and LLRE+BFR (3 sets of 10 repetitions at 30% of 1RM). In regard to special populations, a pilot study using ischemic heart disease patients reported no inflammatory response (measured as C-reactive protein) after an acute session of LLRE+BFR (Haruhiko Madarame et al., 2013). To note, only one study reported increased inflammatory response after LLRE+BFR (Takarada, Nakamura, et al., 2000). However, it was not clear if the increased plasma levels of interleukin-6 were due to an actual inflammatory response or to the exercise energy demand, since it has been speculated that interleukin-6 may play a role in exercise metabolism (Pedersen, 2012).

Oxidative Stress

Increased oxidative stress is another common physiological response to resistance exercise (Hudson et al., 2008). Oxidative stress involves the formation of reactive oxygen species that can cause cell damage by reacting with proteins, lipids, and even DNA within the cell. For this reason, oxidative stress is also considered a risk and an undesired physiological event. Therefore, an elevated oxidative response to exercise would impose major risks for MS patients by further increasing damage to their musculature, which is

already markedly frail due to the disease itself. However, it has been suggested that some of reactive oxygen species produced during exercise can actually serve as signaling molecules driving some of the exercise adaptations (Powers et al., 2010). Therefore, a non-exacerbated oxidative response to exercise may not impose major issues.

Oxidative stress occurs primarily under conditions that require fast oxygen consumption, overloading the electron transport chain, and under low oxygen availability, with the latter corresponding to what happens during LLRE+BFR. The application of restrictive cuffs commonly results in diminished arterial inflow and occlusion of venous outflow, which forces the muscle to operate in a low-oxygen environment. LLRE+BFR may further induce oxidative stress by ischemia/reperfusion post cuff deflation. These characteristics of LLRE+BFR have concerned scientists regarding the oxidative response to this method of training. Takarada et al. (2000) provided the first evidence that LLRE+BFR does not induce significant oxidative stress. Goldfarb et al. (2008) also reported no significant oxidative response after 3 sets to failure of LLRE+BFR (30% of 1RM). Curiously, the authors reported significant oxidative stress immediately post and 15 min post high-intensity resistance exercise as well as after 5 min of BFR application in the absence of exercise. Therefore, the resistance exercise combined with BFR seemed to attenuate the oxidative stress. Additionally, Garten et al. (2015) confirmed this ability of LLRE+BFR to attenuate oxidative stress post-exercise. The authors reported lower protein carbonyl concentrations after a single bout of low-load resistance exercise to failure (30% of 1-RM) with BFR or without BFR compared to high-intensity resistance exercise to failure (80% of 1-RM) with or without BFR, and also after BFR application in the absence of exercise. More research is needed in order to elucidate how LLRE+BFR

actually attenuates oxidative stress, however, there is strong evidence suggesting that LLRE+BFR does not induce significant oxidative stress.

Resistance Training for Multiple Sclerosis Patients

Several studies have demonstrated the ability of progressive resistance training to enhance several fitness parameters in people living with MS, including muscle size, strength, physical function, perceived fatigue, and others. Moreover, these results have been shown to ultimately result in improved quality of life.

White et al. (2004), had 8 individuals diagnosed with MS complete 8 weeks of resistance training twice-weekly consisting of knee flexion and extension, plantarflexion, and spinal flexion and extension, at intensities ranging from 50% to 70% of maximal voluntary contractions. Although 8 weeks of resistance training did not elicit any changes in muscle cross-sectional area, there were significant improvements in several functional parameters, including strength gains (7.4% to 52%) and stepping performance (8.7%), as well as a decrease in the self-reported fatigue (from 32 to 26). In another study, Dalgas et al. (2009) had 19 individuals diagnosed with relapsing-remitting complete 12 weeks of progressive resistance training for the lower-body (leg press, knee extension, hip flexion, hamstrings curl, and hip extension), twice a week, with the number of sets and repetitions ranging from 3 to 4 and 8 to 12, respectively. Maximum isometric strength for the knee extensors and flexors, 1-RM strength, and functional capacity were measured before and after training. Following the 12-week training program, there were significant increases in all strength parameters ($\approx 16\%$ to 37%) and in functional capacity (21.5%) in the training group over the non-exercising control. The same training program has also been

shown to induce muscle fiber hypertrophy of the type II muscle fiber in MS patients (Dalgas et al., 2010). Furthermore, another study from the same research group demonstrated that this progressive resistance training program was also effective at improving fatigue, mood, and quality of life in people living with MS (Dalgas et al., 2010).

Additional studies have confirmed the ability of resistance training to improve muscle size, strength, power and physical function in individuals with MS (Broekmans et al., 2011; de Souza-Teixeira et al., 2009; Dodd et al., 2011). Interestingly, Dodd et al. (2006) performed a qualitative analysis to identify the self-reported positive and negative effects of progressive resistance training. The sample consisted of 8 participants that completed 10 weeks of resistance training performed twice a week. The participants cited improvements in many physical (e.g., strength, endurance, function, less fatigue, etc.), psychosocial (e.g., confidence, mood, etc.), and social (e.g., friendship, encouragement, and others) parameters as positive training outcomes. However, participants also identified muscle soreness, during and after exercise, as a negative short-term effect of resistance training. This highlights the importance of developing new resistance training interventions capable of eliciting positive long-term adaptations, while resulting in less mechanical stress and muscle damage, and, consequently, diminish muscle soreness.

Summary of Review of Literature

In summary, the above review of the literature provides scientific background demonstrating that resistance exercise in combination with BFR has the potential of serving as a clinically relevant non-pharmacological tool to help improve physical fitness

and, consequently, enhance the quality of life of those suffering from MS. Besides being useful at potentially attenuating the effects of MS, LLRE+BFR also imposes low risks in terms of inducing muscle damage, inflammation, oxidative stress, mechanical stress.

The primary mechanisms discussed herein thought to drive the positive adaptations following LLRE+BFR include the exercise-induced metabolic stress, muscle activation, muscle swelling, hormonal responses, and the regulation of biomolecular pathways. However, additional research is needed to confirm if this is also applicable in the context of MS research.

Despite the strong evidence presented above supporting the hypothesis that LLRE-BFR may benefit individuals with MS, there is surprisingly still no studies that were conducted to test that hypothesis. Thus, there is a critical need for studies to investigate the safety of this training method in this specific population as well as to test if individuals with MS can tolerate performing such a training protocol. Then, additional research is needed to prove whether this training modality may potentially result in long-term positive adaptations, such as increased muscle hypertrophy, strength, and physical function.

Chapter III: Methodology

Participants

Originally, five men and fifteen women with a physician confirmed diagnosis of relapsing-remitting MS volunteered for the current study, however, three people were not included for exhibiting higher physical activity levels, which included currently performing high-load resistance exercise, one person was removed for getting injured for reasons unrelated to the study, and one person requested to be withdrawn from the study. Therefore, the study included a total sample size of 15 participants (males: $n = 4$; females: $n = 11$) with a confirmed diagnosis of relapsing-remitting MS, aged 18 to 64 years from the Multiple Sclerosis Oklahoma Medical Research Foundation (OMRF) located in Oklahoma City, OK. Sample size was established based on a power analysis using previous data from our laboratory collected from healthy individuals. According to this analysis, 8 participants would be adequate to reach a statistical power of at least 0.80 (Beck, 2013).

Inclusion Criteria

1. Relapsing-remitting MS diagnosis confirmed by a neurologist.
2. A disability score ≤ 6.5 on the EDSS scale (Kurtzke, 1983).
3. Age between 18 and 64 years.
4. Non-pregnant women.
5. Not resistance trained for the past 6 months.

6. Normotensive or controlled hypertension (arterial brachial blood pressure \leq 140/90 mm Hg).
7. Ankle brachial index between 0.9 and 1.2.

Exclusion Criteria

1. Exacerbation of the disease symptoms during the period of the study.
2. The occurrence of any injuries that could limit the performance of the exercise trials or strength tests included in the study.
3. Failure to follow specific guidelines and instructions.
4. A direct request from participant to be withdrawn from the study.

Experimental Design

This study consisted of a randomized, within-within subjects crossover design that required participants to complete a total of five visits to the Neuromuscular Laboratory. During the first visit, participants were informed about the study protocols and experimental procedures and provided written informed consent before any testing was initiated. Participants also filled out and signed standardized questionnaires. Then, participants completed a familiarization session for the 1-RM test. During visit two, participants rested for five to ten minutes before completing the measurements of brachial arterial blood pressure, ankle-brachial index, and total arterial occlusion pressure for both legs, specifically in this order. Participants then completed the 1-RM test for the horizontal two-leg leg press and bilateral knee extension exercises. At visit three, participants' total body composition and bone mineral density were measured using DXA

scans, with one additional DXA scan at the spine and two at the hip (left and right side). Then, participants completed a second 1-RM test for the same two-leg press and bilateral knee extension exercises. Lastly, participants were familiarized with the sensation of performing resistance exercise while wearing the restrictive cuffs. During visits four and five, participants randomly completed the two experimental exercise trials (LLBFR-RE and HL-RE). There was a minimum three-day interval between visits two, three, and four and a 14-day period between visits four and five.

For each exercise trial visit, participants arrived at the laboratory between 6:00 AM and 8:00 AM, in a fasted state of at least eight hours. Participants, then, consumed a light breakfast consisting of yogurt, fruits, cereal, and orange juice, and rested for 15 minutes in the seated position. After resting, the baseline measures of muscle thickness and thigh circumference were taken, followed by a nurse obtaining a sample of venous blood. Blood samples were used to measure whole blood lactate, cortisol, and interleukin-6, myostatin, and mTOR concentrations. Participants then lifted a load equivalent to their highest 1-RM, previously assessed at visits two and three, which was also used to determine the loads to be lifted during each exercise trial and for superficial electromyography normalization. After that, the exercise session assigned for that day was initiated. Myoelectrical activity was continuously measured during exercise in the vastus medialis and vastus lateralis muscles of both legs. Ratings of pain were measured before and immediately after each set of leg press and knee extension as well as 5 min, 30 min, 60 min, and 24 hours post-exercise. Ratings of perceived exertion were measured immediately after each set of both exercises. Muscle thickness and thigh circumference

were re-assessed immediately post-, 15 min post-, and 60 min post-exercise. Two additional blood samples were taken 5 min post- and 60 min post-exercise.

Forms and Questionnaires

Participants were requested to fill out and sign all the following documents before any testing was carried out:

1. Consent form: To ensure voluntary participation in the study.
2. Health insurance portability and accountability act: To provide authorization for collection and usage of health-related information.
3. Physical activity readiness questionnaire: To ensure that participants were safe to perform exercise (Shephard, 1988).
4. International physical activity questionnaire: To collect information related to the participants' physical activity levels (Hagströmer et al., 2006).
5. Bone-specific physical activity questionnaire: To obtain information related to bone loading physical activity (Weeks & Beck, 2008).
6. Self-administered Kurtzke Expanded Disability Status Scale (EDSS): To quantify the levels of disability of each participant (Kurtzke, 1983).
7. Menstrual history questionnaire: To acquire information related to the regularity of the female participants' menstrual cycle and hormonal replacement therapy history.
8. Medical history questionnaire: to guarantee that participants met the inclusion criteria for this study and did not have any other diseases that would be negatively impacted by the study procedures or that could interfere with the study outcomes.

Arterial Brachial Blood Pressure

Arterial brachial blood pressure was measured using a portable automatic monitor (BP710, OMRON, IL) placed on the left arm and with participants lying down in the supine position. Before the measurement, participants rested in a supine position for 5 to 10 minutes in a quiet room. Measurements were taken in duplicate and the average was used in further analysis.

Ankle-Brachial Index

Ankle-brachial index (ABI) was measured following the measurement of blood pressure with participants lying down in the same position. A pneumatic inflatable cuff was manually inflated and used to measure the systolic blood pressure on both arms and ankles with the help of a handheld doppler placed on the radial and posterior tibial arteries, respectively. ABI was calculated as a ratio of the systolic blood pressure measured in the arms over the systolic blood pressure measured in the ankles.

Total Arterial Occlusion Pressure

Following the ABI measurement, the total amount of pressure required to totally occlude the arterial blood flow to each leg was measured with participants also lying down in the supine position. A 13.5 cm wide nylon cuff (SC12, D.E. Hokanson, Bellevue, WA, USA) connected to a rapid cuff inflator system (E20 Rapid Cuff Inflator, D. E. Hokanson, Bellevue, WA) was placed at the most proximal portion of the thigh and used to occlude arterial blood flow. A handheld Doppler probe (MD6 Doppler, D. E. Hokanson, Bellevue, WA, USA) coated with transmission gel was placed over the

posterior tibial artery and used to detect the auscultatory pulse. The cuff was first inflated to 50 mm Hg for approximately 20 seconds, deflated, and, then, re-inflated to the participant's systolic blood pressure. From this point, the cuff was deflated and re-inflated in increments of 10 mm Hg until the auscultatory pulse was interrupted. Then, the cuff was slowly deflated until the pulse was re-detected by the Doppler. This procedure was repeated in the contralateral limb. The pressure displayed immediately before the pulse was re-detected was considered the total arterial occlusion pressure was used to calculate the 50% BFR pressure to be applied during exercise, as the average of the two legs.

Standing Height and Body Mass

Standing height and body mass was measured and used to calculate body mass index (BMI). Standing height was measured to the nearest 0.5 cm using a calibrated stadiometer (Stadi-O-Meter, Novel Products, Rockton, IL) attached to the wall. Participants were asked to stand straight with their body aligned to the stadiometer and with heels, back, and head touching the wall. Standing height was measured after participants inspired as much air as possible and held their breath for a few seconds. Body mass was measured to the nearest 0.1 kg using a calibrated digital scale (BWB-800A, TANITA, Japan). Participants were wearing as minimal amounts of clothing and as possible, free from accessories such as watches and necklaces, and with empty pockets. Body mass was measured with participants standing immobile on the scale for about 3 seconds. BMI was calculated as body mass (kg) divided by the squared root of standing height (m).

Body Composition and Bone Mineral Density

Dual-energy X-ray absorptiometry (GE Lunar Prodigy DXA, GE Healthcare, Madison, NI) scans were used to assess body composition and bone mineral density. A total of 4 scans were performed: total body, lumbar spine (from L1 to L4), and dual proximal femur (femoral neck, trochanter, and total hip). Body composition was measured for the whole body presented as bone-free lean body mass (BFLBM), fat mass (FM), and bone mineral content (BMC). All scans were analyzed using specific software (enCORE 16, Healthcare, WI). Quality assurance tests were performed at each testing day for calibration and to ensure that the device was working properly. Before each scan, participants were asked to remove shoes and any metal accessories (e.g., earrings, necklace, piercing, etc.) and to wear minimal clothing. During the scans, participants lied down in the supine position, with arms and legs straight, and head positioned 2 to 3 cm below the horizontal line at the top of the measuring table. Hips and shoulders were evenly spaced in the center of the table with arms positioned parallelly to the body without touching it. For the total body scan, straps were wrapped around the knees and ankles and were used to prevent movements and to keep the legs straight during the scan. Following the total body scan, a foam block was placed under both legs and knees, which were bent at 45 to 60 degrees. Participants were asked to maintain their hips and upper body straight, and to point out their navel so that the scan arm could be adjusted to 2 finger widths below the navel, and then to hold their arms upright while the lumbar spine is scanned. Upon lumbar spine completion, the foam block was removed, and were placed on each side of the foot brace using straps. During the hip scans, the leg being measured was kept straight during assessment. The same procedure was repeated in the contralateral limb. Radiation

exposure ranged from 0.08 to 0.18 mrem per scan for each participant. All scans and follow-up analyses were performed by the same technician. The coefficients of variation for the DXA scans in the bone lab range between 1.2 – 1.7% for the total body scans, 1.3 - 1.8% for the dual hip, and 1.8% for the lumbar spine.

One-Repetition Maximum Test

Participants completed bilateral one-repetition maximum (1-RM) tests for the horizontal leg press and knee extension exercises (Cybex International Inc., Medway, MA, USA), which were used to determine the loads to be lifted during each experimental trial. The tests were performed during visits two and three and followed guidelines from the National Strength and Conditioning Association (Baechle & Earle, 2016). The 1-RM test represented the maximum amount of weight that could be lifted in a single attempt through a full range of motion. Before starting the test, participants were introduced to proper technique and performed an initial warmup with a load that easily allowed the completion of 8 to 10 repetitions; then, the weight was increased, and participants completed 4 to 5 repetitions; next, the weight was increased again, and participants performed 2 to 3 repetitions. Following the warmups, the weight was progressively increased until the participant was no longer able to complete a repetition with proper form through a full range of motion. Participants were given 2 to 4 min to rest between warmups and between each maximal attempt. The 1-RM was considered the last load lifted with proper form through a full range of motion. The 1-RM for each participant was obtained within 3 to 5 attempts. There was a minimum rest period of 3 minutes between the 1-RM test for the leg press and the knee extension exercises. The same

trained technician administered all tests for each participant. Reliability estimates for the 1-RM tests are presented in the results section.

Surface Electromyography

Myoelectric activity was measured using surface electromyography (sEMG) and was represented as root mean square (RMS). Data acquisition was carried out using an amplifier system (MP-100, BIOPAC systems Inc, CA) and superficial bipolar electrodes (EL503, BIOPAC systems Inc, CA), placed over the vastus medialis and vastus lateralis muscles of both legs with an inter-electrodes distance of 2 cm. Electrodes' placement followed the recommendations from the SENIAM project (surface ElectroMyoGraphy for the Non-Invasive Assessment of Muscles). The skin was shaved, abraded, and whipped with alcohol prior to electrode placement. Although the intent was electromyography signal to be sampled at a rate of 2000 Hz, due to technical error, the signal was collected at 200 Hz. However, a similar procedure has previously been reported in a study investigating myoelectric activity in individuals with MS (Dalgas et al., 2013). The signal was full-wave rectified and a low pass 4th order Butterworth filter with a 6 Hz cut-off frequency was used prior to the calculation of the root mean squares. Prior to the start of each exercise bout, participants lifted their 1-RM load while sEMG was recorded. This recording was used for signal normalization. Additionally, participants performed 3 repetitions at 70% of their 1RM with 50% of BFR before the $LLRE+BFR$ exercise protocol. The same number of repetitions was performed at 20% of 1-RM without BFR before the HI-RE exercise trial. This procedure was used to investigate the impact of BFR on the sEMG signal. sEMG was also recorded continuously

at each set during exercise and stored in a personal computer. The signal was analyzed at the end of the study using specialized software (Acknowledge 3.9.1, BIOPAC systems Inc, CA) using the concentric and eccentric contractions for all the 4 sets of both exercises for each experimental condition.

Whole-blood Lactate

Whole-blood lactate (WBL) was measured in mmol/L using a portable lactate analyzer (Lactate Plus, Nova Biomedical Corporation, Waltham, MA, USA). Intravenous blood samples of about 0.7 μ L were collected after a 5-min rest period at baseline and 5 min and 60 min post-exercise. The portable lactate analyzer was calibrated at least once a day before data collection using low (1.0 to 1.6 mmol/L) and high (4.0 to 5.4 mmol/L) control solutions (Lactate Plus, Nova Biomedical Corporation, Waltham, MA, USA). All analyses were performed in duplicate and used to calculate intra- and inter-day intraclass correlation coefficients (ICCs) and minimal differences needed to be considered a real change (MD). The inter- and intra-tests ICCs for WBL measurements were 0.985 and 0.994, respectively.

Muscle Thickness

Muscle thickness was measured to the nearest 0.1 cm using a B-mode ultrasound device and a 5 MHz linear probe (UF-750XT, Fukuda Denshi, Japan) at the 50% anterior portion of both thighs, before, immediately post-, 30 min post-, and 60 minutes post-exercise. The measurements were performed at the 50% portion of the thigh, which consisted of the distance from the greater trochanter to the lateral condyle of the femur.

This site was marked with semi-permanent ink to ensure consistency of the measurements between visits. Transmission gel was placed over the linear probe, which was positioned perpendicularly to the skin interface without causing any depression. Muscle thickness was considered as the distance from the subcutaneous adipose tissue-muscle interface to the muscle-bone interface, measured in a straight line. All measurements were performed with participants seating down, feet positioned shoulder width apart, arms straight. All measures were obtained by the same trained technician. The ICCs between visits for muscle thickness measured in the right and left legs were 0.925 and 0.972, respectively.

Thigh Circumference

Thigh circumference was measured to the nearest 0.1 cm using a tape measure at the same time points and in the same sites used to measure muscle thickness. Thigh circumference was measured with the participant seating down, feet positioned shoulder width apart, and arms straight. All measures obtained by the same trained technician. The ICCs between visits for thigh circumference measured in the right and left legs were 0.980 and 0.991, respectively

Hematocrit and Plasma Volume Change

Hematocrit was measured at baseline, 5 min post-, and 60 min post-exercise using venous blood samples (approximately 6 μ L in volume) collected from the antecubital vein and transferred to microhematocrit heparinized capillary tubes. The blood was allowed to rest at room temperature for approximately 5 min and then it was centrifuged

for 2 min at 16,000 rpm (StatSpin, Norwood, MA). The reading was performed using a manual reader plate and all measurements were performed in duplicate. Percent plasma volume change will be calculated using the following equation (Van Beaumont, 1972):

$$\%PV\Delta = \frac{100}{(100 - HCT_{pre})} \times 100 \left(\frac{HCT_{pre} - HCT_{post}}{HCT_{post}} \right)$$

Then, the concentration of the blood markers measured in this study were corrected for the changes in plasma volume using individual values and using the following equation:

$$Corrected_{conc} = Uncorrected_{conc} \times \left(\frac{100 + \%PV\Delta}{100} \right)$$

Blood Handling and Assays

Venipunctures were performed to collect blood samples of approximately 7.5 mL by a certified nurse. Following each blood draw, the blood was allowed to rest and clot at room temperature for about 30 min. Then, it was centrifuged at 2,000 G for 15 min and serum was separated, pipetted into ½ mL aliquots, and frozen at -80 °C until all assays were performed at the end of the study. All assays for mammalian target of rapamycin complex 1 (mTOR), myostatin, interleukin-6 (IL-6), and cortisol were performed using specialized kits following manufacturers' instructions (Appendix D). In summary, the procedures were initiated by letting the serum samples and the assay kit rest for approximately an hour until reaching room temperature. Then, the standards would be diluted following the instructions from each assay kit, and 100 microliters would be transferred to the first wells of the microtiter plate, followed by the transfer of approximately 50 microliters of serum samples to the remaining wells. Blank wells would

be used if determined by the manufacture. The remaining steps included the addition of reagents, that varied according to the ELISA kits being used, and rounds of incubation between 60 and 30 min. Finally, the microtiter plate was transferred to spectrophotometer device and read at 450 nm. The intra and inter-assay CVs for the mTOR, myostatin, IL-6, and cortisol assays were, respectively: 4% and 11.5%; 7% and 13%; 5% and 11%; and 3.5% and 10%. The ELISA kits were purchased from the following manufactures: USA R&D Systems (IL-6), DRG Instruments GmbH (Cortisol), MyBiosource (myostatin), and Bioassay Technology Laboratory (mTOR).

Ratings of Perceived Exertion

RPE was measured using the OMNI perceived exertion scale for resistance exercise (OMNI-RES) (Robertson et al., 2003), designed to measure effort immediately after each set of resistance exercise. In addition to numeric values linked to verbal anchors, the scale also includes figures to help participants rate their perceived exertion. The scale is divided into 11 categories from 0 to 10, as follows: 0 = extremely easy, 2 = easy, 4 = somewhat easy, 6 = somewhat hard, 8 = hard, and 10 = extremely hard. No verbal anchors are given in association with the numbers 2, 4, 6, and 8. The scale was carefully explained to the participants and they were familiarized with the scale during the exercise familiarization session at visit 2 and an anchoring procedure for the scale was performed during visits three. Participants were also reminded on how to properly use the scale prior to each experimental trial session.

Ratings of Pain

The ratings of pain and ratings of delayed onset muscle soreness were assessed using a visual verbal analog scale (Cook et al., 1997). This scale combines numeric values with verbal anchors and is divided into 12 categories from 0 to 10, as follows: 0 = no pain at all, 0.5 = very faint pain (just noticeable), 1 = weak pain, 2 = mild pain, 3 = moderate pain, 4 = somewhat strong pain, 5 = strong pain, 7 = very strong pain, 10 = extremely intense pain (almost unbearable). No verbal anchors were given in association with the numbers 6, 8, and 9. On the top of the scale, there was also a point (•) with the verbal anchor “unbearable pain”. Participants were shown the scale and asked to rate the amount of pain or pain that they felt in their legs before the start of each exercise bout (LP and KE) and immediately after each set of exercise. Participants were also asked to rate their levels of pain at 5 min, 30 min, and 60 min post-exercise. Moreover, participants were contacted via phone 24 hours following each exercise trial and asked to rate their levels of delayed-onset muscle soreness using the scale. The scale was carefully explained to the participants and they were familiarized with the scale during the exercise familiarization session at visit 3. Participants were reminded on how to properly use the scale prior to each experimental trial session.

Modified Fatigue Impact Scale

Participants were required to rate their levels of symptomatic fatigue using the Modified Fatigue Impact Scale (MFIS) (Multiple Sclerosis Council for Clinical Practice Guidelines, 1998) during each visit. The scale contains 21 items, including 9 physical items, 10 cognitive items, and 2 psychosocial items. The maximum score possible is 84,

with higher scores indicating greater fatigue. Previous studies have confirmed the reliability and validity of the MFIS (Flachenecker et al., 2002; Téllez et al., 2005). This scale was used to guarantee that participants will display similar levels of fatigue during both exercise trials. Therefore, participants' levels of fatigue during the last two trial sessions were considered different if a standard deviation greater than 2.5 was observed, calculated based on the score of the first 3 visits. In this case, the visit would be rescheduled for another day, in which the participants' fatigue levels would be reassessed. All participants' MFIS scores were within the 2.5 standard deviation limit and, thus, no visit had to be rescheduled.

Contraction Speed

An iOS-based metronome application (MetroTimer 4.6, ONYX 3) was used to control the speed of both the concentric and eccentric portion of the contraction during all exercise trials. The metronome was set at 40 bpm, which allowed 1.5 second for each portion of the contraction. Participants were familiarized with this pace during the exercise familiarization session and, during the actual experimental trials, participants received verbal encouragement to maintain the pre-determined contraction speed.

Resistance Exercise Protocols

Participants were required to randomly complete the following exercise conditions: low-load resistance exercise with BFR ($LLRE+BFR$) and high-load resistance exercise without BFR (HL-RE). The $LLRE+BFR$ condition consisted of 4 sets of 30+15+15+15 repetitions of both bilateral horizontal leg press and knee extension

exercises, performed at 20% of the individual's 1-RM, with a 1-minute rest interval between sets and 3-min between exercises, and at a metronome-controlled pace of 1.5 second for each portion of the contraction. Arterial blood flow to both legs were restricted by 50% of the total arterial occlusion pressure using a pair of 13.5 cm wide nylon cuffs (SC12, D.E. Hokanson, Bellevue, WA) connected to a rapid inflator device (E20 Rapid Cuff Inflator, D. E. Hokanson, Bellevue, WA) and placed at the most proximal portion of each thigh. The cuffs were inflated immediately before exercise and deflated following completion of the last set of leg press and knee extension exercises. Thus, cuffs remained inflated during the entire exercise period, including the between sets rest intervals, but were deflated during the 3-min interval between leg press and knee extension. For the HL-RE exercise condition, participants completed 4 sets of 8 to 10 repetitions of the same leg press and knee extension exercises, at 70% of 1-RM, with the same rest interval between sets and exercises, and at the same contraction speed. No BFR was applied during the HL-RE testing condition.

Statistical Analyses

Data Distribution

Descriptive and graphical information from histograms and Q-Q plots supplemented by the Shapiro Wilk test were used to determine data distribution. All data was analyzed using RStudio 3.6.1 (R Foundation for Statistical Computing, Vienna, Austria) and significance level was set at $p \leq 0.05$.

Parametric Data

Parametric data consisted of 1-RM strength values, whole-blood lactate, sEMG, muscle thickness, thigh circumference, and all blood markers. These variables were analyzed using two-way (condition x time) repeated measures analyses of variance to test all main effects and interactions. In the case of significant interactions, pairwise *t* tests were used to test the simple effects. Familywise error rate was controlled using the Bonferroni procedure. If the sphericity assumption was not met, the Greenhouse-Geisser correction was used.

Generalized eta-squared (η_G^2) was used as estimates of effect size for all main effects and interactions and was interpreted as follows: 0.02 as small, 0.13 as medium, and 0.26 as a large effect size (Cohen, 1988). Cohen's *d* was calculated as estimates of effect size for the pairwise comparisons, whenever deemed necessary. Intraclass correlation coefficients were calculated as test-retest reliability estimates for the 1-RM tests based on an absolute agreement, two-way mixed-effects model. The standard error of the measurement (SEM) was calculated as the squared root of the mean squared error and was used to calculate the minimum difference (MD) needed to be considered a real change ($MD = SEM \times 1.96 \times \sqrt{2}$) with a 95% confidence interval. All parametric data are presented as means \pm SD, unless stated otherwise.

Nonparametric data

Nonparametric data consisted of RPE, ratings of pain, and MFIS scores. Therefore, these variables were analyzed using nonparametric statistics. The Wilcoxon test was used to compare the two experimental conditions within specific time points. The Friedman's nonparametric test was used to test for significant differences in the

median rank scores across the time points. If a significant difference was detected, pairwise Wilcoxon nonparametric tests with Bonferroni procedure were used to locate the differences. Nonparametric data are presented as Winsorized means \pm Winsorized SD.

Chapter IV: Results and Discussion

Results Section

Descriptive Characteristics

Table 1 presents the descriptive characteristics of all participants included in the study. Out of the 11 female participants included, 3 were post-menopausal, 2 were not taking any oral contraceptives, and 5 were taking hormonal contraceptives (Skyla IUD, Mirena [2 participants], Lo Loestrin Fe, Trinessa, Vivelle-Dot).

Table 1. Participants' descriptive characteristics ($n = 15$).

Variable	Mean \pm SD	Minimum	Maximum
Expanded disability status scale (EDSS)	1.87 \pm 1.51	0.00	5.50
Age (years)	45.67 \pm 9.35	33.40	64.00
Standing height (cm)	170.03 \pm 7.06	155.00	182.50
Total body mass (kg)	91.74 \pm 19.63	61.40	120.70
Body mass index (kg/m ²)	31.91 \pm 7.18	18.83	40.10
Bone-free lean mass (kg)	49.24 \pm 7.11	38.83	60.12
Fat mass (kg)	39.84 \pm 14.64	15.10	63.11
Body fat (%)	43.33 \pm 8.97	25.71	54.00
Total body bone mineral content (kg)	2.66 \pm 0.39	1.90	3.34
Total body BMD (g/cm ²)	1.245 \pm 0.15	0.98	1.50
Spine region BMD (g/cm ²)	1.261 \pm 0.15	1.01	1.53
Total hip BMD (g/cm ²)	1.002 \pm 0.14	0.78	1.24
Femoral neck BMD (g/cm ²)	0.972 \pm 0.16	0.71	1.28
Trochanter BMD (g/cm ²)	0.800 \pm 0.11	0.61	0.96
Z-score for total body BMD	0.59 \pm 1.00	-1.2	2.00
Z-score for spine region BMD	0.16 \pm 0.94	-1.40	1.40
Z-score for total hip BMD	-0.39 \pm 0.76	-1.60	1.30
Z-score for femoral neck BMD	-0.38 \pm 0.92	-1.80	1.40
Z-score for trochanter BMD	-0.81 \pm 0.71	-2.00	0.10
T-score for total body BMD	1.31 \pm 1.39	-1.00	4.10
T-score for spine region BMD	0.77 \pm 1.20	-1.40	2.90
T-score for total hip BMD	-0.04 \pm 1.12	-1.80	1.90
T-score for femoral neck BMD	-0.51 \pm 1.17	-2.40	1.70
T-score for trochanter BMD	-0.44 \pm 0.98	-2.10	0.90
Left leg occlusion pressure (mmHg)	169.33 \pm 26.00	128	214
Right leg occlusion pressure (mmHg)	161.13 \pm 21.17	123.00	194

Regarding their levels of physical activity, 3 participants were classified as high, 3 as moderate, and 9 as low levels of physical activity (Table 2). Table 2 also presents data from the Bone Specific Physical Activity Questionnaire.

Table 2. Participants' physical activity characteristics ($n = 15$).

Variable	Mean \pm SD	Minimum	Maximum
Total physical activity MET	6447.70 \pm 6855.11	803	26898
Walk MET	2603.70 \pm 3848.42	0	11088
Moderate physical activity MET	3465.33 \pm 4322.05	110	13410
Vigorous Physical Activity MET	378.67 \pm 740.98	0	2400
Current BPAQ	0.22 \pm 0.63	0	2.35
Past BPAQ	277.66 \pm 268.55	66.36	1141.28
Total BPAQ	140.47 \pm 133.8	33.18	571.82

BPAQ: Bone Specific Physical Activity Questionnaire, MET: Metabolic equivalent.

Table 3 presents the exercise volumes achieved within each of the experimental exercise conditions, for the leg press and knee extension exercises.

Table 3. Exercise volume (kg) for each exercise condition during leg press and knee extension ($n = 15$).

	Leg press	Knee extension
LLRE+BFR	1727.35 \pm 433.43	790.12 \pm 203.15
HL-RE	3685.01 \pm 924.66**	1573.16 \pm 457.54**

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

**Significantly greater than LLRE+BFR at $p < 0.01$. Data are mean \pm SD.

Physiological responses

Whole-Blood Lactate

A significant condition \times time interaction ($F = 14.905, p = 0.001, \eta_G^2 = 0.15$) and significant condition ($F = 31.118, p < 0.001, \eta_G^2 = 0.16$) and time ($F = 53.046, p = 0.001, \eta_G^2 = 0.60$) main effects were observed for whole-blood lactate (Table 4 and Figure 1). Further analyses revealed that HL-RE resulted in significantly greater lactate levels 5 min post-exercise compared to the LLRE+BFR condition ($p < 0.001, d = 1.03$), with no significant differences between trials at baseline ($p = 0.11$) or 60 min ($p = 0.055$) post-exercise. Furthermore, for both conditions, whole-blood lactate was significantly elevated from baseline levels 5 min post-exercise ($p < 0.001$), however, it returned to pre-exercise levels 60 min post-exercise ($p = 1.00$).

Table 4. Absolute values for whole-blood lactate concentration (mmol/L) before and after each exercise condition ($n = 15$).

	Pre	5 min	60 min
LLRE+BFR	0.94 \pm 0.51	2.20 \pm 0.67 ^{$\alpha\beta$}	0.92 \pm 0.41
HL-RE	1.19 \pm 0.70	3.72 \pm 1.41 ^{$**\alpha\beta$}	1.08 \pm 0.42

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

^{**}Significantly greater than LLRE+BFR at $p < 0.01$, ^{α} Significantly different than pre at $p < 0.05$, ^{β} Significantly different than 60 min at $p < 0.05$. Data are mean \pm SD.

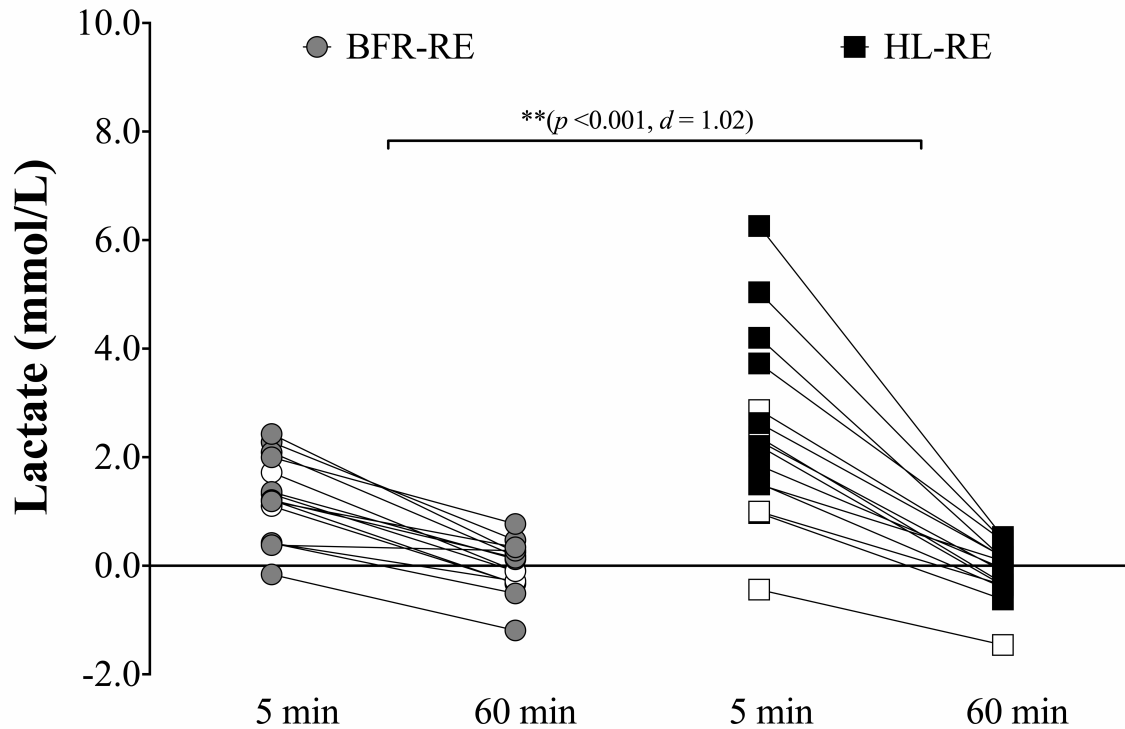


Figure 1. Individual absolute changes from baseline in whole-blood lactate concentration (mmol/L) from pre-exercise at 5 min and 60 min following each exercise condition (n = 15).

$_{LLRE+BFR}$: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

**Significantly greater than $_{LLRE+BFR}$ at $p < 0.05$.

Hematocrit

There was a significant condition \times time interaction ($F = 3.67$, $p < 0.039$, $\eta_G^2 = 0.01$) but no significant condition ($F = 0.02$, $p = 0.866$, $\eta_G^2 < 0.01$) nor time ($F = 0.13$, $p = 0.879$, $\eta_G^2 < 0.01$) main effects for the changes in hematocrit levels (Table 5). Further analysis of the condition \times time interaction using pairwise comparisons revealed that such effect does not actually exist ($p \geq 0.06$).

Table 5. Mean values for hematocrit concentration (%) before and after each exercise condition ($n = 14$).

	Rest	5 min	60 min
LLRE+BFR	43.04 ± 3.04	43.48 ± 2.92	42.71 ± 3.54
HL-RE	43.02 ± 2.92	43.66 ± 3.22	43.45 ± 3.17

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Data are mean ± SD.

Plasma Volume Change

There was not a significant condition × time interaction ($F = 3.67, p < 0.039, \eta_G^2 = 0.01$) nor significant condition ($F = 0.02, p = 0.866, \eta_G^2 < 0.01$) or time ($F = 0.13, p = 0.879, \eta_G^2 < 0.01$) main effects for the changes in plasma volume (Table 6).

Table 6. Mean values for plasma volume changes (%) following each exercise condition ($n = 14$).

	Rest	5 min	60 min
LLRE+BFR	-	-1.67 ± 5.37	1.59 ± 6.85
HL-RE	-	-2.41 ± 5.87	-1.45 ± 7.43

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Data are mean ± SD.

Cortisol

Data from only 13 participants were used for statistical analyses. One female was excluded for missing one blood draw and another female was removed due to exaggeratedly high cortisol levels during the HL-RE testing visit. The uncorrected cortisol concentration for this participant were 1369.65 ng/mL at rest, 2184.382 ng/mL 5 min post-exercise ($\Delta = 814.73$ ng/mL), and 3841.92 ng/mL 60 min post-exercise ($\Delta = 2472.71$ ng/mL).

Plasma cortisol concentrations were corrected for plasma volume changes and thus will be presented as corrected as well as uncorrected concentrations. As displayed

on Table 7 and Figure 2, there was a significant time main effect ($F = 5.61, p = 0.02, \eta_G^2 = 0.05$) but no significant condition main effect ($F = 0.02, p = 0.89, \eta_G^2 < 0.01$) or condition \times time interaction ($F = 2.40, p = 0.112, \eta_G^2 = 0.01$) for the uncorrected cortisol concentration. Pairwise comparisons revealed no significant changes in uncorrected cortisol concentrations from pre-exercise (169.56 ± 24.18 ng/mL) at 5 min post-exercise (152.32 ± 15.57 ng/mL, $p = 0.52$), but there was a significant decrease 60 min post-exercise (125.13 ± 18.94 ng/mL) compared to pre-exercise ($p < 0.01$) and 5 min post-exercise ($p = 0.02$) measures.

Similar results were observed for the corrected plasma cortisol concentrations with a significant time main effect ($F = 5.18, p = 0.029, \eta_G^2 = 0.06$) being detected, but no significant condition main effect ($F = 0.02, p = 0.893, \eta_G^2 < 0.01$) or condition \times time interaction ($F = 2.10, p = 0.143, \eta_G^2 = 0.01$). Follow-up analyses of the time main effect demonstrated no significant changes in corrected cortisol concentrations from pre (169.56 ± 24.18 ng/mL) compared to 5 min post-exercise (148.95 ± 18.65 ng/mL, $p > 0.37$), but there was a significant decrease 60 min post-exercise (123.10 ± 13.38 ng/mL) compared to pre ($p < 0.001$) and 5 min post-exercise ($p = 0.032$).

Table 7. Cortisol responses (ng/mL) before and after each exercise condition ($n = 13$).

<i>Uncorrected values</i>	Pre	5 min	60 min ^a
LLRE+BFR	168.74 \pm 25.44	161.53 \pm 20.20	115.00 \pm 12.98
HL-RE	170.38 \pm 25.78	143.12 \pm 19.56	135.26 \pm 18.93
<i>Corrected values</i>	Pre	5 min	60 min ^a
LLRE+BFR	168.74 \pm 25.44	159.12 \pm 20.47	115.45 \pm 12.44
HL-RE	170.38 \pm 25.78	138.78 \pm 18.55	130.76 \pm 15.41

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

^aSignificantly different than pre at $p < 0.05$. Data are mean \pm SE.

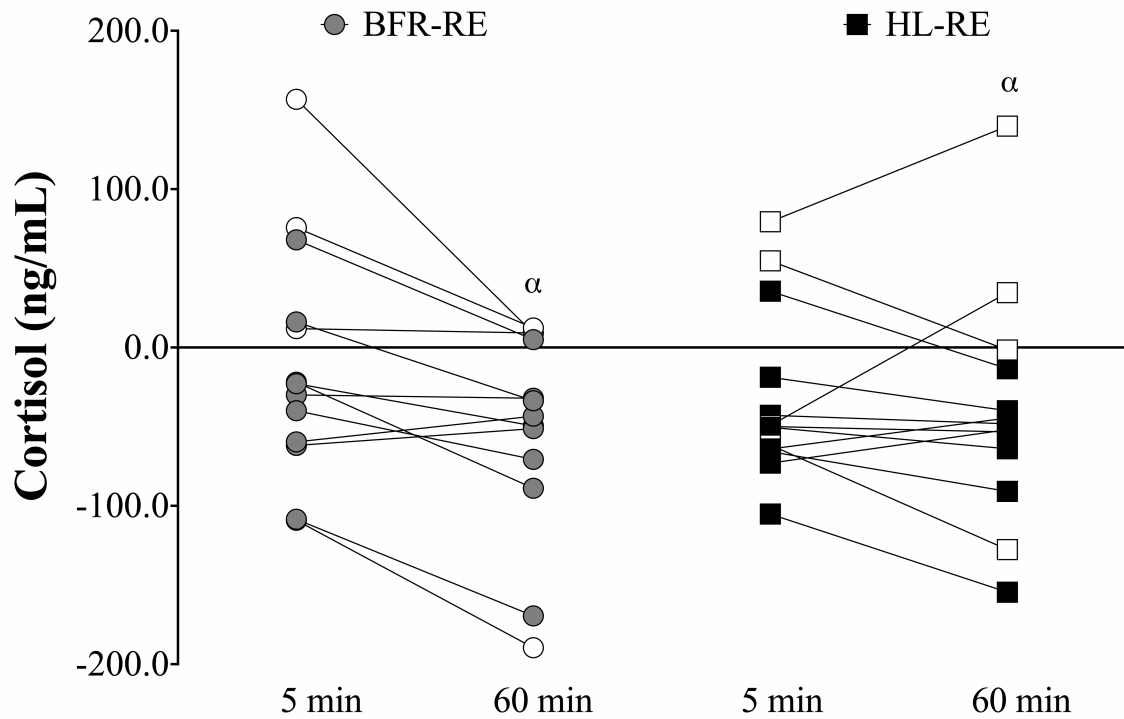


Figure 2. Individual absolute changes in corrected cortisol concentration (ng/mL) from pre-exercise at 5 min and 60 min following each exercise condition ($n = 13$).

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

^αSignificantly different than pre at $p < 0.05$.

Inflammation

As illustrated in Table 8 and Figure 3, there were no significant condition \times time interactions (uncorrected: $F = 0.24$, $p = 0.71$, $\eta_G^2 < 0.01$; corrected: $F = 0.49$, $p = 0.55$, $\eta_G^2 < 0.01$) nor condition (uncorrected: $F = 0.09$, $p = 0.77$, $\eta_G^2 < 0.01$; corrected: $F = 0.13$, $p = 0.72$, $\eta_G^2 < 0.01$) or time (uncorrected: $F = 0.79$, $p = 0.41$, $\eta_G^2 < 0.01$; corrected: $F = 1.1$, $p < 0.32$, $\eta_G^2 < 0.01$) main effects for either uncorrected or corrected serum IL-6 concentrations.

Table 8. IL-6 concentrations (pg/mL) before and after each exercise condition ($n = 14$).

<i>Uncorrected values</i>	Rest	5 min	60 min
LLRE+BFR	2.69 ± 0.64	2.66 ± 0.69	3.02 ± 0.75
HL-RE	2.70 ± 0.50	2.58 ± 0.51	2.80 ± 0.58
<i>Corrected values</i>	Rest	5 min	60 min
LLRE+BFR	2.69 ± 0.64	2.64 ± 0.70	3.14 ± 0.81
HL-RE	2.70 ± 0.50	2.57 ± 0.52	2.80 ± 0.59

BFR: blood flow restriction resistance exercise, HL-RE: high-load resistance exercise. Data are mean ± SE.

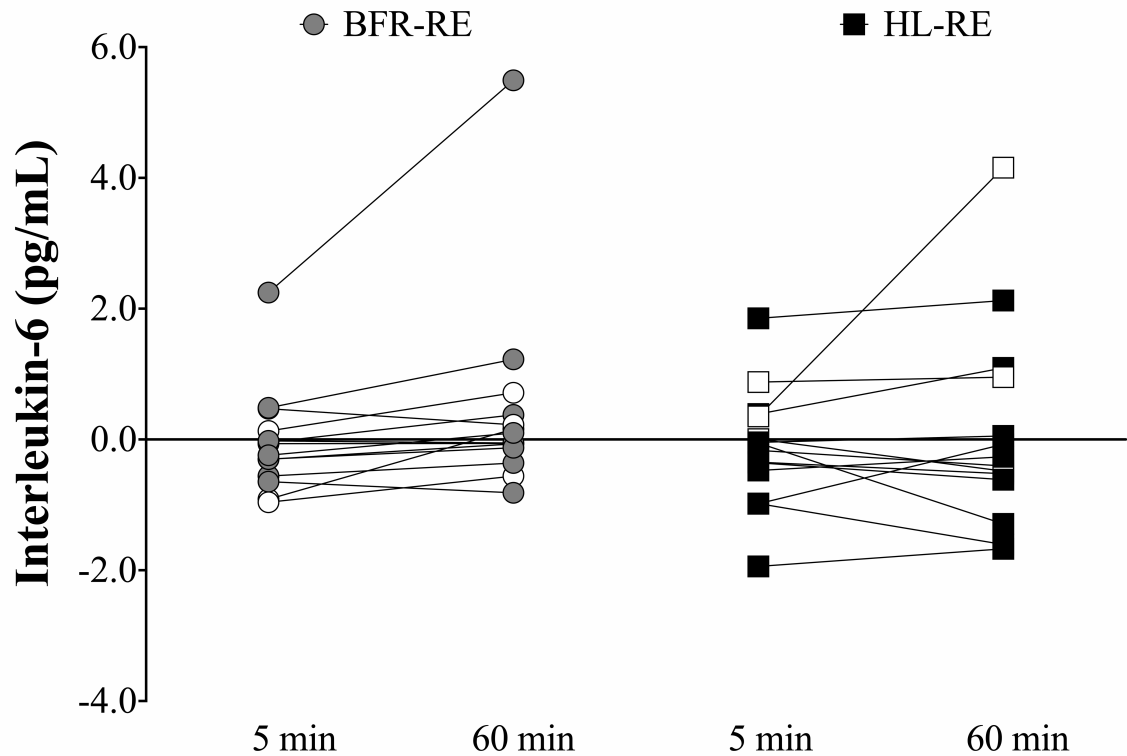


Figure 3. Individual absolute changes in interleukin-6 concentration (pg/mL) from pre-exercise at 5 min and 60 min following each exercise condition ($n = 13$).

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

Mammalian Target of Rapamycin Complex 1

There were no significant condition × time interactions (uncorrected: $F = 2.16$, $p = 0.14$, $\eta_G^2 < 0.01$; corrected: $F = 0.40$, $p = 0.11$, $\eta_G^2 < 0.01$) nor condition (uncorrected: $F = 0.04$, $p = 0.84$, $\eta_G^2 < 0.01$; corrected: $F = 0.29$, $p = 0.60$, $\eta_G^2 < 0.01$) or time

(uncorrected: $F = 0.25$, $p = 0.78$, $\eta_G^2 < 0.01$; corrected: $F = 0.29$, $p = 0.75$, $\eta_G^2 < 0.01$)

main effects for either uncorrected or corrected serum mTOR concentrations (Table 9 and Figure 4).

Table 9. Absolute values for mTOR concentration (pg/mL) before and after each exercise condition ($n = 14$).

<i>Uncorrected values</i>	Rest	5 min	60 min
LLRE+BFR	8.40 ± 1.54	7.82 ± 1.48	8.15 ± 1.51
HL-RE	7.99 ± 1.72	8.34 ± 1.59	7.89 ± 1.45
<i>Corrected values</i>	Rest	5 min	60 min
LLRE+BFR	8.40 ± 1.54	7.73 ± 1.48	8.48 ± 1.66
HL-RE	7.99 ± 1.72	8.30 ± 1.67	7.91 ± 1.55

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Data are mean ± SE.

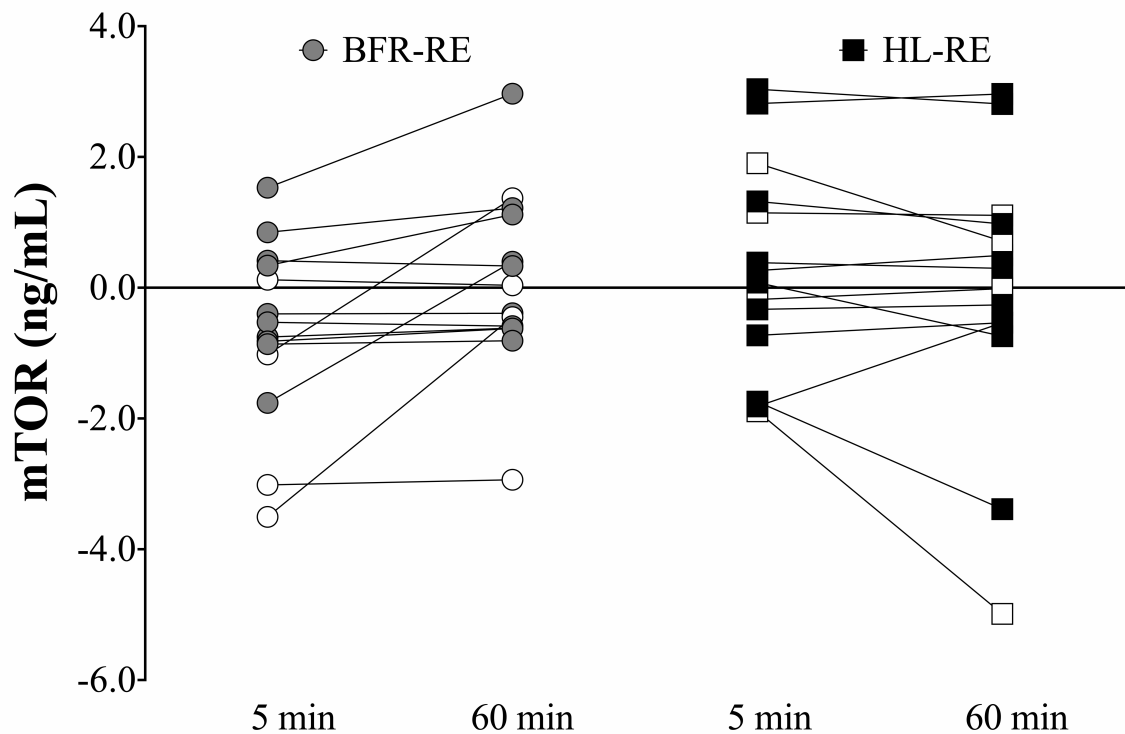


Figure 4. Individual absolute changes in corrected mTOR concentration (pg/mL) from pre-exercise at 5 min and 60 min following each exercise condition ($n = 13$).

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

Myostatin

There were no significant condition \times time interactions (uncorrected: $F = 1.89$, $p = 0.19$, $\eta_G^2 = 0.02$; corrected: $F = 1.75$, $p = 0.21$, $\eta_G^2 = 0.02$) nor condition (uncorrected: $F = 1.23$, $p = 0.29$, $\eta_G^2 < 0.01$; corrected: $F = 1.43$, $p = 0.25$, $\eta_G^2 = 0.01$) or time (uncorrected: $F = 0.63$, $p = 0.46$, $\eta_G^2 = 0.01$; corrected: $F = 1.03$, $p = 0.34$, $\eta_G^2 = 0.01$) main effects for either uncorrected or corrected serum myostatin concentrations (Table 10 and Figure 5).

Table 10. Mean values for myostatin concentration (pg/mL) before and after each exercise condition ($n = 14$).

<i>Uncorrected values</i>	Pre-exercise	5 min	60 min
LLRE+BFR	2.11 \pm 0.42	1.65 \pm 0.17	1.73 \pm 0.19
HL-RE	1.63 \pm 0.18	1.77 \pm 0.22	1.66 \pm 0.18
<i>Corrected values</i>	Pre-exercise	5 min	60 min
LLRE+BFR	2.11 \pm 0.42	1.62 \pm 0.18	1.69 \pm 0.81
HL-RE	1.63 \pm 0.18	1.70 \pm 0.22	1.58 \pm 0.19

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Data are mean \pm SE.

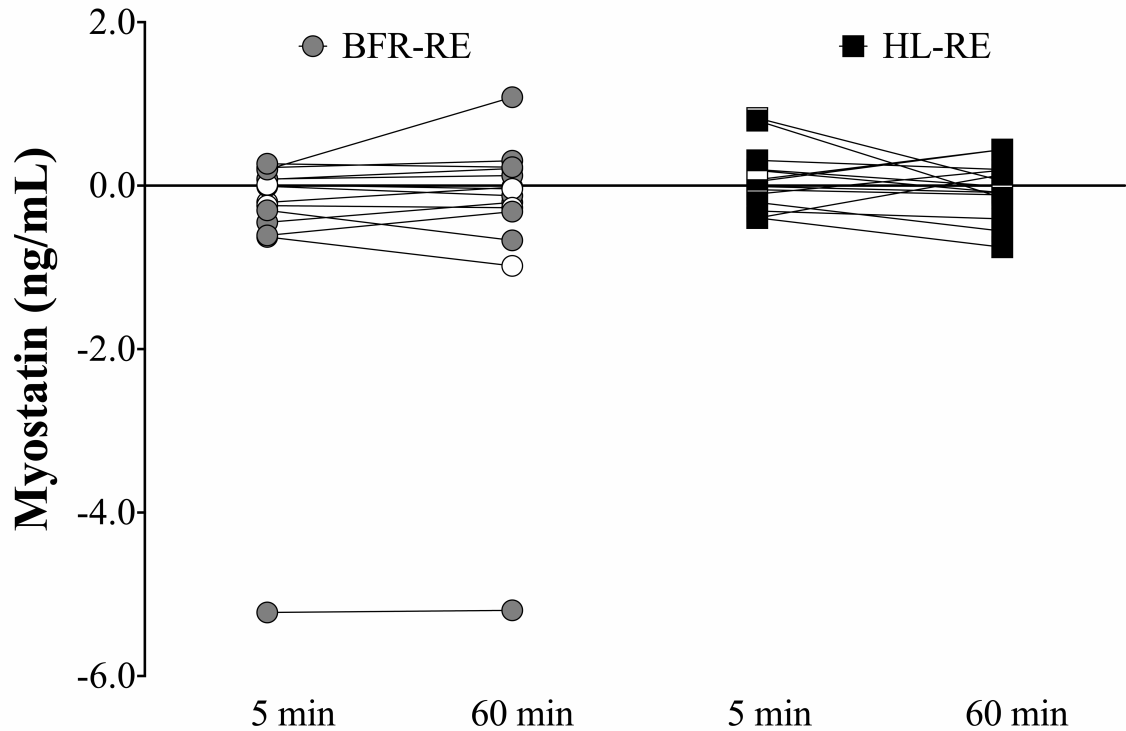


Figure 5. Individual absolute changes in corrected myostatin concentration (pg/mL) from pre-exercise at 5 min and 60 min following each exercise condition ($n = 13$). LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

Muscle Thickness

There was a significant time main effect (right leg: $F = 13.196$, $p < 0.001$, $\eta_G^2 = 0.03$; left leg: $F = 14.921$, $p < 0.001$, $\eta_G^2 = 0.624$), but no significant condition main effect (right leg: $F < 0.01$, $p = 0.98$, $\eta_G^2 < 0.01$; left leg: $F = 0.212$, $p = 0.65$, $\eta_G^2 = 0.023$) or condition \times time interaction (right leg: $F = 0.735$, $p = 0.50$, $\eta_G^2 < 0.01$; left leg: $F = 0.538$, $p = 0.660$, $\eta_G^2 = 0.056$) for muscle thickness measured in both the right and left legs (Table 11 and Figure 6). Further analyses revealed that, for the right leg, muscle thickness significantly increased from pre-exercise levels (3.43 ± 0.70 cm) at immediately post- (3.76 ± 0.68 cm, $p < 0.01$) and 30 min post-exercise (3.58 ± 0.73 cm, $p = 0.03$) and

returned to resting levels 60 min post-exercise (3.48 ± 0.70 cm, $p = 1.00$). Additionally, immediately post- ($p < 0.01$) and 30 min ($p = 0.02$) post-exercise measures were also significantly greater than 60 min post-exercise levels. Similar results were observed for the left leg with muscle thickness peaking immediately post-exercise (3.81 ± 0.66 cm) compared to resting (3.44 ± 0.61 cm, $p < 0.01$), 30 min (3.54 ± 0.68 cm, $p < 0.01$) and 60 min (3.45 ± 0.68 cm, $p < 0.01$) post-exercise values.

Table 11. Absolute values for muscle thickness before and after each exercise condition ($n = 10$).

<i>Right Leg</i>	Pre-exercise^α	0 min^β	30 min^β	60 min^α
LLRE+BFR	3.39 ± 0.64	3.75 ± 0.61	3.59 ± 0.63	3.52 ± 0.61
HL-RE	3.46 ± 0.80	3.77 ± 0.78	3.57 ± 0.85	3.44 ± 0.82
<i>Left Leg</i>	Pre-exercise^α	0 min^β	30 min^α	60 min^α
LLRE+BFR	3.44 ± 0.60	3.82 ± 0.58	3.61 ± 0.64	3.49 ± 0.61
HL-RE	3.44 ± 0.65	3.80 ± 0.76	3.48 ± 0.84	3.43 ± 0.78

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

^{αβ}Different Greek letters represent significant ($p < 0.05$) time main effect differences. Data are mean \pm SD.

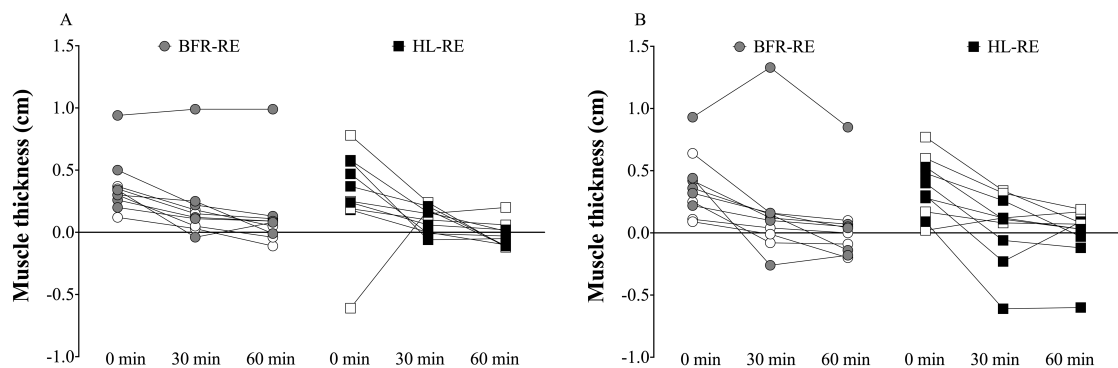


Figure 6. Individual absolute changes in muscle thickness (cm) from pre-exercise immediately post- and at 30 and 60 min following each exercise condition ($n = 10$).

A: Right leg, B: left Leg, LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

Thigh Circumference

There was a significant time main effect for thigh circumference (Table 12 and Figure 7) for both right and left legs (right leg: $F = 13.16, p < 0.001, \eta_G^2 = 0.01$; left leg: $F = 16.45, p < 0.001, \eta_G^2 < 0.01$), but no significant condition main (right leg: $F = 0.02, p = 0.897, \eta_G^2 < 0.01$; left leg: $F = 0.46, p = 0.510, \eta_G^2 < 0.01$) effect or significant condition \times time interaction (right leg: $F = 0.33, p = 0.806, \eta_G^2 < 0.01$; left leg: $F = 0.143, p = 0.860, \eta_G^2 = 0.01$). Follow up analysis of the time main effect, for the right leg, revealed that thigh circumference increased significantly compared to pre-exercise levels (61.52 ± 9.36 cm) immediately post- (62.36 ± 9.30 cm, $p < 0.01$) and 30 min post-exercise (61.99 ± 9.36 cm, $p = 0.01$), which were both significantly greater than 60 min-post-exercise (61.25 ± 9.25 cm, $p < 0.01$ for both). Regarding the left leg, significant increases from baseline (61.15 ± 9.63 cm) were detected only immediately post-exercise (62.14 ± 9.75 cm), which was also significantly greater than 30 min (61.52 ± 9.57 cm, $p < 0.01$) and 60 min (60.72 ± 9.32 cm) post-exercise measures.

Table 12. Absolute values for thigh circumference (cm) before and after each exercise condition ($n = 15$).

<i>Right Leg</i>	Pre-exercise ^α	0 min ^β	30 min ^β	60 min ^α
LLRE+BFR	61.57 ± 9.58	62.37 ± 9.62	61.85 ± 9.51	61.24 ± 9.20
HL-RE	61.47 ± 9.47	62.35 ± 9.32	62.12 ± 9.54	61.26 ± 9.62
<i>Left Leg</i>	Pre-exercise ^α	0 min ^β	30 min ^α	60 min ^α
LLRE+BFR	61.29 ± 9.63	62.21 ± 9.8	61.59 ± 9.95	60.88 ± 9.45
HL-RE	61.03 ± 9.95	61.99 ± 10.08	61.43 ± 9.74	60.57 ± 9.51

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

^{αβ}Different Greek letters represent significant ($p < 0.05$) time main effect differences. Data are mean \pm SD.

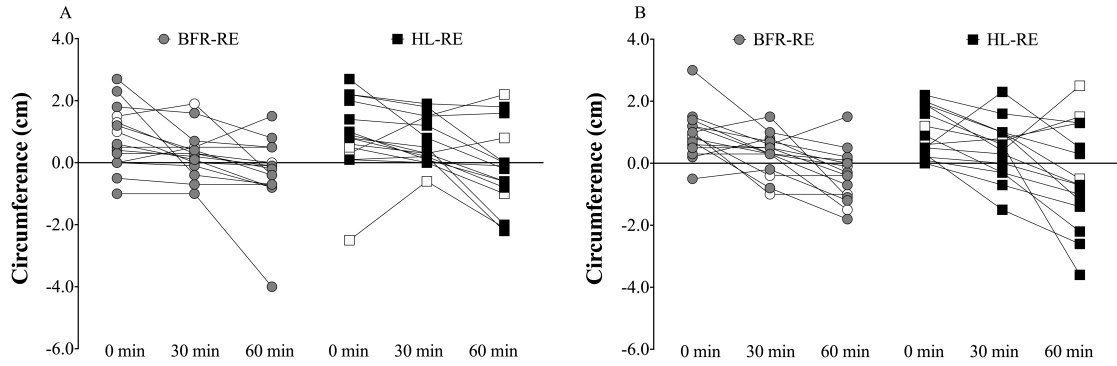


Figure 7. Individual absolute changes in thigh circumference (cm) from Pre-exercise immediately post- and at 30 and 60 min following each exercise condition ($n = 10$). A: Right leg, B: left Leg, $_{LL}RE+BFR$: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Filled symbols (i.e., ●/■) represent females and clear symbols (i.e., □/○) represent males.

Electromyography

Table 13 displays the changes in the myoelectric activity of the vastus medialis and vastus lateralis of both right and left leg during the leg press exercise. No significant condition \times time interactions ($F \leq 1.10$, $p \geq 0.333$, $\eta_G^2 < 0.01$) or time main effects ($F \leq 2.90$, $p \geq 0.065$, $\eta_G^2 \leq 0.01$) were observed for any of the muscles or legs analyzed; however, significant condition main effects ($F \geq 101.42$, $p < 0.001$, $\eta_G^2 \geq 0.75$) were observed in all analyses, demonstrating that HL-RE tends to elicit greater myoelectric activity than $_{LL}RE+BFR$, regardless of the muscle or leg utilized during the leg press exercise.

Table 13. Electromyography amplitude values (% of 1-RM) within each set of both exercise conditions during leg press ($n = 15$).

	Set 1	Set 2	Set 3	Set 4	Condition Effect	Time Effect	Interaction
<i>RL_VM</i>							
$_{LL}RE+BFR$	29.29 ± 10.36	27.29 ± 11.04	26.63 ± 10.02	25.53 ± 11.39	F = 160.11 $p < 0.001$	F = 2.39 $p = 0.082$	F = 0.52 $p = 0.333$
HL-RE	75.84 ± 8.72	77.90 ± 10.44	74.64 ± 8.58	75.58 ± 11.65	$\eta_G^2 = 0.86$	$\eta_G^2 = 0.01$	$\eta_G^2 < 0.01$
<i>RL_VL</i>							
$_{LL}RE+BFR$	28.11 ± 10.30	26.07 ± 10.86	26.13 ± 10.41	25.41 ± 11.71	F = 127.17 $p < 0.001$	F = 2.59 $p = 0.065$	F = 0.80 $p = 0.499$
HL-RE	80.60 ± 12.87	80.31 ± 16.52	76.66 ± 14.33	76.74 ± 14.71	$\eta_G^2 = 0.81$	$\eta_G^2 = 0.01$	$\eta_G^2 < 0.01$
<i>LL_VM</i>							
$_{LL}RE+BFR$	32.59 ± 10.73	28.74 ± 9.30	28.60 ± 9.99	28.28 ± 9.22	F = 101.42 $p < 0.001$	F = 1.31 $p = 0.283$	F = 1.10 $p = 0.361$
HL-RE	74.36 ± 16.82	75.68 ± 16.86	74.46 ± 16.56	73.64 ± 15.61	$\eta_G^2 = 0.75$	$\eta_G^2 < 0.01$	$\eta_G^2 < 0.01$
<i>LL_VL</i>							
$_{LL}RE+BFR$	32.38 ± 13.02	29.27 ± 12.47	28.93 ± 12.75	27.86 ± 10.88	F = 112.67 $p < 0.001$	F = 2.90 $p = 0.075$	F = 0.29 $p = 0.71$
HL-RE	83.40 ± 16.87	82.39 ± 17.10	80.18 ± 17.86	80.77 ± 16.30	$\eta_G^2 = 0.77$	$\eta_G^2 = 0.01$	$\eta_G^2 < 0.01$

$_{LL}RE+BFR$: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. RL_VM: Right leg vastus medialis muscle, RL_VL: Right leg vastus lateralis muscle, LL_VM: Left leg vastus medialis muscle, LL_VL: Left leg vastus lateralis muscle.

Table 14 displays the changes in the myoelectric activity of the vastus medialis and vastus lateralis of both right and left leg during the knee extension exercise. Differently than what was observed during knee extension exercise, there was a significant condition \times time interaction ($F = 4.31, p = 0.043, \eta_G^2 = 0.01$) and a significant condition main effect ($F = 81.82, p < 0.001, \eta_G^2 = 0.53$), but no significant time main effect ($F = 3.83, p < 0.057, \eta_G^2 = 0.01$) for the electromyography amplitude measured in the vastus medialis of the right leg. Although a significant condition \times time interaction was observed, further analyses demonstrated that significantly ($p < 0.001$) greater EMG

amplitude were observed during HL-RE compared to $_{LL}RE+BFR$ during all sets, and that no significant differences were observed across sets within HL-RE ($p \geq 0.26$) or $_{LL}RE+BFR$ ($p \geq 0.37$). For the remaining muscle groups of both legs, there were no significant condition \times time interactions ($F \leq 1.98$, $p \geq 0.16$, $\eta_G^2 < 0.01$) and although a significant ($F = 6.22$, $p = 0.01$, $\eta_G^2 = 0.02$) time main effect was detected for the vastus medialis of the left leg, pairwise comparisons revealed that such difference does not actually exist ($p \geq 0.077$). Finally, no additional significant time main effects were observed for the remaining muscle groups ($F \leq 1.86$, $p \geq 0.19$, $\eta_G^2 < 0.01$), whereas significant ($F \geq 56.24$, $p < 0.001$, $\eta_G^2 \leq 0.53$) condition main effects were observed for all, demonstrating that HL-RE tends to elicit greater myoelectric activity than $_{LL}RE+BFR$, regardless of the muscle or leg utilized during the knee extension exercise.

Table 14. Electromyography amplitude values (% of 1-RM) within each set of both exercise conditions during knee extension ($n = 15$).

	Set 1	Set 2	Set 3	Set 4	Condition Effect	Time Effect	Interaction
<i>RL_VM</i>							
LLRE+BFR	57.08 ± 13.78	54.73 ± 14.84	55.62 ± 15.07	57.28 ± 16.30	F = 81.82 $p < 0.001$ $\eta_G^2 = 0.53$	F = 3.83 $p = 0.057$ $\eta_G^2 = 0.01$	F = 4.31 $p = 0.043$ $\eta_G^2 = 0.01$
HL-RE	100.24 21.12	105.44 27.79	110.48 34.51	115.46 42.22			
<i>RL_VL</i>							
LLRE+BFR	55.55 ± 16.69	51.94 ± 15.29	52.64 ± 16.20	54.17 ± 16.91	F = 66.01 $p < 0.001$ $\eta_G^2 = 0.58$	F = 1.14 $p = 0.29$ $\eta_G^2 < 0.01$	F = 1.98 $p = 0.16$ $\eta_G^2 < 0.01$
HL-RE	101.64 ± 20.33	103.97 ± 23.54	107.48 ± 29.60	109.08 ± 35.17			
<i>LL_VM</i>							
LLRE+BFR	51.49 ± 15.63	51.64 ± 17.46	52.98 ± 16.84	54.30 ± 16.67	F = 100.06 $p < 0.001$ $\eta_G^2 = 0.62$	F = 6.22 $p = 0.01$ $\eta_G^2 = 0.02$	F = 1.98 $p = 0.17$ $\eta_G^2 < 0.01$
HL-RE	97.27 ± 17.29	103.02 ± 21.12	105.22 ± 25.73	108.58 ± 30.60			
<i>LL_VL</i>							
LLRE+BFR	57.18 ± 15.45	57.88 ± 15.90	60.86 ± 16.78	60.22 ± 16.97	F = 56.24 $p < 0.001$ $\eta_G^2 = 0.55$	F = 1.86 $p = 0.19$ $\eta_G^2 < 0.01$	F = 0.38 $p = 0.64$ $\eta_G^2 < 0.01$
HL-RE	100.77 ± 19.87	102.30 ± 22.99	102.34 ± 22.48	104.96 ± 28.84			

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. RL_VM: Right leg vastus medialis muscle, RL_VL: Right leg vastus lateralis muscle, LL_VM: Left leg vastus medialis muscle, LL_VL: Left leg vastus lateralis muscle.

Table 15 outlines a comparison of the myoelectric activity of the right versus the left leg during both leg press and knee extension exercises using the vastus lateralis and vastus medialis muscles. In all analyses, there were no significant condition \times leg interactions ($F \leq 1.04$, $p \geq 0.32$, $\eta_G^2 < 0.01$), except for the vastus lateralis muscle during knee extension ($F = 4.77$, $p = 0.046$, $\eta_G^2 = 0.01$), however, pairwise comparisons reveal

that such effect does not actually exist. Furthermore, there were no significant leg main effects for any of the analyzes ($F \leq 1.86$, $p \geq 0.19$, $\eta_G^2 \leq 0.01$), demonstrated that participants did not display any limb asymmetry, when comparing the right and left legs. Finally, there were also significant condition main effects, with HL-RE being significantly greater than $_{LL}RE+BFR$.

Table 15. Electromyography amplitude values within each set of both exercise conditions during knee extension ($n = 15$).

	Right Leg	Left Leg	Condition Effect	Leg Effect	Interaction
<i>LP_VM</i>					
$_{LL}RE+BFR$	27.34 ± 10.32	29.55 ± 9.32	F = 155.29 $p < 0.001$ $\eta_G^2 = 0.82$	F = 0.02 $p = 0.89$ $\eta_G^2 < 0.01$	F = 1.04 $p = 0.32$ $\eta_G^2 < 0.01$
HL-RE	75.99 ± 8.87	74.53 ± 15.34			
<i>LP_VL</i>					
$_{LL}RE+BFR$	26.43 ± 10.49	29.61 ± 11.87	F = 135.37 $p < 0.001$ $\eta_G^2 = 0.81$	F = 1.86 $p = 0.19$ $\eta_G^2 < 0.01$	F < 0.01 $p = 0.98$ $\eta_G^2 < 0.01$
HL-RE	78.58 ± 13.54	81.69 ± 16.03			
<i>KE_VM</i>					
$_{LL}RE+BFR$	56.18 ± 14.58	52.60 ± 16.46	F = 107.34 $P < 0.001$ $\eta_G^2 = 0.59$	F = 0.80 $P = 0.39$ $\eta_G^2 = 0.01$	F = 0.03 $P = 0.86$ $\eta_G^2 < 0.01$
HL-RE	107.91 ± 30.62	103.52 ± 22.87			
<i>KE_VL</i>					
$_{LL}RE+BFR$	53.58 ± 16.01	59.03 ± 15.89	F = 67.85 $P < 0.001$ $\eta_G^2 = 0.59$	F = 0.17 $P = 0.69$ $\eta_G^2 < 0.01$	F = 4.77 $P = 0.046$ $\eta_G^2 = 0.01$
HL-RE	105.54 ± 26.14	102.59 ± 22.71			

$_{LL}RE+BFR$: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. LP_VM: Myoelectric activity of the vastus medialis muscle during leg press, LP_VL: Myoelectric activity of the vastus lateralis muscle during leg press, KE_VM: Myoelectric activity of the vastus medialis muscle during knee extension, KE_VL: Myoelectric activity of the vastus lateralis muscle during knee extension.

Data are mean ± SD.

Figure 8 illustrates the changes in myoelectric activity of the vastus medialis and vastus lateralis muscle of right and left legs from the initial 3 repetitions to the final 3 repetitions of the leg press and knee extension exercise, within both experimental conditions. During leg press, $_{LL}BFR-RE$ resulted in significantly greater myoelectric activity than $HL-RE$ for all muscle groups (all $p < 0.001$), with no significant differences between the initial and last 3 repetitions within both conditions ($p > 0.05$), except for the vastus lateralis muscle of the right leg, in which a significant increase from the first to the last 3 sets was observed within the $_{LL}BFR-RE$ condition ($p < 0.001$). During knee extension, there were similar results with $_{LL}BFR-RE$ inducing significantly greater myoelectric activity than $HL-RE$ for all muscle groups (all $p < 0.001$). Additionally, there were also significant time effects (all $p < 0.001$), with greater myoelectric activity being observed during the last 3 repetitions compared to the first three repetitions for both exercise protocols.

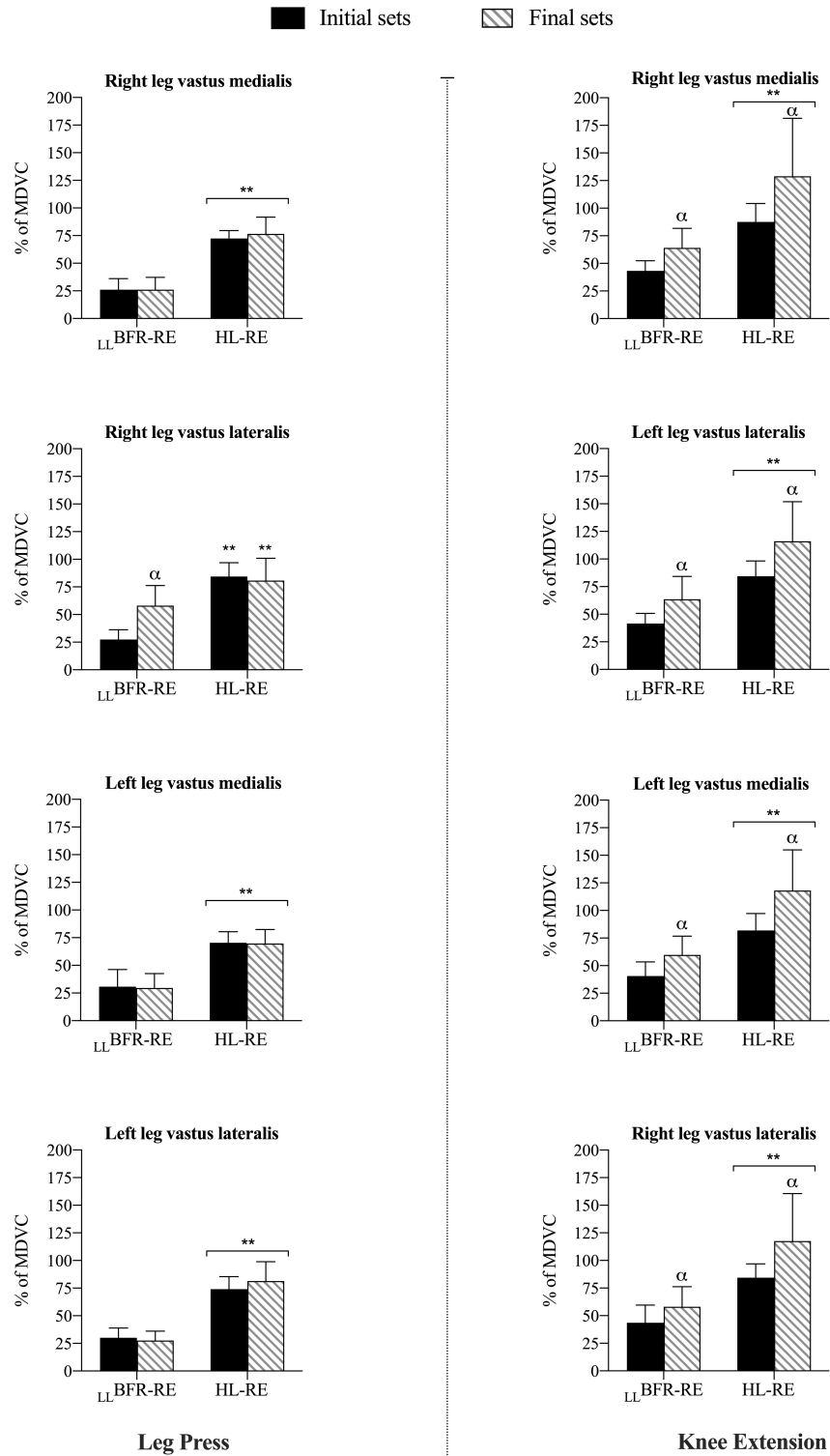


Figure 8. Surface electromyography during the initial 3 repetitions and final 3 repetitions of leg press and knee extension within each experimental trial ($n = 15$). LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. **Significantly greater than LLRE+BFR at $p < 0.01$, α Significantly greater than the initial repetitions at $p < 0.05$. Data are mean \pm SD.

1-RM test re-test reliability

As presented on Table 16, 1-RM reliability was exercise dependent. For the leg press exercise, although a significant intraclass correlation coefficient was observed ($p < 0.001$, ICC = 0.847), there was also a significant ($t = 4.36$, $p < 0.001$) 14.32 % increase in the mean 1-RM score. On the other hand, a larger significant ICC ($p < 0.001$, ICC = 0.932) was observed for the 1-RM test for the knee extension exercise, with no significant ($t = 0.71$, $p < 0.491$) difference in the mean 1-RM scores from trial 1 and 2.

Table 16. Changes in 1-RM values (kg) from Trial 1 to Trial 2 for both Leg Press and Knee Extension ($n = 15$).

<i>Leg Press</i>							
Trial 1	Trial 2	Δ	Δ%	SEM	t	p	ICC
101.93 ± 29.46	114.48 ± 27.75**	12.55 ± 11.15	14.32 ± 13.05	7.88	4.36	< 0.001	0.847**
<i>Knee Extension</i>							
Trial 1	Trial 2	Δ	Δ%	SEM	t	p	ICC
50.77 ± 12.97	51.67 ± 13.30	0.90 ± 4.91	2.61 ± 10.20	3.47	0.71	0.491	0.932**

**Significant p-value at $p \leq 0.001$. Data are mean ± SD. Δ: Absolute change from Trial 1 to Trial 2, Δ%: Percent change from Trial 1 to Trial 2, SEM: Standard error of the measurement, t: paired t-test value, p: p-value, ICC: Intraclass correlation coefficient.

Perceptual Responses

Ratings of Perceived Exertion

As presented on Table 17, HL-RE elicited significantly ($p \leq 0.01$) greater RPE than LLRE+BFR following all sets of leg press, and after sets 2 to set 4 of knee extension. Additionally, RPE levels observed following set 3 were significantly ($p < 0.05$) than those observed after set 2 for both experimental trials during leg press, whereas no significant ($p > 0.05$) differences were observed across sets for either exercise condition during knee extension. Finally, Figure 9 presents the RPE scores for each participant averaged across all four sets of LP and KE within each experimental trial, and it demonstrates that HL-RE elicited a significantly greater overall RPE response than LLRE+BFR during both LP ($p < 0.01$) and KE ($p = 0.01$).

Table 17. Ratings of perceived of exertion for both experimental conditions during each set of leg press and knee extension ($n = 15$).

					Time ($p < 0.05$)
<i>Leg Press</i>	Set 1	Set 2	Set 3	Set 4	
LLRE+BFR	4.0 ± 1.1	3.5 ± 1.4	4.5 ± 1.4	4.4 ± 1.4	2 < 3
HL-RE	6.8 ± 0.9**	6.7 ± 1.4*	7.6 ± 1.3**	7.6 ± 1.7**	2 < 3
<i>Knee Extension</i>	Set 1	Set 2	Set 3	Set 4	
LLRE+BFR	6.8 ± 1.7	6.7 ± 1.5	7.0 ± 1.7	7.2 ± 1.4	N.S.
HL-RE	8.1 ± 1.0	8.9 ± 0.9**	8.9 ± 0.9**	9.2 ± 1.0**	N.S.

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

*Significant condition effect at $p \leq 0.05$, **Significant condition effect at $p \leq 0.01$. Data are Winsorized mean ± Winsorized SD.

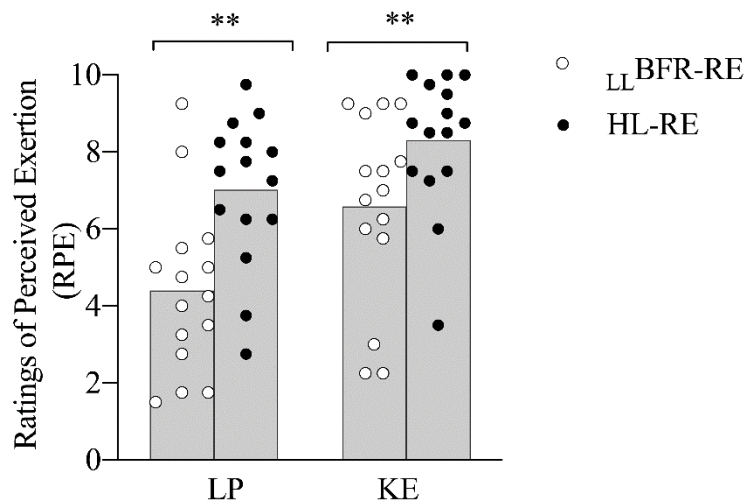


Figure 9. Individual Ratings of Perceived Exertion (RPE) values averaged across sets within conditions during leg press and knee extension ($n = 15$).

LLBFR+RE: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

**Significant condition effect at $p \leq 0.01$. Winsorized means (vertical bars) and individual data (dots) from each participant is presented.

Ratings of Pain

Table 18 presents the ratings of pain values assessed immediately before and immediately after each set of leg press and knee extension during the LLRE+BFR and HL-RE trials. For the ratings of pain measured prior to each set, LLBFR+RE elicited significantly ($p < 0.05$) greater pain than HL-RE before sets 3 and 4 of leg press and before sets 2, 3, and 4 of knee extension. Curiously, immediately after each set of the leg press and knee extension exercises, both LLBFR+RE and HL-RE protocols resulted in similar ($p > 0.05$) ratings of pain; except after the first set of knee extension, when LLBFR+RE was significantly ($p < 0.05$) greater than HL-RE.

Regarding the comparisons across sets within each condition, the ratings of pain measured before sets 2, 3, and 4 were significantly ($p < 0.05$) greater than those measured before set 1 (i.e., pre-exercise), with no significant ($p > 0.05$) differences across sets 2,

3, and 4 for either leg press or knee extension during BFR+RE. During HL-RE, significant ($p < 0.05$) differences from pre-set 1 values were observed only before set 4, for leg press, and before sets 3 and 4, for knee extension. Regarding the pain levels measured immediately after each set, post-sets 3 and 4 were significantly ($p < 0.05$) different than post-set 1 values, and post-set 4 was also significantly ($p < 0.05$) different than post-set 2 for during $_{LL}$ BFR+RE for the knee leg press exercise, whereas no significant ($p > 0.05$) differences existed across post-set measures within the $_{LL}$ RE+BFR during knee extension or for the HL-RE exercise condition during either leg press or knee extension exercises.

Finally, there were significant ($p \leq 0.05$) increases in the ratings of pain from immediately before to immediately after sets 1 and 4 of leg press and sets 1, 3, and 4 of knee extension, for the $_{LL}$ RE+BFR condition (Figure 10). During HL-RE, significant ($p \leq 0.05$) pre- to post-set elevations in pain levels were observed during sets 1, 3, and 4 of leg press and all sets of knee extension.

Table 18. Ratings of pain immediately before and immediately after each set for both experimental conditions during leg press and knee extension ($n = 15$).

<i>Pre-set pain levels</i>					
<i>Leg press</i>	Pre-Set 1	Pre-Set 2	Pre-Set 3	Pre-Set 4	Time ($p < 0.05$)
LLRE+BFR	0.0 ± 0.0	2.7 ± 0.9*	2.9 ± 1.4**	3.2 ± 1.7**	$I < 2, 3, 4$
HL-RE	0.0 ± 0.0	0.3 ± 0.5	0.4 ± 0.5	0.9 ± 0.9	$I < 4$
<i>Knee extension</i>					
	0.6 ± 0.9	3.60 ±	2.85 ±	2.93 ±	$I < 2, 3, 4$
LLBFR-RE		1.7**	1.8**	1.90*	
HL-RE	0.3 ± 0.5	1.17 ± 1.3	0.98 ± 0.7	1.48 ± 1.18	$I < 2, 4$
<i>Post-set pain levels</i>					
<i>Leg press</i>	Post-Set 1	Post-Set 2	Post-Set 3	Post-Set 4	Time ($p < 0.05$)
LLRE+BFR	2.4 ± 1.2	2.7 ± 1.3	3.4 ± 1.2	4.0 ± 1.8	$I < 3, 4; 2 < 4$
HL-RE	1.2 ± 0.8	1.5 ± 1.6	2.1 ± 1.5	2.4 ± 1.9	<i>N.S.</i>
<i>Knee extension</i>					
LLRE+BFR	4.6 ± 1.8**	3.6 ± 1.3	3.8 ± 1.6	3.9 ± 2.1	<i>N.S.</i>
HL-RE	2.2 ± 1.6	3.0 ± 1.7	3.1 ± 1.7	3.4 ± 2.2	<i>N.S.</i>

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

*Significant condition effect at $p \leq 0.05$, **Significant condition effect at $p \leq 0.01$. Data are Winsorized mean ± Winsorized SD.

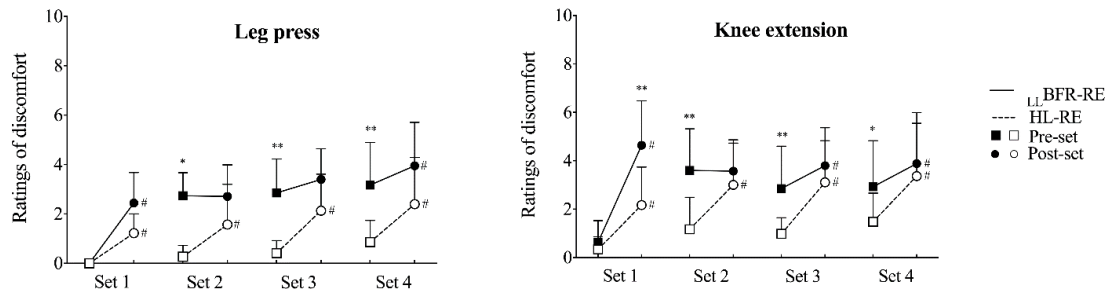


Figure 10. Changes in the ratings of pain from pre to post each set within both experimental trials ($n = 15$).

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise.

*Significant condition effect at $p \leq 0.05$, **Significant condition effect at $p \leq 0.01$, #Significant pre- to post-set difference within each individual set at $p \leq 0.05$. Data are Winsorized means ± Winsorized SD.

Soreness

Table 19 presents the changes in soreness following each bout of exercise. There were no significant ($p > 0.05$) differences in soreness levels between conditions at any time point from 5 min up to 24 h post-exercise. The pairwise comparisons across time within the **LLRE+BFR** condition revealed that soreness levels after 30 min and 60 min post-exercise were significantly ($p = 0.028$ and 0.029 , respectively) lower than 5 min post-exercise measures, but not significantly ($p = 1.00$) different than 24 h post-exercise. For the **HL-RE** exercise trial, the soreness measured 24 h post-exercise was significantly ($p = 0.025$) greater than that from 60 min after exercise. No other significant time differences were observed for the **HL-RE** protocol.

Table 19. Soreness levels following each experimental trial ($n = 15$).

	5 min	30 min	60 min	24 h	Time ($p < 0.05$)
LLRE+BFR	0.9 ± 0.9	0.1 ± 0.2	0.0 ± 0.0	0.8 ± 0.9	$5 > 30, 60$
HL-RE	0.9 ± 1.0	0.1 ± 0.2	0.0 ± 0.0	1.3 ± 1.3	$24 > 60$

LLRE+BFR: low-load resistance exercise with blood flow restriction, HL-RE: high-load resistance exercise. Data are Winsorized means \pm Winsorized SD.

Modified Fatigue Impact Scale

The score for each domain of the MFIS is presented below on Table 20. The Friedman Rank test demonstrated that no significant ($p = 0.063$) differences existed in the physical domain scores across visits, although significant time effects were observed for the cognitive ($p = 0.001$) and psychological ($p = 0.004$) domains. Pairwise comparisons utilizing the Wilcoxon Signed-Rank test revealed that significantly ($p = 0.05$) lower scores for the cognitive domain were observed during visit 5 in comparison

to visit 1, while no actual significant ($p > 0.05$) difference existed across visits for the psychological domain.

Table 20. Modified Fatigue Impact Scale Scores for each visit ($n = 15$).

	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5
Physical	14.7 ± 8.9	9.1 ± 8.6	10.0 ± 9.0	10.0 ± 8.7	8.0 ± 6.9
Cognitive	10.2 ± 5.6	6.2 ± 5.0	4.6 ± 4.3	5.9 ± 4.7	5.3 ± 4.4 ^α
Psychological	3.2 ± 2.9	0.9 ± 1.1	0.9 ± 1.0	1.9 ± 1.8	1.4 ± 1.8

^αSignificantly different than Visit 1 at $p = 0.05$. Data are Winsorized means ± Winsorized SD.

Discussion

Physiological Responses

Whole-Blood Lactate

Whole-blood lactate was measured as a means to estimate the exercise-induced metabolic response. This study demonstrated that HL-RE induced a greater metabolic response 5 min post-exercise compared to the LLRE+BFR trial, which corresponded to approximately a 212% increase for the former and 134% increase for the latter. These data suggest that, although LLRE+BFR seems to induce a smaller metabolic response than HL-RE, it is still capable of evoking a considerable metabolic stress.

Although there are no studies that have investigated the metabolic response of individuals living with MS to LLRE+BFR, the results of the current investigation are in line with previous data from our laboratory in a cohort of healthy young individuals (Freitas, Galletti, et al., 2020; Freitas, Miller, et al., 2020). In these studies, it was also observed a smaller metabolic response following LLRE+BFR in comparison to HL-RE. Previous studies from other research groups have also corroborated these findings. For instance, Suga et al. (2009) used P-magnetic resonance spectroscopy to compare the metabolic stress during a single bout of LLRE+BFR (20% of 1-RM) and HL-RE (65% of 1-RM) and observed that HL-RE induced a greater metabolic response in the form of a greater decrease in pH, higher concentration of H₂PO₄, and greater phosphocreatine utilization. Curiously, in a follow-up study, Suga et al. (2010) were able to replicate their previous findings that LLRE+BFR (20% of 1-RM) elicits a greater metabolic response than HL-RE (65% of 1-RM), but in addition to that, the authors also observed that the gap in the metabolic response to both exercise conditions shrinks and is eventually

reversed once higher exercise loads are used. In fact, similar changes in intramuscular pH and metabolites were observed between LLRE+BFR, performed at 30% of 1-RM, and HL-RE (65% of 1-RM). Conversely, the decrease in intramuscular pH, creatine phosphate utilization, and increase in H₂PO₄ were much greater following LLRE+BFR, performed at 40% of 1-RM compared to HL-RE (65% of 1-RM). Therefore, the smaller metabolic response to LLRE+BFR compared to HL-RE observed in the present study may be due to the lower exercise intensity of 20% of 1-RM used in the LLRE+BFR condition while loads of 70% of the participants' 1-RM were used in the HL-RE condition.

It should also be noted that, in comparison to previous literature, the lactate response observed in the current study was much smaller than that of healthy individuals. For instance, a previous study from our research group using a similar protocol reported lactate levels of 5.82 ± 2.28 mmol/L and 9.42 ± 2.14 mmol/L 5 min following LLRE+BFR and HL-RE, respectively (Freitas, Miller, et al., 2020), whereas lactate values of 2.28 ± 0.72 and 3.85 ± 1.65 were observed following the same respective exercise conditions in the current study. These results may be partially explained by the fact that the participants enrolled in the present investigation displayed smaller absolute maximal dynamic strength (leg press: 115.16 ± 28.90 versus 203.15 ± 38.65 , knee extension: 52.67 ± 13.54 versus 95.97 ± 17.36). Maximal strength levels play a vital role on the exercise-induced lactate response as individuals displaying higher maximal strength levels are capable of exercising using higher loads and exerting more strength compared to less strong counterparts. It should also be highlighted that the majority of the individuals included in the current investigation were females, which have been shown to display a smaller

lactate response than males following both $LLRE+BFR$ and $HL-RE$ (Freitas, Galletti, et al., 2020).

Altogether, these results demonstrate that $LLRE+BFR$ may elicit a significant metabolic response, in the form of lactate accumulation, in individuals living with MS. Although such metabolic response was lower than that observed with $HL-RE$ and the accumulating evidence (Takada et al., 2012) suggesting that the exercise-induced metabolic stress may play an important role in the long-term skeletal muscle hypertrophic adaptation to $LLRE+BFR$, this study demonstrates that $LLRE+BFR$ may serve as a potential alternative to people living with MS that cannot withstand higher training loads or that would simply prefer to lift lighter weights.

Electromyography

Myoelectric activity of the vastus medialis and lateralis of both legs was measured using superficial electromyography (sEMG). The data demonstrate that $HL-RE$ induced greater myoelectric activity than $LLRE+BFR$ for both muscle groups during the leg press ($\approx 28\%$ versus $\approx 75\%$, respectively) and the knee extension exercises ($\approx 60\%$ versus $\approx 100\%$, respectively). Although inferior in comparison to $HL-RE$, $LLRE+BFR$ was capable of resulting in a substantial increase in the myoelectric activity of the tested muscle groups.

The findings of the current study are in accordance with previous literature demonstrating that $LLRE+BFR$ tends to induce lower changes in sEMG amplitude compared to $HL-RE$. For instance, Fatela et al. (2018) employed a similar experimental design to the one used in the current study, in which 10 healthy young men completed 4

sets of 30+15+15+15 isokinetic concentric contractions of $LLRE+BFR$ (at 20% of 1-RM) with 80% of BFR and 4 sets of 10 isokinetic concentric contractions each of HL-RE (at 75% of 1-RM) for the knee extension. Myoelectric activity of the vastus medialis and rectus femoris muscles was reported as root mean square amplitude. The authors demonstrated that, similarly to the current investigation, HL-RE elicited greater myoelectric activity of both muscles measured compared to $LLRE+BFR$. Furthermore, in agreement with the current study, the authors also demonstrated that $LLRE+BFR$ was able to induce a substantial increase in the myoelectric activity during $LLRE+BFR$ and suggested that it may serve as an effective training alternative to HL-RE.

Although global sEMG amplitude has several drawbacks to estimate the order of motor unit recruitment, previous studies using P-magnetic resonance spectroscopy and split inorganic phosphate has demonstrated that $LLRE+BFR$ is capable of inducing the recruitment of the fast-twitch muscle fibers (Suga et al., 2012).

To the best of my knowledge, this study was the first to compare the myoelectric activity between the right and left legs of individuals with MS. Although no previous study has performed such comparison, Chung et al., (2008) reported limb asymmetries in peak torque and power in people with MS compared to healthy controls. However, in their analysis, the authors did not identify which leg (i.e., right or left) greater or smaller torque or power levels. A few differences between the current study and that by Chung et al., (2008) should be highlighted. Firstly, in the current study, both legs performed the physical task simultaneously, whereas Chung et al., (2008) test each leg separately. Secondly, participants in Chung's et al., (2008) study had slightly higher EDSS scores

(4 ± 1) compared to the ones included in the current study (1.87 ± 1.51), which could have contributed to attenuate any potential leg differences.

Muscle Swelling

In this study, exercise-induced muscle swelling was estimated utilizing measures of muscle thickness and thigh circumference. Additionally, changes in plasma volume were also used to indirectly estimate fluid shifts into the muscle. This study demonstrated that both the $LLRE+BFR$ and the $HL-RE$ conditions resulted in similar increases in muscle thickness and thigh circumference that lasted for up to 30 min post-exercise, compared to baseline levels. However, although plasma volume slightly decreased 5 min post-exercise, it did not reach statistical significance.

Although only a few studies have directly compared the acute effects of $LLRE+BFR$ and the $HL-RE$ on post-exercise muscle swelling, the findings of the current study are in agreement with previous studies demonstrating that $LLRE+BFR$ is capable of eliciting significant muscle swelling post-exercise (Freitas et al., 2017; Nyakayiru et al., 2019; Wilson et al., 2013; Tomohiro Yasuda et al., 2015). Nonetheless, there is conflict findings regarding the separate effects of the $LLRE+BFR$ and $HL-RE$ protocols on muscle swelling. For example, Freitas et al. (2017) reported greater muscle thickness, measured via ultrasound, immediately post-exercise for $LLRE+BFR$ compared to $HL-RE$, whereas no differences in muscle swelling were observed between conditions 15 min post-exercise when estimated through either muscle cross sectional area (measured via peripheral quantitative tomography) or thigh circumference. Follow-up studies from our laboratory

have confirmed that LLRE+BFR and HL-RE seems to induce similar muscle swelling (Freitas, Galletti, et al., 2020; Freitas, Miller, et al., 2020).

The potential contributions of the exercise-induced muscle swelling to the skeletal muscle chronic hypertrophic response commonly observed following LLRE+BFR emerged with previous studies demonstrating that the application of BFR in the absence of exercise attenuates muscle atrophy of the quadriceps following surgery of anterior crucial ligament (Takarada, Takazawa, & Ishii, 2000) or immobilization (Kubota et al., 2008). Although the contributions of muscle swelling to prevent muscle atrophy still warrants further investigation, there is accumulating evidence that LLRE+BFR increases rates of myofibrillar hypertrophy to a much greater extent than low-load resistance exercise without BFR.

Inflammation

Inflammation was estimated by measuring post-exercise plasma levels of IL-6. The data demonstrated that no significant changes in IL-6 levels occurred in response to either LLRE+BFR or HL-RE, up to 1 hour post-exercise. Although the IL-6 concentrations observed in the current study were slightly lower than those previously reported in healthy individuals (MacDonald et al., 2003), they were similar to those reported in older subjects (Nicklas et al., 2008) and other clinical populations such as obese and diabetic patients (Abd El-Kader, 2011).

The findings from this study are in accordance with previous literature. For instance, Clark et al. (2011) investigated the acute and chronic (4 weeks) effects of LLRE+BFR and HL-RE on inflammation in healthy young males, estimated in this case

by changes in plasma levels of high-sensitivity c-reactive protein. The $LLRE+BFR$ protocol consisted of 3 sets of knee extension at 30% of 1-RM performed to volitional failure and with BFR set at 130% of the individuals resting systolic blood pressure, while the HL-RE protocols consisted of the same exercise performed at 80% of 1-RM. Similarly to the current study, Clark et al. (2011) reported no acute changes in high-sensitivity c-reactive protein levels up to 1 hour post-exercise, as well as no chronic changes in baseline plasma levels of high-sensitivity c-reactive protein 4 weeks following either $LLRE+BFR$ or HL-RE. Interestingly, in the same study, the authors also reported similar increases in isometric strength following both training methods ($LLRE+BFR = \approx 8\%$ versus $HL-RE = \approx 13\%$) without any changes chronic changes in important safety parameters such as pulse wave velocity, ankle-brachial index, prothrombin time, nerve conduction, fibrinogen, and D-dimer. Karabulut et al. (2013) also compared the long-term (6 weeks) effects of $LLRE+BFR$ (30+15+15+15 repetitions of leg press and knee extension at 20% of 1-RM and with BFR set at 160 mmHg to 240 mmHg) and HL-RE (8+8+8 repetitions of the same exercises at 80% 1-RM) on inflammation (i.e., IL-6) in older men (≈ 56 years old). The authors reported no significant pre to post training differences in plasma IL-6 levels, but, surprisingly, no significant increase in muscle cross-sectional area were detected. In another study investigating the acute effects of $LLRE+BFR$ on plasma IL-6 levels, Bugera et al. (2018) detected no changes in IL-6 levels following a single bout of either $LLRE+BFR$ or HL-RE immediately, 1 hour post-, or 24 hours post-exercise in 1-year resistance trained young males.

Nonetheless, the capacity of $LLRE+BFR$ to cause inflammation should not be completely ruled out. In an earlier study and classic study, Takarada et al. (2000)

demonstrated that LLRE+BFR induced a greater inflammatory response than the same exercise protocol performed without BFR by inducing greater accumulation of plasma IL-6 starting at 30 minutes post-exercise and maintained up to 24 h post-exercise. Similar results, were also observed by Patterson et al. (2013), except that both LLRE+BFR and low-load resistance exercise without BFR induced similar increases in plasma levels of IL-6.

Therefore, based on the finding from the current investigation and the previous research performed on post LLRE+BFR, it seems that LLRE+BFR does not seem to trigger an exaggerated inflammatory response post-exercise in people with MS; at the most, inducing resulting in similar inflammation to HL-RE.

Mammalian Target of Rapamycin Complex 1

This study demonstrated no time or condition differences for plasma levels of mTOR. Before contrasting the aforementioned findings with the current literature on the topic, it is important to highlight that the majority of the studies investigating mTOR activity in response to exercise utilized muscle biopsy samples rather than plasma. Thus, it represents a major limitation of the current study, as changes in plasma levels may not necessarily reflect what is occurring within the intramuscular environment.

Fujita et al. (2007) and Fry et al. (2010) were one of the first to investigate mTOR expression following LLRE+BFR. Although not observing an increase in protein kinase B (also known as Akt) or mTOR up to 3 hours after either LLRE+BFR or low-load resistance exercise without BFR, Fujita et al. (2007) reported a three-fold increase in ribosomal protein S6 kinase beta-1 (S6K1) phosphorylation, a downstream target of

mTOR, 3 hours following LLRE+BFR, whereas no time effects occurred in the control low-load resistance exercise condition. Moreover, Fry et al. (2010) reported an increase in mTOR expression 1 hour after a single bout of LLRE+BFR, which was maintained up to 3 hours post-exercise. Additionally, the 1-hour post-exercise increase was greater than that observed with low-load resistance exercise without BFR. Furthermore, the authors also reported increased phosphorylation of S6K1 at 1 and 3 hours post-exercise, whereas no changes occurred in the control resistance exercise condition. Follow-up studies have confirmed the ability of LLRE+BFR to enhance mTOR signaling pathways in both human and animal models (Gundermann et al., 2012; Nakajima et al., 2016).

Myostatin

Similar to mTOR, no significant condition or time effects were observed for plasma levels of myostatin. Once again, it should be considered that plasma levels of myostatin were measured in the current study, while previous studies have relied on biopsy samples, thus, limiting our ability to interpret the aforementioned findings.

Only a few studies have investigated myostatin expression following LLRE+BFR. A pioneer study in the area was conducted by Drummond et al. (2008), who demonstrated that an acute bout of LLRE+BFR was capable of reducing myostatin gene expression 3 hours post-exercise, although to the same extent as low-load resistance exercise. However, no further comparison to a HL-RE condition was included in the referred study. A recent study using an animal model design (Nakajima et al., 2016) had similar findings to the current investigation in which no significant changes in myostatin expression were detected up to 3 hours post-exercise. There is also evidence that walking with BFR does

not seem to alter myostatin expression acutely (Khoubi et al., 2020), although it has been demonstrated that walking combined with BFR may induce skeletal muscle hypertrophy (Abe et al., 2006; Ozaki et al., 2017). In another study, Laurentino et al. (2012) compared the long-term effects of low-load resistance exercise with and without BFR and HL-RE on myostatin expression as well as skeletal cross-sectional area and dynamic strength. In this study, 8 weeks of LLRE+BFR (knee extension at 20% of 1-RM and 50% of BFR) and HL-RE (knee extension at 80% of 1-RM) promoted significant increases in muscle cross-sectional area (6.3% and 6.1%, respectively) and dynamic strength (40.1% and 36.2%, respectively), which were accompanied by a significant decrease in myostatin gene expression (45% and 41%, respectively).

Although some concerns may be raised regarding the validity of measuring myostatin through ELISA assays using plasma samples, it should however be noted that several previous studies have been capable of detecting significant changes in myostatin concentration using the same procedures (Bagheri et al., 2019; Hittel et al., 2010; Saremi et al., 2010).

Cortisol

Interesting findings for changes in plasma cortisol levels were observed in the current study. Both LLRE+BFR and HL-RE conditions resulted in a decreased in cortisol levels 1 hour post-exercise, compared to baseline values.

These findings contradict many of the previous research investigating the hormonal response to LLRE+BFR, which have demonstrated either significant increases or no post-exercise changes. For instance, Fry et al. (2010) observed significant increases

in cortisol levels from baseline 15 min after $LLRE+BFR$, lasting up to 2 hours post-exercise. Similar results were observed by Fujita et al. (2007) who reported significant increases from baseline at 10 min up to 40 minutes following $LLRE+BFR$. In both Fry et al. (2010) and Fujita et al. (2007), blood samples were taken from 6:00 AM, following an 8-hour fast period, and 15:00 PM. In another study, Madarame et al. (2010) also reported significant increases from baseline in cortisol following $LLRE+BFR$ for either the upper- or lower-body. On the other hand, Patterson et al. (2013) observed no changes in cortisol levels at any time point following an acute bout of $LLRE+BFR$, between 7:00 and 9:00 AM. However, similar findings to the current observation were reported by Chen, Wu, and Cai (2018) who observed significant decreases in cortisol following compared to baseline from immediately post up to 30 min post resistance exercise with and without BFR (between 9:00 and 11:00 AM), however, in the referred study, the authors investigated the effects of BFR combined with local vibration, which was not performed in the current observation. I speculate that the decline in cortisol levels observed in the current study may be due to the fact that baseline levels were measured using blood samples collected following an overnight 8-hour fasting period, after which participants consumed a standardize meal. Food ingestion is known for decreasing cortisol levels (Stachowicz & Lebedzińska, 2016), therefore, the observed reductions in cortisol levels may be due to food consumption and not related to any of the exercise protocols performed. Additionally, another potential mechanism influencing the decreased cortisol concentration observed in this study may be related to natural fluctuations due to the circadian rhythm. Cortisol levels is well known for peaking early in the morning and to decrease towards the end of the day (Hayes et al., 2010).

Only a few studies have directly compared the effects of $LLRE+BFR$ and $HL-RE$ on the post-exercise cortisol response. In this regard, Kim et al. (2014) reported similar increases in cortisol from pre to immediately post both $LLRE+BFR$ (30+15+15+15 repetitions of knee extension and leg press exercises at 20% of 1-RM and BFR pressure set at 200 mmHg) and $HL-RE$ (3 \times 10 repetitions of the same exercises at 80% of 1-RM without BFR). Altogether, the current study demonstrated that both $LLRE+BFR$ and $HL-RE$ induce the same stress response post-exercise in people with MS.

Perceptual Responses

Ratings of Perceived Exertion

Although several studies have investigated the perceptual responses of healthy individuals to different exercise modalities, including resistance (Martín-Hernández et al., 2017; Santos et al., 2019) and endurance exercise (da Silva et al., 2019), the scientific literature is scarce of studies exploring this topic in the context of MS. Nonetheless, Kiselka et al. (Kiselka et al., 2013) reported that people with MS are capable of providing RPE in a similar fashion to healthy individuals during near maximal and submaximal isometric contractions, demonstrating that the disease does not seem to affect a person's perception of muscular exertion. Additionally, although using a different RPE scale, Cleland et al. (2016) demonstrated that individuals with MS were also able to provide reliable RPE estimates during endurance exercise. Therefore, the findings from the current investigation provide novel insight into the perceptual responses of those with MS to different forms of resistance exercise. Specifically, the findings that $_{LL}BFR-RE$ requires less muscular exertion than $HL-RE$ is of great relevance as it would likely translate into $_{LL}BFR-RE$ being an appealing alternative to traditional exercise, which may drive increases in exercise adherence for this clinical population. Several studies have demonstrated that $_{LL}BFR-RE$ leads to positive long-term neuromuscular adaptations in many clinical populations (Alves et al., 2020; Erickson et al., 2019; Groennebaek et al., 2019) and that sometimes it may even match the hypertrophy gains observed following traditional high-load resistance exercise (Lixandrão, Ugrinowitsch, et al., 2018a), although involving less mechanical stress to the joints, no to minimal muscle damage, and, as demonstrated in the current study, lower muscular exertion.

Ratings of Pain

Rating of pain is an additional perceptual variable that impacts an individual's tolerance to a specific exercise modality. In this study, I measured pain immediately before and immediately after each set of exercise. Measuring pain prior to a subsequent set of repetitions provides an indirect measure of recovery status from a previously completed set. Further, given that strict resistance exercise guidelines (e.g., rest intervals) may not be adamantly followed outside of the laboratory setting, this measure provides a baseline for comparison between protocols at similar time points. Hence, it was observed that both resistance exercise protocols tested resulted in similar pain levels immediately after sets, however, pain remained elevated during the rest period between sets and was still elevated prior to a subsequent set during LLBFR-RE compared to HL-RE. This finding is not surprising considering that the restrictive cuffs used to reduce blood flow during LLBFR-RE remained inflated during the entire exercise period. Therefore, deflating the cuffs during the rest intervals between sets may diminish the pain perceived during exercise. Although one may argue that deflating the cuffs may compromise the efficacy of LLBFR-RE, previous research has demonstrated that similar increases in the physiological markers of muscle hypertrophy occur regardless the cuffs remain inflated or are deflated during the rest periods between sets (Freitas, Miller, et al., 2020).

Delayed-Onset Muscle Soreness

Regarding the DOMS response up to 24 h post-exercise. Curiously, similar DOMS were observed between protocols at all time points. Considering that the HL-RE

protocol was performed using higher-loads (70% of 1-RM), it was expected that the greater stress would translate into higher DOMS levels 24 hours following the exercise session, which did not happen. Considering that people with MS commonly display lower absolute strength levels than healthy age matched individuals (Jørgensen et al., 2017) and that participants included in the current study were not resistance trained, it is possible that it resulted in relatively lower loads being lifted during the HL-RE trial. However, DOMS was significantly elevated 24 h post-exercise in comparison to 60 minutes post-exercise for the HL-RE trial only. As mentioned earlier, such increase represents only a “mild pain” as it was rated in the lower end of the pain scale used and should also be highlighted that it did not represent a significant difference from the BFR-RE protocol. Therefore, the clinical and practical relevance of such observation is unknown.

Chapter V: Conclusions

The purpose of this study was to compare the acute physiological and perceptual responses of people living with MS to a single bout of low-load (20% 1RM) resistance exercise with BFR ($LLRE+BFR$) and high-load (70% 1RM) resistance exercise without BFR (HL-RE).

Research Questions

1. *Does $LLRE+BFR$ induce the same metabolic response (whole-blood lactate) as traditional HL-RE?*

$LLRE+BFR$ did not induce the same metabolic response as HL-RE in terms of whole-blood lactate concentrations post-exercise. Although both resistance exercise conditions significantly increased lactate levels 5 min post-exercise, the increases observed following HL-RE ($\approx 210\%$) were significantly greater than those observed following $LLRE+BFR$ ($\approx 130\%$).

2. *Are changes in electromyography amplitude similar between $LLRE+BFR$ and HL-RE?*

Changes in surface electromyography (sEMG) amplitude were significantly greater during HL-RE compared to $LLRE+BFR$, during the leg press and knee extension exercises as well as for the vastus lateralis and vastus medialis muscles of both legs. On average, there was approximately a 28% increase in sEMG during $LLRE+BFR$ versus $\approx 75\%$ increase during HL-RE, during leg press, and approximately 60% versus $\approx 100\%$, respectively, during knee extension.

3. *Is there a difference in the acute exercise-induced muscle swelling response (muscle thickness and thigh circumference) between LLRE+BFR and HL-RE?*

There were no significant differences in the exercise-induced muscle swelling response following both LLRE+BFR and HL-RE, measured in both legs as changes in muscle thickness and thigh circumference. Additionally, the increases in muscle thickness remained elevated up to 30 min post-exercise.

4. *Is the hormonal stress response (cortisol) similar between LLRE+BFR and HL-RE?*

A similar hormonal stress response, in the form of plasma cortisol concentration, was detected following both LLRE+BFR and HL-RE protocols. Further, a reduction from baseline was observed in plasma cortisol levels 1 hour post-exercise, however, such decrease may be related to the fact that participants consumed a light meal before exercise, which may have contributed to the observed decrease in cortisol, rather than the exercise protocols performed. Lastly, diurnal variations of cortisol levels are well known for causing declining cortisol concentrations from morning to afternoon levels.

5. *Do biomolecular markers of muscle anabolism (mTOR) and catabolism (myostatin) display similar responses to LLRE+BFR -RE and HL-RE?*

Similar responses were observed for both mTOR and myostatin following LLRE+BFR and HL-RE. In fact, no significant changes in both markers were detected up to 1 hour following the LLRE+BFR and HL-RE trials.

6. *Is the exercise-induced inflammatory response (interleukin-6) similar between LLRE+BFR and HL-RE?*

The post-exercise inflammatory response was similar between the LLRE+BFR and HL-RE experimental conditions.

7. *Are the post-exercise changes in plasma volume and hematocrit levels similar between LLRE+BFR and HL-RE?*

Similar changes in plasma volume were observed following both LLRE+BFR and HL-RE conditions. Additionally, there were no significant time differences up to 1 hour post-exercise for both testing conditions. Such responses may be attributed to the fact that MS is well known for causing sweating impairments (Saari et al., 2009).

8. *Do LLRE+BFR and HL-RE elicit similar ratings of perceived exertion?*

LLRE+BFR elicited significantly lower ratings of perceived exertion (RPE) than HL-RE during both leg press and knee extension exercises. On average, LLRE+BFR resulted in a RPE score of approximately 4 while an average score of about 7 was observed for HL-RE, during leg press. During knee extension, average scores of approximately 6 and 8 were observed for the LLRE+BFR and HL-RE, respectively.

9. *Are pain levels perceived during LLRE+BFR similar to those perceived during HL-RE?*

LLRE+BFR tended to induce greater pain than HL-RE immediately before sets, meaning that maintaining the restrictive cuffs inflated during the rest interval between

sets diminishes the full recovery from a previous set. On the other hand, when measured immediately after each set, both exercise conditions resulted in similar levels of pain.

10. *Is the 24-h post-exercise delayed-onset muscle soreness response similar between $LLRE+BFR$ and $HL-RE$.*

The 24-h post-exercise muscle soreness response was similar between $LLRE+BFR$ and $HL-RE$. In fact, only a score of 0.8, in a scale from 0 to 10, was detected 24 hours after the $LLRE+BFR$ trial, and a score of 1.3 24 hours after the $HL-RE$ trial.

Research Subquestions

1. *Were participants able to complete the pre-determined standard BFR protocol (4 sets of 30+15+15+15 repetitions at 20% of 1-RM)?*

Participants were able to complete the 4 sets of 30+15+15+15 repetitions at 20% of 1-RM for the $LLRE+BFR$ experimental trial for both leg press and knee extension exercises.

2. *Were participants able to complete the pre-determined high-load resistance exercise protocol (4 sets of 10 repetitions)?*

Participants were able to complete the scheme of 4 sets of 10 repetitions for the leg press exercise but not for the knee extension, unless the load was decreased.

- 3. Is there any difference in exercise volume between leg press and knee extension exercises within the same exercise protocol?*

Exercise volume was slightly greater for leg press compared to knee extension during HL-RE due to the fact that most participants were unable to complete the pre-determined number of repetitions for the latter.

- 4. Were individuals with MS able to tolerate the application of BFR during exercise?*

Most participants were able to tolerate the BFR stimulus during exercise, with only one participant feeling lightheaded immediately post-exercise, which was dissipated following a few minutes of rest.

- 5. Was there any difference in electromyography amplitude when comparing muscles of the right and left legs?*

There were no differences in electromyography amplitude within the vastus medialis or vastus lateralis muscles, during the leg press or knee extension exercise, when comparing right and left legs.

- 6. Was 1-RM testing a reliable method to measure maximum dynamic strength in people living with MS?*

Conflicting findings were observed regarding the reliability of the 1-RM test in people living with MS. Although high reliability scores were observed for the 1-RM test performed in both the leg press and the knee extension exercise, a significant

increase from trial 1 to trial 2 was observed during leg press, potentially due to a learning effect, but not during knee extension.

Hypotheses

1. *Considering the literature suggesting the exercise-induced metabolic response as one of the potential mechanisms contributing for muscle hypertrophy following LLRE+BFR and the several studies reporting similar hypertrophy gains following both LLRE+BFR and HL-RE, it was hypothesized that a similar metabolic response (whole-blood lactate) would be observed between the LLRE+BFR and HL-RE protocols*

This hypothesis was rejected as HL-RE resulted in a greater post-exercise whole-blood lactate accumulation compared to LLRE+BFR.

2. *Myoelectric activity during exercise would be greater during HL-RE in comparison to LLRE+BFR. This hypothesis was based on multiple studies demonstrating smaller myoelectric activity during LLRE+BFR compared to HL-RE.*

This hypothesis was confirmed as LLRE+BFR resulted in greater myoelectric activity than HL-RE for all muscles tested during both leg press and knee extension exercises.

3. *There are also several studies demonstrating that LLRE+BFR and HL-RE may induce similar post-exercise responses. Thus, it was hypothesized that the exercise-induced*

muscle swelling response (muscle thickness and thigh circumference) would be similar between $LLRE+BFR$ and $HL-RE$.

This hypothesis was also confirmed as a similar muscle swelling response was observed following both the $LLRE+BFR$ and $HL-RE$ experimental trials, in the form of increases in muscle thickness and thigh circumference.

- 4. Although only a few studies have directly compared the hormonal stress response following $LLRE+BFR$ and $HL-RE$, considering the higher mechanical stress involved with $HL-RE$, it was hypothesized that a greater hormonal stress (cortisol) response would be observed following $HL-RE$ compared to $LLRE+BFR$.*

This hypothesis was rejected as no significant differences were observed between the $LLRE+BFR$ and $HL-RE$ conditions up to 1 hour post-exercise.

- 5. As the regulation of biomolecular pathways has also been suggested as potential mechanisms through which both $LLRE+BFR$ and $HL-RE$ elicit the positive adaptations, it was hypothesized that similar levels of biomolecular markers of muscle anabolism (mTOR) and catabolism (myostatin) would be observed following $LLRE+BFR$ and $HL-RE$.*

This hypothesis was confirmed as no significant differences were observed between $LLRE+BFR$ and $HL-RE$ for the post-exercise changes in mTOR and myostatin plasma concentrations.

6. *The higher mechanical loads used during HL-RE have been well documented to induce muscle damage after an exercise bout, whereas the current literature is yet to demonstrate that LLRE+BFR induces any muscle damage. Considering the common inflammatory response taking place following damaging exercise, it was hypothesized greater inflammation (interleukin-6) would be observed following HL-RE compared to LLRE+BFR.*

This hypothesis was rejected as no significant differences were observed for the post-exercise interleukin-6 plasma concentration following either LLRE+BFR or HL-RE.

7. *There would be no difference in changes in plasma volume and hematocrit levels between LLRE+BFR and HL-RE. This hypothesis was based on previous literature demonstrating minimal to no changes in plasma volume and hematocrit levels.*

This hypothesis was confirmed as no significant differences were detected between LLRE+BFR and HL-RE for changes in plasma volume and hematocrit levels.

8. *Considering the higher mechanical loads used during HL-RE, it was hypothesized that HL-RE would result in greater ratings of perceived exertion (RPE) compared to LLRE+BFR.*

This hypothesis was confirmed as HL-RE resulted in greater RPE values during both leg press and knee extension exercises in comparison to LLRE+BFR.

9. *Although one would naturally expect HL-RE to result in lower ratings of pain compared to LLRE+BFR, due to the use of lower loads, it should also be considered that the restriction of blood flow may, on the other hand, contribute to increase the ratings of pain during exercise. Therefore, it was hypothesized that LLRE+BFR would result in similar ratings of pain when compared to HL-RE.*

This hypothesis was partially confirmed as similar levels of pain were observed between the LLRE+BFR and HL-RE trials. However, for the levels of pain measured immediately before each set, the LLRE+BFR trial tended to result in greater pain.

10. *As HL-RE is expected to result in greater muscle damage than LLRE+BFR, it was hypothesized that HL-RE would also result in greater ratings of delayed-onset muscle soreness 24 h post-exercise, while LLRE+BFR will not induce any delayed-onset muscle soreness.*

This hypothesis was confirmed as no significant differences existed between LLRE+BFR and HL-RE for the delayed-onset muscle soreness score. Moreover, only small non-significant increases were observed 24 hours following both experimental conditions.

Subhypotheses

1. *Based on fact that participants are resistance untrained, not familiar with LLRE+BFR, and that individuals with MS fatigue more quickly compared to healthy individuals, it was hypothesized that most participants would not be able to complete all the repetitions for the 4 sets of the BFR exercise protocol.*

Participants were able to perform the required number of repetitions for the LLRE+BFR condition during leg press, but not during the last set of the knee extension exercise.

- 2. Also considering the fact that participants are resistance untrained and have a compromised ability to perform high-load resistance exercise for prolonged periods of time, it was hypothesized that most participants would not be able to complete the 10 repetitions of the last 2 sets of the high-load resistance exercise protocol.*

Participants were able to perform the required number of repetitions for the HL-RE condition during leg press, but not during the last set of the knee extension exercise.

- 3. It was hypothesized that greater exercise volume would be observed with the leg press compared to knee extension exercise, as participants may experience greater fatigue during knee extension, which will be performed after the leg press.*

This hypothesis was partially confirmed as participants completed all predetermined number of repetitions for the HL-RE condition during leg press but not during knee extension, resulting in a slightly greater exercise volume being observed during leg press in comparison with knee extension.

- 4. Although unpleasant, considering the lower levels of BFR applied, it was hypothesized that most participants would be able to tolerate the application of BFR during exercise.*

This hypothesis was conformed as, all participants tolerated well the application of BFR during exercise without any apparent adverse effects. Only one participant felt light-headed following $LLRE+BFR$, but fully recovered within a few minutes.

5. *Taking into consideration the studies demonstrating limb asymmetry in people suffering from MS, it was hypothesized that left and right legs would display differences in sEMG amplitude.*

These data did not support the hypothesis that differences in sEMG amplitude would be observed when comparing the right and left legs.

6. *Considering limb asymmetry and the fact that participants not familiar with the technique of resistance training, it was hypothesized that 1-RM would not be a reliable testing method to assess maximum dynamic strength in MS patients.*

This hypothesis was not confirmed as high reliability scores were observed for leg press and knee extension and 1-RM testing. However, it should be considered that a significant 14% mean increase in the 1-RM score was observed during leg press, but not during knee extension.

Clinical Significance

This study was the first to provide scientific evidence that $LLBFR-RE$ may potentially serve as a resistance training modality for people living with MS. These findings have profound relevance for individuals suffering from MS, considering that training at high intensities may potentially increase body temperature leading to a

temporary exacerbation of the symptoms of the disease. Considering, the compromised physical function parameters commonly observed in these individuals, performing HL-RE would be difficult and potentially increase the risk of injury. Finally, the use of higher training loads could also induce muscle damage and impose a further temporary decline in physical function.

This study demonstrated that people living with MS are capable of tolerating and performing LLBFR+RE without any major adverse effects. This study also demonstrated that LLBFR+RE is capable of acutely increasing many of the physiological parameters commonly thought to contribute the skeletal muscle hypertrophic often observed following traditional resistance exercise without BFR, indicating that it may potentially serve as a training alternative to HL-RE for MS patients unable or unwilling to lift heavy loads. The perceptual data from this study also demonstrated that LLBFR+RE requires less muscular exertion compared to HL-RE, and does not cause exaggerated pain during exercise or elevated delayed-onset muscle soreness up to 24 h post-exercise, which altogether makes it more attractive and appealing for people living with MS.

Future Directions

The findings of the current study demonstrated that LLRE+BFR is capable of acutely increasing many of the physiological parameters commonly used to explain the positive adaptations often observed following LLRE+BFR. Additionally, this study also demonstrated that people living with MS are capable of performing a typical LLRE+BFR protocol without any major adverse effects. Lastly, this investigation also demonstrated that LLRE+BFR requires less muscular effort than traditional high-load resistance training.

Therefore, future studies should investigate the long-term effects of low-load resistance training combined with blood flow restriction in individuals with MS on skeletal muscle size and strength levels, as well as parameters of physical function (e.g., mobility, coordination, balance, etc.).

The current investigation demonstrated that low-load resistance training combined with BFR may serve as a potential training alternative to traditional high-load resistance training capable of inducing positive neuromuscular adaptations. Nonetheless, further research is needed to confirm the long-term benefits of this training modality in this clinical population.

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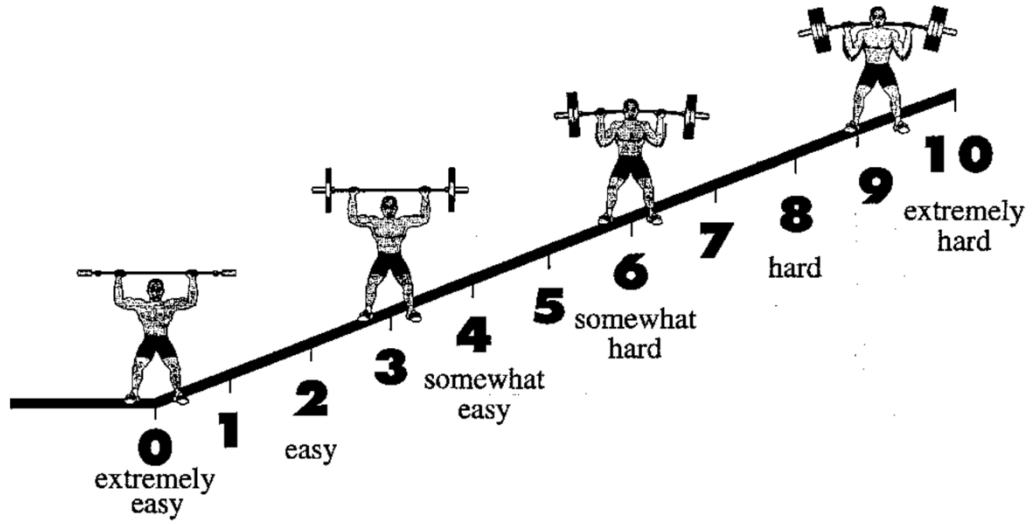
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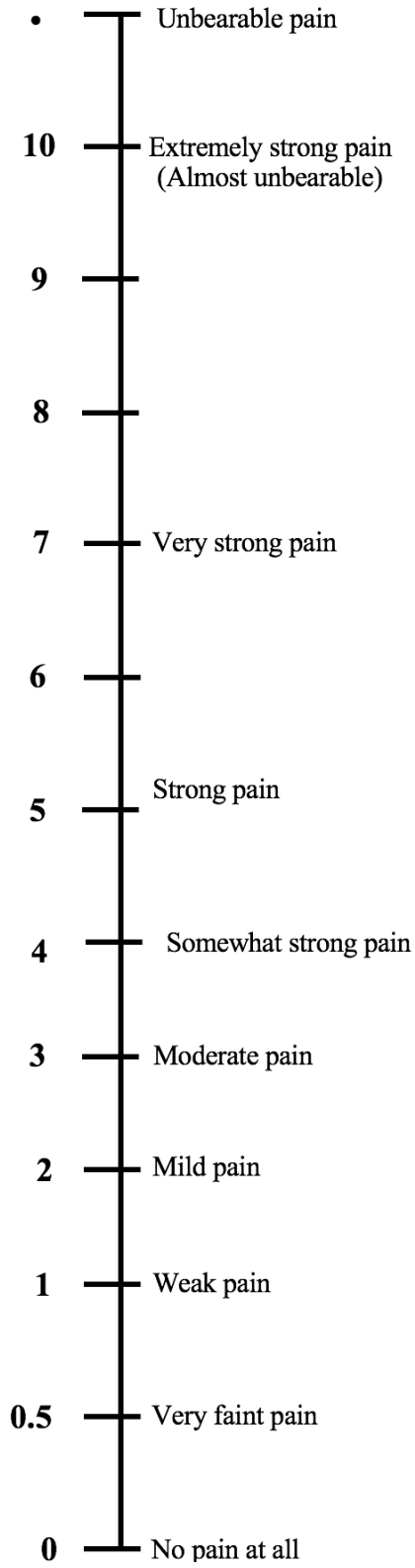
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Appendices

Appendix A: OMNI Scale for Resistance Exercise



Appendix B: Pain Scale



Appendix C: IBR Letter of Approval



Institutional Review Board for the Protection of Human Subjects

Initial Submission – Board Approval

Date: October 9, 2018

IRB#: 9779

To: Michael G Bemben, PhD

Meeting Date: 10/01/2018

Approval Date: 10/08/2018

Expiration Date: 09/30/2019

Study Title: Acute Physiological Responses to Low-Load Resistance Exercise with Blood Flow Restriction Compared to Traditional High-Load Resistance Exercise in Multiple Sclerosis Patients

Reference Number: 682221

Study Status: Active - Open

At its regularly scheduled meeting the IRB reviewed the above-referenced research study. Study documents associated with this submission are listed on page 2 of this letter. To review and/or access the submission forms as well as the study documents approved for this submission, open this study from the *My Studies* option, click to open this study, look under Protocol Items to click on the current *Application*, *Informed Consent* and *Other Study Documents*.

If this study required routing through the Office of Research Administration (ORA), you may not begin your study yet, as per OUHSC Institutional policy, until the contract through ORA is finalized and signed.

As principal investigator of this research study, it is your responsibility to:

- Conduct the research study in a manner consistent with the requirements of the IRB and federal regulations at 45 CFR 46 and/or 21 CFR 50 and 56.
- Request approval from the IRB prior to implementing any/all modifications.
- Promptly report to the IRB any harm experienced by a participant that is both unanticipated and related per IRB Policy.
- Maintain accurate and complete study records for evaluation by the HRPP quality improvement program and if applicable, inspection by regulatory agencies and/or the study sponsor.
- Promptly submit continuing review documents to the IRB upon notification approximately 60 days prior to the expiration date indicated above.

In addition, it is your responsibility to obtain informed consent and research privacy authorization using the currently approved, stamped forms and retain all original, signed forms, if applicable.

If you have questions about this notification or using iRIS, contact the IRB at 405-271-2045 or irb@ouhsc.edu.

Sincerely,

Karen Beckman, MD, Chair
Institutional Review Board

Appendix D: ELISA Kits' Instructions

Interleukin-6

Quantikine® ELISA

Human IL-6 Immunoassay

Catalog Number D6050
S6050
PD6050

For the quantitative determination of human Interleukin 6 (IL-6) concentrations in cell culture supernates, serum, and plasma.

This package insert must be read in its entirety before using this product.
For research use only. Not for use in diagnostic procedures.

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INTRODUCTION

Interleukin 6 (IL-6) is a pleiotropic, α -helical, 22-28 kDa phosphorylated and variably glycosylated cytokine that plays important roles in the acute phase reaction, inflammation, hematopoiesis, bone metabolism, and cancer progression (1-5). Mature human IL-6 is 183 amino acids (aa) in length and shares 39% aa sequence identity with mouse and rat IL-6 (6). Alternative splicing generates several isoforms with internal deletions, some of which exhibit antagonistic properties (7-10). Cells known to express IL-6 include CD8⁺ T cells, fibroblasts, synovocytes, adipocytes, osteoblasts, megakaryocytes, endothelial cells (under the influence of endothelins), sympathetic neurons, cerebral cortex neurons, adrenal medulla chromaffin cells, retinal pigment cells, mast cells, keratinocytes, Langerhans cells, fetal and adult astrocytes, neutrophils, monocytes, eosinophils, colonic epithelial cells, B1 B cells and pancreatic islet beta cells (2, 11-33). IL-6 production is generally correlated with cell activation and is normally kept in control by glucocorticoids, catecholamines, and secondary sex steroids (2). Normal human circulating IL-6 is in the 1 pg/mL range, with slight elevations during the menstrual cycle, modest elevations in certain cancers, and large elevations after surgery (34-38).

IL-6 induces signaling through a cell surface heterodimeric receptor complex composed of a ligand binding subunit (IL-6 R α) and a signal transducing subunit (gp130). IL-6 binds to IL-6 R α , triggering IL-6 R α association with gp130 and gp130 dimerization (39). gp130 is also a component of the receptors for CLC, CNTF, CT-1, IL-11, IL-27, LIF, and OSM (40). Soluble forms of IL-6 R α are generated by both alternative splicing and proteolytic cleavage (5). In a mechanism known as trans-signaling, complexes of soluble IL-6 and IL-6 R α elicit responses from gp130-expressing cells that lack cell surface IL-6 R α (5). Trans-signaling enables a wider range of cell types to respond to IL-6, as the expression of gp130 is ubiquitous, while that of IL-6 R α is predominantly restricted to hepatocytes, monocytes, and resting lymphocytes (2, 5). Soluble splice forms of gp130 block trans-signaling from IL-6/IL-6 R α but not from other cytokines that use gp130 as a co-receptor (5, 41).

IL-6, along with TNF- α and IL-1, drives the acute inflammatory response. IL-6 is almost solely responsible for fever and the acute phase response in the liver, and it is important in the transition from acute inflammation to either acquired immunity or chronic inflammatory disease (1-5). When dysregulated, it contributes to chronic inflammation in conditions such as obesity, insulin resistance, inflammatory bowel disease, arthritis, and sepsis (2, 5). IL-6 modulates bone resorption and is a major effector of inflammatory joint destruction in rheumatoid arthritis through its promotion of Th17 cell development and activity (1). It contributes to atherosclerotic plaque development and destabilization as well as the development of inflammation-associated carcinogenesis (1, 2). IL-6 can also function as an anti-inflammatory molecule, as in skeletal muscle where it is secreted in response to exercise (2). In addition, it enhances hematopoietic stem cell proliferation and the differentiation of memory B cells and plasma cells (42).

The Quantikine® Human IL-6 Immunoassay is a 4.5 hour solid phase immunoassay designed to measure human IL-6 in cell culture supernates, serum, and plasma. It contains *E. coli*-expressed recombinant human IL-6, and antibodies raised against the recombinant protein. Natural human IL-6 showed dose-response curves that were parallel to the standard curves obtained using the Quantikine® kit standards, indicating that this kit can be used to determine relative levels of natural human IL-6.

www.RnDSystems.com

It has been observed in our laboratories that the measurement of IL-6 is insensitive to the addition of the recombinant form of the IL-6 soluble receptor. Therefore it is probable that experimental sample measurements reflect the total amount of IL-6 present, i.e., the total amount of free IL-6 plus the amount of IL-6 initially bound to soluble receptors, if any are present in the samples. High levels of high-affinity autoantibodies to IL-6 in the serum of some blood donors have been reported (36, 37). Such autoantibodies have the potential to interfere with the measurement of IL-6 by ELISA immunoassays.

PRINCIPLE OF THE ASSAY

This assay employs the quantitative sandwich enzyme immunoassay technique. A monoclonal antibody specific for human IL-6 has been pre-coated onto a microplate. Standards and samples are pipetted into the wells and any IL-6 present is bound by the immobilized antibody. After washing away any unbound substances, an enzyme-linked polyclonal antibody specific for human IL-6 is added to the wells. Following a wash to remove any unbound antibody-enzyme reagent, a substrate solution is added to the wells and color develops in proportion to the amount of IL-6 bound in the initial step. The color development is stopped and the intensity of the color is measured.

LIMITATIONS OF THE PROCEDURE

- FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.
- The kit should not be used beyond the expiration date on the kit label.
- Do not mix or substitute reagents with those from other lots or sources.
- It is important that the calibrator diluent selected for the standard curve be consistent with the samples being assayed.
- If samples generate values higher than the highest standard, dilute the samples with the appropriate calibrator diluent and repeat the assay. If cell culture supernate samples require larger dilutions, perform an intermediate dilution with culture media and the final dilution with the appropriate calibrator diluent.
- Any variation in diluent, operator, pipetting technique, washing technique, incubation time or temperature, and kit age can cause variation in binding.
- Variations in sample collection, processing, and storage may cause sample value differences.
- This assay is designed to eliminate interference by other factors present in biological samples. Until all factors have been tested in the Quantikine® Immunoassay, the possibility of interference cannot be excluded.

TECHNICAL HINTS

- When mixing or reconstituting protein solutions, always avoid foaming.
- To avoid cross-contamination, change pipette tips between additions of each standard level, between sample additions, and between reagent additions. Also, use separate reservoirs for each reagent.
- To ensure accurate results, proper adhesion of plate sealers during incubation steps is necessary.
- When using an automated plate washer, adding a 30 second soak period following the addition of Wash Buffer, and/or rotating the plate 180 degrees between wash steps may improve assay precision.
- Substrate Solution should remain colorless until added to the plate. Keep Substrate Solution protected from light. Substrate Solution should change from colorless to gradations of blue.
- Stop Solution should be added to the plate in the same order as the Substrate Solution. The color developed in the wells will turn from blue to yellow upon addition of the Stop Solution. Wells that are green in color indicate that the Stop Solution has not mixed thoroughly with the Substrate Solution.

For research use only. Not for use in diagnostic procedures.

MATERIALS PROVIDED & STORAGE CONDITIONS

Store the unopened kit at 2-8 °C. Do not use past kit expiration date.

PART	PART #	CATALOG #	CATALOG #	DESCRIPTION	STORAGE OF OPENED/ RECONSTITUTED MATERIAL
Human IL-6 Microplate	890045	1 plate	6 plates	96 well polystyrene microplate (12 strips of 8 wells) coated with a monoclonal antibody specific for human IL-6.	Return unused wells to the foil pouch containing the desiccant pack. Reseal along entire edge of zip-seal. May be stored for up to 1 month at 2-8 °C.*
Human IL-6 Standard	890047	1 vial	6 vials	Recombinant human IL-6 in a buffered protein base with preservatives; lyophilized. Refer to the vial label for reconstitution volume.	Aliquot and store for up to 1 month at ≤ -20 °C in a manual defrost freezer.* Avoid repeated freeze-thaw cycles.
Human IL-6 Conjugate	890046	1 vial	6 vials	21 mL/vial of a polyclonal antibody specific for human IL-6 conjugated to horseradish peroxidase with preservatives.	May be stored for up to 1 month at 2-8 °C.*
Assay Diluent RD1W	895117	1 vial	6 vials	11 mL/vial of a buffered protein base with preservatives.	
Calibrator Diluent RDST	895175	1 vial	6 vials	21 mL/vial of a buffered protein base with preservatives. For cell culture supernate samples.	
Calibrator Diluent RD6F	895018	1 vial	6 vials	21 mL/vial of animal serum with preservatives. For serum/plasma samples.	
Wash Buffer Concentrate	895003	1 vial	6 vials	21 mL/vial of a 25-fold concentrated solution of buffered surfactant with preservative. May turn yellow over time.	
Color Reagent A	895000	1 vial	6 vials	12 mL/vial of stabilized hydrogen peroxide.	
Color Reagent B	895001	1 vial	6 vials	12 mL/vial of stabilized chromogen (tetramethylbenzidine).	
Stop Solution	895032	1 vial	6 vials	6 mL/vial of 2 N sulfuric acid.	
Plate Sealers	N/A	4 strips	24 strips	Adhesive strips.	

* Provided this is within the expiration date of the kit.

D6050 contains sufficient materials to run an ELISA on one 96 well plate. S6050 (SixPak) contains sufficient materials to run ELISAs on six 96 well plates.

This kit is also available in a PharmPak (R&D Systems®, Catalog # PD6050). PharmPaks contain sufficient materials to run ELISAs on 50 microplates. Specific vial counts of each component may vary. Refer to the PharmPak Contents section for specific vial counts.

www.RnDSystems.com

3

PHARMPAK CONTENTS

Each PharmPak contains reagents sufficient for the assay of 50 microplates (96 wells/plate). The package inserts supplied are the same as those supplied in the single kit packs and because of this, a few minor differences related to the number of reagents and their container sizes should be noted.

• Sufficient material is supplied to perform at least 50 standard curves; reuse of each vial may be required. The number of vials, and the number of standard curves obtained per vial will vary with the analyte.

• Wash Buffer 25X Concentrate is bulk packed in 125 mL bottles containing 100 mL, and not in the glass vials described in the package insert. **Note:** Additional wash buffer is available for purchase (R&D Systems®, Catalog # W1A126).

The reagents provided in this PharmPak are detailed below.

PART	PART #	QUANTITY
Human IL-6 Microplate	890045	50 plates
Human IL-6 Conjugate	890046	50 vials
Human IL-6 Standard	890047	25 vials
Calibrator Diluent RDST	895175	50 vials
or		
Calibrator Diluent RD6F	895018	50 vials
Assay Diluent RD1W	895117	50 vials
Color Reagent A	895000	50 vials
Color Reagent B	895001	50 vials
Wash Buffer Concentrate, 25X	895126	9 bottles
Stop Solution	895032	50 vials
Plate sealers	N/A	100 sheets
Package inserts	749909	2 booklets

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OTHER SUPPLIES REQUIRED

- Microplate reader capable of measuring absorbance at 450 nm, with the correction wavelength set at 540 nm or 570 nm.
- Pipettes and pipette tips.
- Deionized or distilled water.
- 500 mL graduated cylinder.
- Squirter bottle, manifold dispenser, or automated microplate washer.
- Test tubes for dilution of standards.
- Human IL-6 Controls (optional; R&D Systems®, Catalog # QC01-1).

PRECAUTIONS

Calibrator Diluent RD6F contains sodium azide which may react with lead and copper plumbing to form explosive metallic azides. Flush with large volumes of water during disposal. The Stop Solution provided with this kit is an acid solution.

Some components in this kit contain a preservative which may cause an allergic skin reaction. Avoid breathing mist.

Color Reagent B may cause skin, eye, and respiratory irritation. Avoid breathing fumes.

Wear protective gloves, clothing, eye, and face protection. Wash hands thoroughly after handling. Refer to the SDS on our website prior to use.

SAMPLE COLLECTION & STORAGE

The sample collection and storage conditions listed below are intended as general guidelines. Sample stability has not been evaluated.

Cell Culture Supernates - Remove particulates by centrifugation and assay immediately or aliquot and store samples at ≤ -20 °C. Avoid repeated freeze-thaw cycles.

Serum - Use a serum separator tube (SST) and allow samples to clot for 30 minutes at room temperature before centrifugation for 15 minutes at 1000 x g. Remove serum and assay immediately or aliquot and store samples at ≤ -20 °C. Avoid repeated freeze-thaw cycles.

Plasma - Collect plasma using EDTA, heparin, or citrate as an anticoagulant. Centrifuge for 15 minutes at 1000 x g within 30 minutes of collection. Assay immediately or aliquot and store samples at ≤ -20 °C. Avoid repeated freeze-thaw cycles.

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REAGENT PREPARATION

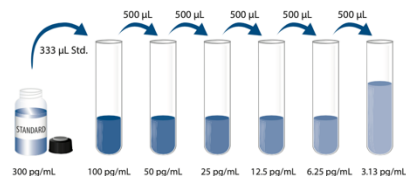
Bring all reagents to room temperature before use.

Wash Buffer - If crystals have formed in the concentrate, warm to room temperature and mix gently until the crystals have completely dissolved. Add 20 mL of Wash Buffer Concentrate to 480 mL of deionized or distilled water to prepare 500 mL of Wash Buffer.

Substrate Solution - Color Reagents A and B should be mixed together in equal volumes within 15 minutes of use. Protect from light. 200 µL of the resultant mixture is required per well.

Human IL-6 Standard - Refer to the vial label for reconstitution volume. Reconstitute the Human IL-6 Standard with Calibrator Diluent RDST (for cell culture supernate samples) or Calibrator Diluent RD6F (for serum/plasma samples). This reconstitution produces a stock solution of 300 pg/mL. Allow the standard to sit for a minimum of 15 minutes with gentle agitation prior to making dilutions.

Pipette 667 µL of Calibrator Diluent RDST (for cell culture supernate samples) or Calibrator Diluent RD6F (for serum/plasma samples) into the 100 pg/mL tube. Pipette 500 µL of the appropriate calibrator diluent into each remaining tube. Use the stock solution to produce a dilution series (below). Mix each tube thoroughly before the next transfer. The undiluted Human IL-6 Standard (300 pg/mL) serves as the high standard. The appropriate calibrator diluent serves as the zero standard (0 pg/mL).



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ASSAY PROCEDURE

Bring all reagents and samples to room temperature before use. It is recommended that all standards, controls, and samples be assayed in duplicate.

1. Prepare all reagents and working standards as directed in the previous sections.
2. Remove excess microplate strips from the plate frame, return them to the foil pouch containing the desiccant pack, and reseal.
3. Add 100 μ L of Assay Diluent RD1W to each well.
4. Add 100 μ L of standard, control, or samples per well. Cover with the adhesive strip provided. Incubate for 2 hours at room temperature. A plate layout is provided to record standards and samples assayed.
5. Aspirate each well and wash, repeating the process three times for a total of four washes. Wash by filling each well with Wash Buffer (400 μ L) using a squirt bottle, manifold dispenser, or autowasher. Complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against clean paper towels.
6. Add 200 μ L of Human IL-6 Conjugate to each well. Cover with a new adhesive strip. Incubate for 2 hours at room temperature.
7. Repeat the aspiration/wash as in step 5.
8. Add 200 μ L of Substrate Solution to each well. Incubate for 20 minutes at room temperature. **Protect from light.**
9. Add 50 μ L of Stop Solution to each well. The color in the wells should change from blue to yellow. If the color in the wells is green or the color change does not appear uniform, gently tap the plate to ensure thorough mixing.
10. Determine the optical density of each well within 30 minutes, using a microplate reader set to 450 nm. If wavelength correction is available, set to 540 nm or 570 nm. If wavelength correction is not available, subtract readings at 540 nm or 570 nm from the readings at 450 nm. This subtraction will correct for optical imperfections in the plate. Readings made directly at 450 nm without correction may be higher and less accurate.

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CALCULATION OF RESULTS

Average the duplicate readings for each standard, control, and sample and subtract the average zero standard optical density (O.D.).

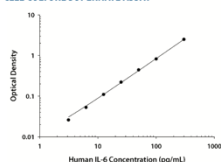
Create a standard curve by reducing the data using computer software capable of generating a four parameter logistic (4-PL) curve fit. As an alternative, construct a standard curve by plotting the mean absorbance for each standard on the y-axis against the concentration on the x-axis and draw a best fit curve through the points on the graph. The data may be linearized by plotting the log of the human IL-6 concentrations versus the log of the O.D. and the best fit line can be determined by regression analysis. This procedure will produce an adequate but less precise fit of the data.

If samples have been diluted, the concentration read from the standard curve must be multiplied by the dilution factor.

TYPICAL DATA

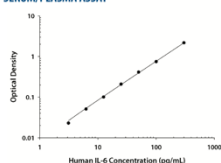
These standard curves are provided for demonstration only. A standard curve should be generated for each set of samples assayed.

CELL CULTURE SUPERNATE ASSAY



(pg/mL)	O.D.	Average	Corrected
0	0.022	0.025	---
3.13	0.050	0.051	0.026
6.25	0.078	0.078	0.053
12.5	0.134	0.135	0.110
25	0.247	0.246	0.221
50	0.472	0.468	0.443
100	0.865	0.850	0.825
300	2.524	2.520	2.495
	2.515		

SERUM/PLASMA ASSAY



(pg/mL)	O.D.	Average	Corrected
0	0.025	0.027	---
3.13	0.049	0.050	0.023
6.25	0.078	0.078	0.051
12.5	0.127	0.128	0.101
25	0.236	0.236	0.209
50	0.438	0.440	0.413
100	0.773	0.776	0.749
300	2.176	2.198	2.171
	2.221		

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PRECISION

Intra-Assay Precision (Precision within an assay)

Three samples of known concentration were tested twenty times on one plate to assess intra-assay precision.

Inter-Assay Precision (Precision between assays)

Three samples of known concentration were tested in twenty separate assays to assess inter-assay precision. Assays were performed by at least three technicians using two lots of components.

CELL CULTURE SUPERNATE ASSAY

Sample	Intra-Assay Precision			Inter-Assay Precision		
	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	15.8	95.6	179	16.4	98.8	188
Standard deviation	0.7	3.0	3.1	0.6	2.5	3.7
CV (%)	4.4	3.1	1.7	3.7	2.5	2.0

SERUM/PLASMA ASSAY

Sample	Intra-Assay Precision			Inter-Assay Precision		
	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	16.8	97.7	186	17.2	101	191
Standard deviation	0.7	1.6	3.8	1.1	3.3	7.2
CV (%)	4.2	1.6	2.0	6.4	3.3	3.8

RECOVERY

The recovery of human IL-6 spiked to three different levels in samples throughout the range of the assay in various matrices was evaluated.

Sample Type	Average % Recovery	Range
Cell culture media (n=5)	98	94-103%
Serum (n=5)	93	86-99%
EDTA plasma (n=5)	95	84-101%
Heparin plasma (n=5)	90	88-98%
Citrate plasma (n=5)	91	82-95%

SENSITIVITY

The minimum detectable dose (MDD) of human IL-6 is typically less than 0.70 pg/mL.

The MDD was determined by adding two standard deviations to the mean O.D. value of twenty zero standard replicates and calculating the corresponding concentration.

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LINEARITY

To assess the linearity of the assay, samples were spiked with high concentrations of human IL-6 in various matrices and diluted with the appropriate calibrator diluent to produce samples with values within the dynamic range of the assay.

	Cell culture media (n=4)	Serum (n=4)	EDTA plasma (n=4)	Heparin plasma (n=4)	Citrate plasma (n=4)
1:2	Average % of Expected: 99 Range (%): 96-101	97 92-100	101 98-105	103 96-109	101 96-106
1:4	Average % of Expected: 100 Range (%): 93-110	101 93-107	104 97-110	106 97-113	105 101-109
1:8	Average % of Expected: 96 Range (%): 92-100	102 96-108	100 86-112	104 93-111	106 101-111
1:16	Average % of Expected: 94 Range (%): 83-108	103 93-111	99 90-110	105 99-107	101 90-114

CALIBRATION

This immunoassay is calibrated against highly purified *E. coli*-expressed recombinant human IL-6 produced at R&D Systems*. The NIBSC/WHO 1st International Standard for IL-6 (89/548), which was intended as a potency standard, was evaluated in this kit. The NIBSC/WHO standard is a CHO cell-derived recombinant human IL-6.

The dose response curve of the International Standard (89/548) parallels the Quantikine® standard curve. To convert sample values obtained with the Quantikine® Human IL-6 kit to approximate NIBSC 89/548 units, use the equation below.

NIBSC (89/548) approximate value (IU/mL) = 0.131 x Quantikine® Human IL-6 value (pg/mL)

SAMPLE VALUES

Serum/Plasma - Forty serum and plasma samples from apparently healthy volunteers were evaluated for the presence of human IL-6 in this assay. Thirty-three samples measured less than the lowest standard, 3.13 pg/mL. Seven samples measured between 3.13 and 12.5 pg/mL. No medical histories were available for the donors used in this study.

Cell Culture Supernates - Human peripheral blood mononuclear cells (1×10^6 cells/mL) were cultured in RPMI supplemented with 10% fetal bovine serum, 50 μ M β -mercaptoethanol, 2 mM L-glutamine, 100 IU/mL penicillin, and 100 IU/mL streptomycin sulfate and stimulated for 1, 3, and 5 days with 10 μ g/mL PHA. Aliquots of the culture supernates were removed on days 1, 3, and 5 and assayed for levels of human IL-6.

Condition	Day 1 (pg/mL)	Day 3 (pg/mL)	Day 5 (pg/mL)
Unstimulated	575	311	660
Stimulated	17,130	17,520	16,340

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SPECIFICITY

This assay recognizes natural and recombinant human IL-6. The factors listed below were prepared at 50 ng/mL in Calibrator Diluent RDST and at 100 ng/mL in Calibrator Diluent RD6F and assayed for cross-reactivity. Preparations of the following factors at 50 ng/mL in a mid-range recombinant human IL-6 control prepared in Calibrator Diluent RDST and 100 ng/mL in a mid-range IL-6 control prepared in Calibrator Diluent RD6F were assayed for interference. No significant cross-reactivity or interference was observed.

Recombinant human:

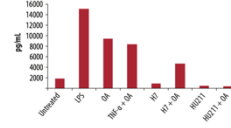
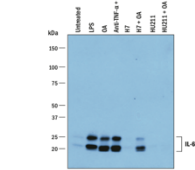
- CNTF IL-7
- G-CSF IL-8
- GM-CSF IL-11
- gp130 IL-12
- IL-1a LIF
- IL-1β LIF R
- IL-2 OSM
- IL-3 TNF-α
- IL-4 TNF-β
- IL-6 Ra
- IL-6 Ra/gp130

Recombinant mouse:

- GM-CSF IL-2
- IL-3 IL-3
- IL-4 IL-4
- IL-5 IL-5
- IL-6 IL-6
- IL-7 IL-7
- IL-11 IL-11
- IL-12 IL-12

Recombinant rat:

- CNTF
- Natural proteins:**
- bovine FGF acidic
- bovine FGF basic
- human PDGF
- porcine PDGF
- human TGF-β1
- porcine TGF-β1,2
- porcine TGF-β2



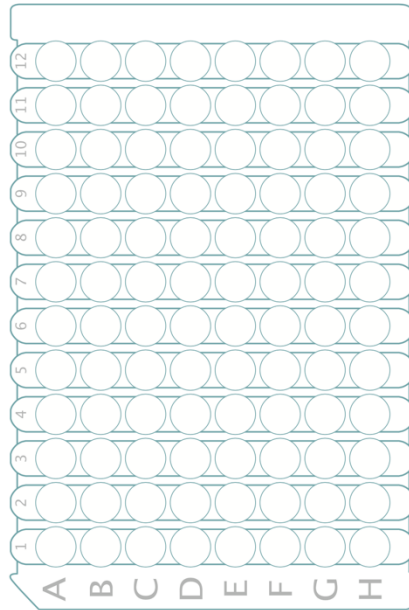
Monocytes were prepared from human PBMCs by adherence to plastic. Adherent monocytes were washed, replated, and allowed to rest for 24 hours. Pretreatments were for 30 minutes: neutralizing anti-human TNF-α (R&D Systems®, Catalog # MAB610) at 5.0 μg/mL, H7 serine kinase inhibitor (Tocris, Catalog # 0542) at 10 μM, or HU211 NFκB inhibitor (Tocris, Catalog # 2861) at 10 μM. Following the pretreatment, 500 ng/mL LPS or 30 ng/mL okadaic acid (OA, Tocris, Catalog # 1136) was added for 20 hours as indicated. Conditioned media was tested in the Quantikine® ELISA, resolved by SDS-PAGE, transferred to a PVDF membrane, and immunoblotted with the detection antibody used in this kit. The immunoprecipitation/Western Blot shows direct correlation with the ELISA value for these samples.

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PLATE LAYOUT

Use this plate layout to record standards and samples assayed.



NOTES

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Cortisol



Instructions for Use

Cortisol ELISA RUO

RUO

REF EIA-1887R

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Cortisol ELISA RUO EIA-1887R

Version 12.1
Effective, May 2019

(V12.0_2019/05 - v)

Please use only the valid version of the Instructions for Use provided with the kit.

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Cortisol ELISA RUO EIA-1887R

For Research Use Only

Not for use in diagnostic procedures

1 INTRODUCTION

1.1 Intended Use

The DRG Cortisol ELISA is an enzyme immunoassay for the quantitative measurement of Cortisol in serum and plasma (EDTA-, heparin- or citrate plasma).

2 PRINCIPLE OF THE TEST

The DRG Cortisol ELISA Kit is a solid phase enzyme-linked immunosorbent assay (ELISA), based on the principle of competitive binding. The microtiter wells are coated with a monoclonal antibody directed towards an antigenic site on the Cortisol molecule. Endogenous Cortisol of a sample competes with a Cortisol-horse radish peroxidase conjugate for binding to the coated antibody. After incubation the unbound conjugate is washed off. The amount of bound peroxidase conjugate is inversely proportional to the concentration of Cortisol in the sample. After addition of the substrate solution, the intensity of color developed is inversely proportional to the concentration of Cortisol in the sample.

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Cortisol ELISA RUO EIA-1887R

3 WARNINGS AND PRECAUTIONS

1. This kit is for research use only. For professional use only.
2. All reagents of this test kit which contain human serum or plasma have been tested and confirmed negative for HIV III, HBsAg and HCV by FDA approved procedures. All reagents, however, should be treated as potential biohazards in use and for disposal.
3. Before starting the assay, read the instructions completely and carefully. Use the valid version of instructions for use provided with the kit. Be sure that everything is understood.
4. The microplate contains snap-off strips. Unused wells must be stored at 2 °C - 8 °C in the sealed foil pouch and used in the frame provided.
5. Pipetting of samples and reagents must be done as quickly as possible and in the same sequence for each step.
6. Use reservoirs only for single reagents. This especially applies to the substrate reservoirs. Using a reservoir for dispensing a substrate solution that had previously been used for the conjugate solution may turn solution colored. Do not pour reagents back into vials as reagent contamination may occur.
7. Mix the contents of the microplate wells thoroughly to ensure good test results. Do not reuse microwells.
8. Do not let wells dry during assay; add reagents immediately after completing the rinsing steps.
9. Allow the reagents to reach room temperature (21 °C - 26 °C) before starting the test. Temperature will affect the absorbance readings of the assay. However, values for the samples will not be affected.
10. Never pipet by mouth and avoid contact of reagents and specimens with skin and mucous membranes.
11. Do not smoke, eat, drink or apply cosmetics in areas where specimens or kit reagents are handled.
12. Wear disposable latex gloves when handling specimens and reagents. Microbial contamination of reagents or specimens may give false results.
13. Handling should be done in accordance with the procedures defined by an appropriate national biohazard safety guideline or regulation.
14. Do not use reagents beyond expiry date as shown on the kit labels.
15. All indicated volumes have to be performed according to the protocol. Optimal test results are only obtained when using calibrated pipettes and microtiterplate readers.
16. Do not mix or use components from kits with different lot numbers. It is advised not to exchange wells of different plates even of the same lot. The kits may have been shipped or stored under different conditions and the binding characteristics of the plates may result slightly different.
17. Avoid contact with Stop Solution containing 0.5 M H₂SO₄. It may cause skin irritation and burns.
18. Some reagents contain Proclin 300, BND and/or MIT as preservatives. In case of contact with eyes or skin, flush immediately with water.
19. TMB substrate has an irritant effect on skin and mucosa. In case of possible contact, wash eyes with an abundant volume of water and skin with soap and abundant water. Wash contaminated objects before reusing them. If inhaled, take the person to open air.
20. Chemicals and prepared or used reagents have to be treated as hazardous waste according to the national biohazard safety guideline or regulation.
21. For information on hazardous substances included in the kit please refer to Safety Data Sheets. Safety Data Sheets for this product are available upon request directly from DRG.

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4 REAGENTS**4.1 Reagents provided**

- Microtiter wells.** 12 x 8 (break apart) strips, 96 wells; Wells coated with anti-Cortisol antibody (monoclonal).
- Standard (Standard 0-6).** 7 vials, 1 mL, ready to use; Concentrations: 0, 20, 50, 100, 200, 400, 800 ng/mL, thus corresponding to 0, 55.2, 138, 276, 552, 1104, 2208 nmol/L. Conversion factor: 1 ng/mL = 2.76 nmol/L. Contains non-mercury preservative.
- Enzyme Conjugate.** 1 vial, 25 mL, ready to use, Cortisol conjugated to horseradish peroxidase; Contains non-mercury preservative.
- Substrate Solution.** 1 vial, 14 mL, ready to use, Tetramethylbenzidine (TMB).
- Stop Solution.** 1 vial, 14 mL, ready to use, contains 0.5 M H₂SO₄. Avoid contact with the stop solution. It may cause skin irritations and burns.
- Wash Solution.** 1 vial, 30 mL (40X concentrated). See "Reagent Preparation".

Note: Additional Standard 0 for sample dilution is available upon request.

4.2 Materials required but not provided

- A microtiter plate calibrated reader (450 ± 10 nm) (e.g. the DRG Instruments Microtiter Plate Reader).
- Calibrated variable precision micropipettes.
- Absorbent paper.
- Distilled or deionized water
- Timer
- Graph paper or software for data reduction

4.3 Storage Conditions

When stored at 2 °C - 8 °C unopened reagents will retain reactivity until expiration date. Do not use reagents beyond this date. Opened reagents must be stored at 2 °C - 8 °C. Microtiter wells must be stored at 2 °C - 8 °C. Once the foil bag has been opened, care should be taken to close it tightly again. Opened kits retain activity for 5 weeks if stored as described above.

4.4 Reagent Preparation

Bring all reagents and required number of strips to room temperature prior to use.

Wash Solution

Add deionized water to the 40X concentrated Wash Solution. Dilute 30 mL of concentrated Wash Solution with 1170 mL deionized water to a final volume of 1200 mL. The diluted Wash Solution is stable for 2 weeks at room temperature.

4.5 Disposal of the Kit

The disposal of the kit must be made according to the national regulations. Special information for this product is given in the Material Safety Data Sheet.

4.6 Damaged Test Kits

In case of any severe damage to the test kit or components, DRG has to be informed in writing, at the latest, one week after receiving the kit. Severely damaged single components should not be used for a test run. They have to be stored until a final solution has been found. After this, they should be disposed according to the official regulations.

5 SPECIMEN COLLECTION AND PREPARATION

Serum or plasma (EDTA-, heparin- or citrate plasma) can be used in this assay.

Do not use hemolytic, icteric or lipemic specimens. Please note: Samples containing sodium azide should not be used in the assay.

5.1 Specimen Collection**Serum:**

Collect blood by venipuncture (e.g. Sarstedt Monovette for serum), allow to clot, and separate serum by centrifugation at room temperature. Do not centrifuge before complete clotting has occurred. Sample containing anticoagulant may require increased clotting time.

Plasma:

Whole blood should be collected into centrifuge tubes containing anti-coagulant (e.g. Sarstedt Monovette with the appropriate plasma preparation) and centrifuged immediately after collection.

5.2 Specimen Storage and Preparation

Specimens should be capped and may be stored for up to 7 days at 2 °C - 8 °C prior to assaying. Specimens held for a longer time should be frozen only once at -20 °C prior to assay. Thawed samples should be inverted several times prior to testing.

5.3 Specimen Dilution

If in an initial assay, a specimen is found to contain more than the highest standard, the specimens can be diluted with Standard 0 and reassayed as described in Assay Procedure. For the calculation of the concentrations this dilution factor has to be taken into account.

Example:

- 1:10: 10 µL sample + 90 µL Standard 0 (mix thoroughly)
- 1:100: 10 µL dilution a) 1:10 + 90 µL Standard 0 (mix thoroughly).

6 ASSAY PROCEDURE**6.1 General Remarks**

- All reagents and specimens must be allowed to come to room temperature before use. All reagents must be mixed without foaming.
- Once the test has been started, all steps should be completed without interruption.
- Use new disposal plastic pipette tips for each standard, control or sample in order to avoid cross contamination.
- Absorbance is a function of the incubation time and temperature. Before starting the assay, it is recommended that all reagents are ready, caps removed, all needed wells secured in holder, etc. This will ensure equal elapsed time for each pipetting step without interruption.
- As a general rule the enzymatic reaction is linearly proportional to time and temperature.

6.2 Test Procedure

Each run must include a standard curve.

- Secure the desired number of Microtiter wells in the frame holder.
- Dispense 20 µL of each Standard, Control and samples with new disposable tips into appropriate wells.
- Dispense 200 µL Enzyme Conjugate into each well. Thoroughly mix for 10 seconds. It is important to have a complete mixing in this step.
- Incubate for 60 minutes at room temperature.
- Briskly shake out the contents of the wells. Rinse the wells 3 times with diluted Wash Solution (400 µL per well). Strike the wells sharply on absorbent paper to remove residual droplets. **Important note:** The sensitivity and precision of this assay is markedly influenced by the correct performance of the washing procedure!
- Add 100 µL of Substrate Solution to each well.
- Incubate for 15 minutes at room temperature.
- Stop the enzymatic reaction by adding 100 µL of Stop Solution to each well.
- Determine the absorbance (OD) of each well at 450 ± 10 nm with a microtiter plate reader. It is recommended that the wells be read within 10 minutes after adding the Stop Solution.

6.3 Calculation of Results

- Calculate the average absorbance values for each set of standards, controls and samples.
- Using semi-logarithmic graph paper, construct a standard curve by plotting the mean absorbance obtained from each standard against its concentration with absorbance value on the vertical (Y) axis and concentration on the horizontal (X) axis.
- Using the mean absorbance value for each sample determine the corresponding concentration from the standard curve.
- Automated method: The results in the Instructions for Use have been calculated automatically using a 4-Parameter curve fit. (4 Parameter Robust or 4 Parameter Marquand are the preferred methods.) Other data reduction functions may give slightly different results.
- The concentration of the samples can be read directly from this standard curve. Samples with concentrations higher than that of the highest standard have to be further diluted or reported as > 800 ng/mL. For the calculation of the concentrations this dilution factor has to be taken into account.

6.3.1 Example of Typical Standard Curve

The following data is for demonstration only and cannot be used in place of data generations at the time of assay.

Standard	Optical Units (450 nm)
Standard 0 (0 ng/mL)	2.30
Standard 1 (20 ng/mL)	1.67
Standard 2 (50 ng/mL)	1.24
Standard 3 (100 ng/mL)	0.87
Standard 4 (200 ng/mL)	0.57
Standard 5 (400 ng/mL)	0.35
Standard 6 (800 ng/mL)	0.23

7 EXPECTED NORMAL VALUES

It is strongly recommended that each laboratory should determine its own normal and abnormal values.

8 QUALITY CONTROL

Good laboratory practice requires that controls be run with each calibration curve. A statistically significant number of controls should be assayed to establish mean values and acceptable ranges to assure proper performance. It is recommended to use control samples according to state and federal regulations. The use of control samples is advised to assure the day to day validity of results. The controls and the corresponding results of the QC-Laboratory are stated in the QC certificate added to the kit. The values and ranges stated on the QC sheet always refer to the current kit lot and should be used for direct comparison of the results.

It is also recommended to make use of national or international Quality Assessment programs in order to ensure the accuracy of the results. Employ appropriate statistical methods for analyzing control values and trends. If the results of the assay do not fit to the established acceptable ranges of control materials results should be considered invalid.

In this case, please check the following technical areas: Pipetting and timing devices; photometer, expiration dates of reagents, storage and incubation conditions, aspiration and washing methods. After checking the above mentioned items without finding any error contact your distributor or DRG directly.

9 ASSAY CHARACTERISTICS**9.1 Assay Dynamic Range**

The range of the assay is between 1.3 - 800 ng/mL.

9.2 Specificity of Antibodies (Cross Reactivity)

The following substances were tested for cross reactivity of the assay:

Steroid	Cross reactivity (%)
Cortisol	100
Corticosterone	45
Progesterone	9
Deoxycortisol	< 2
Dexamethasone	< 2
Cortisone	0.9
Estrone	< 0.01
Estrilol	< 0.01
Testosterone	< 0.01

10 LIMITATIONS OF USE

Reliable and reproducible results will be obtained when the assay procedure is performed with a complete understanding of the package insert instruction and with adherence to good laboratory practice. Any improper handling of samples or modification of this test might influence the results.

10.1 Interfering Substances

Hemoglobin (up to 4 mg/mL), Bilirubin (up to 0.5 mg/mL) and Triglyceride (up to 7.5 mg/mL) have no influence on the assay results.

10.2 Drug Interferences

Until today no substances (drugs) are known to us, which have an influence to the measurement of Cortisol in a sample.

10.3 High-Dose-Hook Effect

A High-Dose-Hook Effect is not known for competitive assays.

11 LEGAL ASPECTS**11.1 Reliability of Results**

The test must be performed exactly as per the manufacturer's instructions for use. Moreover the user must strictly adhere to the rules of GLP (Good Laboratory Practice) or other applicable national standards and/or laws. This is especially relevant for the use of control reagents. It is important to always include, within the test procedure, a sufficient number of controls for validating the accuracy and precision of the test.

The test results are valid only if all controls are within the specified ranges and if all other test parameters are also within the given assay specifications. In case of any doubt or concern please contact DRG.

11.2 Liability

Any modification of the test kit and/or exchange or mixture of any components of different lots from one test kit to another could negatively affect the intended results and validity of the overall test. Such modification and/or exchanges invalidate any claim for replacement.

Claims submitted due to customer misinterpretation of laboratory results are also invalid. In the event of any claim, the manufacturer's liability is not to exceed the value of the test kit. Any damage caused to the test kit during transportation is not subject to the liability of the manufacturer.

SYMBOLS USED

Symbol	English
RUO	For research use only
	Consult instructions for use
REF	Catalogue number
LOT	Batch code
	Contains sufficient for tests
	Temperature limit
	Use-by date
	Manufacturer
	Caution
Distributed by	Distributed by
Content	Content
Volume/No.	Volume / No.

(13-May-2019_ia)

Myostatin

Human Myostatin (MSTN) ELISA Kit

Cat No: MBS779358

Standard Curve Range: 0.2ng/ml -8ng/ml

Sensitivity: 0.1ng/ml

Expiration date: six months .

Storage: 2-8°C.

For samples: Serum, plasma, cell culture supernatants, body fluid and tissue homogenate

When stored at 2-8 °C unopened reagents will retain reactivity until expiration date.

Opened reagents must be stored at 2-8 °C.

Read this manual carefully before using. The ELISA kit is based on the principle of double antibody sandwich technology.

And the ELISA kits only be used for research purposes, not for medical diagnosis.

Reagent preparation: Bring all reagents to room temperature before using.

**FOR RESEARCH USE ONLY; NOT FOR THERAPEUTIC OR DIAGNOSTIC APPLICATIONS!
PLEASE READ THROUGH ENTIRE PROCEDURE BEFORE BEGINNING!****Intended Use**

For the quantitative determination of Human Myostatin(MSTN) concentrations in serum, plasma, saliva, urine, tissue homogenate, cell culture supernatants and other biological fluids.

Test Principle

The kit was used to test the level of Human Myostatin(MSTN), based on the principle of double antibody sandwich technology enzyme linked immunosorbent assay (ELISA).

Add Standard and Sample to the wells that pre-coated with objective antibody, then add HRP-Conjugate reagent to form an immune complex, incubation, by incubation and washing, removal of unbound enzyme, and then add the substrate A and B, then the solution will turn blue and finally change into yellow at the effect of acid. The color depth or light was positively correlated with the concentration of Myostatin (MSTN).

Precautions

1. Do not substitute reagents from one kit lot to another. Standard, conjugate and microtiter plates are matched for optimal performance. Use only the reagents supplied by manufacturer.

2. It is highly recommended to use the remaining reagents within 1 month before the deadline. For the expiration date, please refer to the label on the kit box. All components are stable before this expiration date. Do not use kit components beyond their expiration date.

3. Remove all kit reagents from refrigerator and allow them to reach room temperature (20-25°C) before use. Do not use water baths to thaw samples or reagents.

4. Use only deionized or distilled water to dilute reagents.

5. Each step add sample, should use sampler, and often proofread the accuracy to avoid the test error. Use fresh disposable pipette tips for each transfer to avoid contamination.

6. Test should strict accordance with the instructions of the operation, the test results must be determined by the microplate reader.

7. Do not remove microtiter plate from the storage bag until needed. Unused strips should be stored at 2-8°C in their pouch with the desiccant provided.

8. Do not mix acid and sodium hypochlorite solutions.

9. Serum and plasma should be handled as potentially hazardous and capable of transmitting disease. Disposable gloves must be worn during the assay procedure, since no known test method can offer complete assurance that products derived from Rat blood will not transmit infectious agents. Therefore, all blood derivatives should be considered potentially infectious and good laboratory practices should be followed.

10. All samples should be disposed of in a manner that will inactivate viruses.

11. Liquid Waste: Add sodium hypochlorite to a final concentration of 1.0%. The waste should be allowed to stand for a minimum of 30 minutes to inactivate the viruses before disposal.

12. Substrate Solution is easily contaminated. If bluish prior to use, do not use. Substrate B is sensitive to light and avoid prolonged exposure to light.

MATERIALS PROVIDED WITH THE KIT

All reagents provided are stored at 2-8°C. Refer to the expiration date on the label.

	Reagents components	96 determinations	48 determinations
1.	Microelisa stripplate	12*8strips	12*4strips
2.	Standard A	0ng/ml	0ng/ml
3.	Standard B	0.5ng/ml	0.5ng/ml
4.	Standard C	1ng/ml	1ng/ml
5.	Standard D	2ng/ml	2ng/ml
6.	Standard E	4ng/ml	4ng/ml
7.	Standard F	8ng/ml	8ng/ml
8.	Sample Diluent	6.0ml	3.0ml
9.	HRP-Conjugate reagent	10.0ml	5.0ml
10.	20X Wash solution	25ml	15ml
11.	Chromogen Solution A	6.0ml	3.0ml
12.	Chromogen Solution B	6.0ml	3.0ml
13.	Stop Solution	6.0ml	3.0ml
14.	Closure plate membrane	2	2
15.	User manual	1	1
16.	Sealed bags	1	1

Note: Standard (A-F) concentration was followed by: 0ng/ml, 0.5ng/ml, 1ng/ml, 2ng/ml, 4ng/ml, 8ng/ml.

Materials required but not supplied

- 1.37 °C incubator
- Microplate reader capable of measuring absorbance at 450 nm.
- Precision pipettes to deliver 2 ml to 1 ml volumes.
- 100 ml and 1 liter graduated cylinders.
- Distilled water,
- Disposable test tube
- Absorbent paper
- Precision pipettes and disposable tip

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Specimen Requirements

- Serum:** Allow the serum to clot for 10-20 minutes at room temperature. Centrifuge (at 2000-3000 RPM) for 20 minutes. Collect the supernatants carefully. When sediments occurred during storage, centrifugation should be performed again.
 - Blood plasma:** In accordance with the requirements of sample collection, EDTA or sodium citrate should be used as anti coagulation. Add EDTA or sodium citrate and mix them for 10-20 minutes. Centrifuge (at 2000-3000 RPM) for approximately 20 minutes. Collect the supernatants carefully. When sediments occurred during storage, centrifugation should be performed again.
 - Urine:** Collect by sterile tube. Centrifuge (at 2000-3000 RPM) for approximately 20 minutes. Collect the supernatants carefully. When sediments occurred during storage, centrifugation should be performed again.
 - Cell culture supernatant:** Collect by sterile tubes when examining secrete components. Centrifuge (at 2000-3000 RPM) for approximately 20 minutes. Collect the supernatants carefully. When examining the components within the cell, use PBS (PH 7.2-7.4) to dilute cell suspension to the cell concentration of approximately 1 million/ml. Damage cells through repeated freeze-thaw cycles to let out the inside components. Centrifuge (at 2000-3000 RPM) for approximately 20 minutes. Collect the supernatants carefully. When sediments occurred during storage, centrifugation should be performed again.
 - Tissue sample:** Incise sample and weigh up. Add a certain amount of PBS (PH 7.4). Freeze with liquid nitrogen immediately for later use. Thaw the sample and keep it at 2-8°C. Add a certain amount of PBS (PH 7.4) and then homogenize the sample thoroughly by hand or homogenizer. Centrifuge (at 2000-5000 RPM) for approximately 20 minutes. Collect the supernatants carefully. Aliquot and keep one for examination and freeze the others for later use.
- Note: 1. Samples to be used within 5 days may be stored at 2-8°C, otherwise samples must be stored at -20°C (≤1month) or -80°C (≤2 months) to avoid loss of bioactivity and avoid contamination.
- Sample hemolysis will influence the result, so the samples should be centrifuged adequately and no hemolysis or granule was allowed.
 - When performing the assay, bring samples to room temperature.
- Samples containing NaN₃ can't be tested as it inhibits the activity of Horse Radish Peroxidase (HRP).
- After collecting the sample, extraction should be immediately carried out in accordance with related documents. After extraction, experiment should be conducted immediately as well. Otherwise, keep the sample at -20°C. Avoid repeated freeze-thaw cycles.

Washed plate method

- Hand-washed plate method: get rid of the liquid within the ELISA plate; in the experimental bench paved a few layers of absorbent paper, put hard the ELISA plate several times downward; the diluted washing solution at least 0.35ml inject into the well, soaking 1-2 minutes. Repeat this process several times as needed.
- Automatic plate washing: If you have automatic washing machine, Should be skilled use, and then used in the formal experiment process.

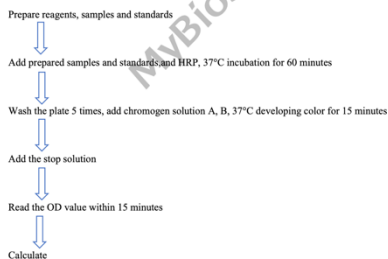
Assay procedure

- Prepare all reagents before starting assay procedure. It is recommended that all Standards and Samples be added in duplicate to the Microelisa Stripplate.

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- Add standard. Set Standard wells, testing sample wells. Add standard 50µl to standard well.
- Add Sample: ① Add Sample 10µl to testing sample well, then add sample diluent 40µl to testing sample well; Blank well doesn't add anything.
② Add 100µl of HRP-conjugate reagent to each well(Standard wells and testing sample wells), then cover it with seal plate membrane, gently shake and mix for 60 minutes at 37 ° C incubation.
- Preparation of washing solution: Dilute the washing concentration (20X) with distilled or deionized water for later use.
- Washing by hand: carefully remove the sealing film, drain the liquid, dried up, each well filled with washing solution, put it aside for 1 min then drain the liquid, so repeat 5 times, pat dry. (Automatic washing: Each wells inject into the wash solution 350µL, soak 1min, wash plate 5 times.)
- Color developing: firstly add 50µl chromogen solution A to each wells, then add 50µl chromogen solution B to each well as well. Shake gently to mix up. Incubate for 15 minutes at 37°C, away from light for color developing.
- Stop: Add 50µl Stop Solution to each well to stop the reaction (the blue color changes into yellow immediately at that moment). If the color in the wells is green or the color change does not appear uniform, gently tap the plate to ensure thorough mixing.
- Assay: Take blank well as zero, measure the absorbance (OD) of each well one by one under 450nm wavelength, which should be carried out within 15 minutes after having added the stop solution.
- According to standards' concentrations and the corresponding OD values, to calculate the linear regression equation of the standard curve. Then according to the OD value of samples, calculate the concentration of the corresponding sample. Also can use related application software.

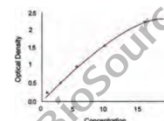
Summary of operating procedures



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Calculation of results

- This standard curve is used to determine the amount in an unknown sample. The standard curve is generated by plotting the average O.D. (450 nm) obtained for each of the six standard concentrations on the vertical (Y) axis versus the corresponding concentration on the horizontal (X) axis.
- First, calculate the mean O.D. value for each standard and sample. All O.D. values, are subtracted by the mean value of the zero standard before result interpretation. Construct the standard curve using graph paper or statistical software.
- To determine the amount in each sample, first locate the O.D. value on the Y-axis and extend a horizontal line to the standard curve. At the point of intersection, draw a vertical line to the X-axis and read the corresponding concentration.
- Any variation in operator, pipetting and washing technique, incubation time or temperature, and kit age can cause variation in result. Each user should obtain their own standard curve.
- Intra-assay CV(%) is less than 10% and Inter-assay CV(%) is less than 15%.
- Standard curve : The following standard curve only for demonstration purposes, each standard curve should be generated with each assay.



- Any variation in operator, pipetting and washing technique, incubation time or temperature, and kit age can cause variation in result. Each user should obtain their own standard curve.
- If samples have been diluted, the concentration read from the standard curve must be multiplied by the dilution factor.
- If specimens generate values higher than the highest standard, dilute the specimens and repeat the assay.


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Mammalian Target of Rapamycin

If you have any question on the order please contact us via: order@bt-laboratory.com;
technical assistance please contact us via: support@bt-laboratory.com
More product visit www.bt-laboratory.com

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Human Mammalian Target of Rapamycin ELISA Kit

USER INSTRUCTION

Cat.No E3693Hu
Standard Curve Range: 0.1ng/ml - 40ng/ml
Sensitivity: 0.04ng/ml
Size: 96 wells

Storage: Store the reagents at 2-8°C. For over 6-month storage refer to the expiration date keep it at -20°C. Avoid repeated thaw cycles. If individual reagents are opened it is recommended that the kit be used within 1 month.

***This product is for research use only, not for use in diagnosis procedures. It's highly recommend to read this instruction entirely before use.**

Precision

Intra-Assay Precision (Precision within an assay) Three samples of known concentration were tested on one plate to assess intra-assay precision.
Inter-Assay Precision (Precision between assays) Three samples of known concentration were tested in separate assays to assess inter-assay precision.
CV(%) = SD/mean x 100
 Intra-Assay: CV<8%
 Inter-Assay: CV<10%

Intended Use

This sandwich kit is for the accurate quantitative detection of human Mammalian Target of Rapamycin (also known as MTOR) in serum, plasma, cell culture supernates, cell lysates, tissue homogenates.

Assay Principle

This kit is an Enzyme-Linked Immunosorbent Assay (ELISA). The plate has been pre-coated with human MTOR antibody. MTOR present in the sample is added and binds to antibodies coated on the wells. And then biotinylated human MTOR Antibody is added and binds to MTOR in the

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sample. Then Streptavidin-HRP is added and binds to the Biotinylated MTOR antibody. After incubation unbound Streptavidin-HRP is washed away during a washing step. Substrate solution is then added and color develops in proportion to the amount of human MTOR. The reaction is terminated by addition of acidic stop solution and absorbance is measured at 450 nm.

Reagent Provided

Components	Quantity
Standard Solution (48ng/ml)	0.5ml x1
Pre-coated ELISA Plate	12 * 8 wells strips x1
Standard Diluent	3ml x1
Streptavidin-HRP	6ml x1
Stop Solution	6ml x1
Substrate Solution A	6ml x1
Substrate Solution B	6ml x1
Wash Buffer Concentrate (25x)	20ml x1
Biotinylated human MTOR Antibody	1ml x1
User Instruction	1
Plate Sealer	2 pcs
Zipper bag	1 pic

Material Required But Not Supplied

- 37°C±0.5°C incubator
- Absorbent paper
- Precision pipettes and disposable pipette tips
- Clean tubes
- Deionized or distilled water
- Microplate reader with 450 ± 10nm wavelength filter

Precautions

- Prior to use, the kit and sample should be warmed naturally to room temperature 30 minutes.
- This instruction must be strictly followed in the experiment.
- Once the desired number of strips has been removed, immediately reseal the bag to protect the remain from deterioration. Cover all reagents when not in use.
- Make sure pipetting order and rate of addition from well-to-well when pipetting reagents.

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- Pipette tips and plate sealer in hand should be clean and disposable to avoid cross-contamination.
- Avoid using the reagents from different batches together.
- Substrate solution B is sensitive to light, don't expose substrate solution B to light for a long time.
- Stop solution contains acid. Please wear eye, hand and skin protection when using this material. Avoid contact of skin or mucous membranes with kit reagent.
- The kit should not be used beyond the expiration date.

Specimen Collection

Serum Allow serum to clot for 10-20 minutes at room temperature. Centrifuge at 2000-3000 RPM for 20 minutes.

Plasma Collect plasma using EDTA or heparin as an anticoagulant. Centrifuge samples for 15 minutes at 2000-3000 RPM at 2 - 8°C within 30 minutes of collection.

Urine Collect by sterile tube. Centrifuge at 2000-3000 RPM for approximately 20 minutes. When collecting pleuroperitoneal fluid and cerebrospinal fluid, please follow the procedures above-mentioned.

Cell Culture Supernatant Collect by sterile tubes when examining secrete components. Centrifuge at 2000-3000 RPM for approximately 20 minutes. Collect the supernatants carefully. When examining the components within the cell, use PBS (pH 7.2-7.4) to dilute cell suspension to the cell concentration of approximately 1 million/ml. Damage cells through repeated freeze-thaw cycles to let out the inside components. Centrifuge at 2000-3000 RPM for approximately 20 minutes.

Tissue and other body fluids Rinse tissues in PBS (pH 7.4) to remove excess blood thoroughly and weigh before homogenization. Mince tissues and homogenize them in PBS (pH7.4) with a glass homogenizer on ice. Thaw at 2-8°C or freeze at -20°C. Centrifuge at 2000-3000 RPM for approximately 20 minutes.

Note

- Sample concentrations should be predicted before being used in the assay. If the sample concentration is not within the range of the standard curve, users must **contact us** to determine the optimal sample for their particular experiments.
- Samples to be used within 5 days should be stored at 2-8°C. Samples should be aliquoted or must be stored at -20°C within 1 month or -80°C within 6 months. Avoid repeated freeze

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thaw cycles.

- Samples should be brought to room temperature before starting the assay.
- Centrifuge to collect sample before use.
- Samples containing NaN₃ can't be tested as it inhibits the activity of Horse Radish Peroxidase (HRP).
- Collect the supernatants carefully. When sediments occurred during storage, centrifugation should be performed again.
- Hemolysis can greatly impact the validity of test results. Take care to minimize hemolysis.

***Sample can't be diluted with this kit. Owing to the material we use to prepare the kit, the sample matrix interference may falsely depress the specificity and accuracy of the assay.**

Reagent Preparation

- All reagents should be brought to room temperature before use.
- **Standard** Reconstitute the 120µl of the standard (48ng/ml) with 120µl of standard diluent to generate a 24ng/ml standard stock solution. Allow the standard to sit for 15 mins with gentle agitation prior to making dilutions. Prepare duplicate standard points by serially diluting the standard stock solution (24ng/ml) 1:2 with standard diluent to produce 12ng/ml, 6ng/ml, 3ng/ml and 1.5ng/ml solutions. Standard diluent serves as the zero standard(0 ng/ml). Any remaining solution should be frozen at -20°C and used within one month. Dilution of standard solutions suggested are as follows:

24ng/ml	Standard No.5	120µl Original Standard + 120µl Standard Diluent
12ng/ml	Standard No.4	120µl Standard No.5 + 120µl Standard Diluent
6ng/ml	Standard No.3	120µl Standard No.4 + 120µl Standard Diluent
3ng/ml	Standard No.2	120µl Standard No.3 + 120µl Standard Diluent
1.5ng/ml	Standard No.1	120µl Standard No.2 + 120µl Standard Diluent



Standard Concentration	Standard No.5	Standard No.4	Standard No.3	Standard No.2	Standard No.1
48ng/ml	24ng/ml	12ng/ml	6ng/ml	3ng/ml	1.5ng/ml

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- **Wash Buffer** Dilute 20ml of Wash Buffer Concentrate 25x into deionized or distilled water to yield 500 ml of 1x Wash Buffer. If crystals have formed in the concentrate, mix gently until the crystals have completely dissolved.

Assay Procedure

1. Prepare all reagents, standard solutions and samples as instructed. Bring all reagents to room temperature before use. The assay is performed at room temperature.
2. Determine the number of strips required for the assay. Insert the strips in the frames for use. The unused strips should be stored at 2-8°C.
3. Add 50µl standard to standard well. **Note:** Don't add antibody to standard well because the standard solution contains biotinylated antibody.
4. Add 40µl sample to sample wells and then add 10µl anti-MTOR antibody to sample wells, then add 50µl streptavidin-HRP to sample wells and standard wells (Not blank control well). Mix well. Cover the plate with a sealer. Incubate 60 minutes at 37°C.
5. Remove the sealer and wash the plate 5 times with wash buffer. Soak wells with at least 0.35 ml wash buffer for 30 seconds to 1 minute for each wash. For automated washing, aspirate all wells and wash 5 times with wash buffer, overfilling wells with wash buffer. Blot the plate onto paper towels or other absorbent material.
6. Add 50µl substrate solution A to each well and then add 50µl substrate solution B to each well. Incubate plate covered with a new sealer for 10 minutes at 37°C in the dark.
7. Add 50µl Stop Solution to each well, the blue color will change into yellow immediately.
8. Determine the optical density (OD value) of each well immediately using a microplate reader set to 450 nm within 10 minutes after adding the stop solution.

Summary

1. Prepare all reagents, samples and standards.
2. Add sample and ELISA reagent into each well. Incubate for 1 hour at 37°C.
3. Wash the plate 5 times.
4. Add substrate solution A and B. Incubate for 10 minutes at 37°C.
5. Add stop solution and color develops.
6. Read the OD value within 10 minutes.

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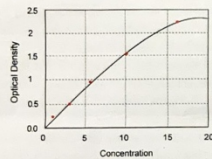
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Calculation of Result

Construct a standard curve by plotting the average OD for each standard on the vertical (Y) axis against the concentration on the horizontal (X) axis and draw a best fit curve through the points on the graph. These calculations can be best performed with computer-based curve-fitting software and the best fit line can be determined by regression analysis.

Typical Data

This standard curve is only for demonstration purposes. A standard curve should be generated with each assay.



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Troubleshooting

Possible Case	Solution
High Background	
<ul style="list-style-type: none"> • Improper washing • Substrate was contaminated • Non-specific binding of antibody • Plate are not be sealing incompletely • Incorrect incubation temperature • Substrate exposed to light prior to use • Contaminated wash buffer 	<ul style="list-style-type: none"> • Increasing duration of soaking steps • Replace. Substrate should be clean and avoid crossed contamination by using the sealer • Replace another purified antibody or blocking buffer • Make sure to follow the instruction strictly • Incubate at room temperature • Keep substrate in a dark place • Use a clean buffers and sterile filter
Weak Signal	
<ul style="list-style-type: none"> • Improper washing • Incorrect incubation temperature • Antibody are not enough • Reagent are contaminated • Pipette are not clean 	<ul style="list-style-type: none"> • Increasing duration of soaking steps • Incubate at room temperature • Increase the concentration of the antibody • Use new one • Pipette should be clean
No Signal	
<ul style="list-style-type: none"> • Reagent are contaminated • Sample prepared incorrectly • Antibody are not enough • Wash buffer contains sodium azide • HRP was not added 	<ul style="list-style-type: none"> • Use new one • Make sure the sample workable/dilution • Increase the antibody concentration • Use a new wash buffer and avoid sodium azide in it • Add HRP according to the instruction
Poor Precision	
<ul style="list-style-type: none"> • Imprecise/ inaccurate pipetting • Incomplete washing of the wells 	<ul style="list-style-type: none"> • Check/ calibrate pipettes • Make sure wells are washed adequately by filling the wells with wash buffer and all residual antibody solutions crossed well before washing.

www.bt-laboratory.com | 1008 Junjiang Inter. Bldg. 228 Ningguo Rd. Yangpu Dist. Shanghai, China
Tel: 86 21 31007137 | Fax: 86 21 65109711 816 | E-mail: save@bt-laboratory.com

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Appendix E: IRB Approved Forms

Consent Form

701G Consent | OUHSC IRB Version Date: 3/12/2019
IRB Number: 9779

Consent Form to Participate in a Research Study University of Oklahoma Health Sciences Center (OUHSC)

Study Title: Acute Physiological Responses to Low-Load Resistance Exercise with Blood Flow Restriction Compared to Traditional High-Load Resistance Exercise in Multiple Sclerosis Patients

Sponsor: National Multiple Sclerosis Society

Principal Investigator: Michael Bemben, PhD

Phone Number: (405) 325-5211

KEY INFORMATION ABOUT THE RESEARCH STUDY

WHY HAVE I BEEN ASKED TO PARTICIPATE IN THIS STUDY?

You are being asked to participate in this research study because you have been diagnosed with multiple sclerosis.

WHY IS THIS STUDY BEING DONE AND HOW LONG WILL IT LAST?

The purpose of this study is to investigate the acute physiological responses to low-load resistance exercise with blood flow restriction and to high-load resistance exercise without blood flow restriction in multiple sclerosis patients. Blood flow restriction resistance exercise involves lifting low weights while the blood flow to the working limbs (arms or legs) is reduced by using specially designed restrictive cuffs. High-load resistance exercise consists of lifting loads that require greater effort from you, but it does not involve restriction of blood flow. We think that you will be in the study for approximately 5 weeks.

WHAT WILL I BE ASKED TO DO IN THIS STUDY?

If you decide to participate in this study, you will be asked to participate in a total of 5 visits (1.5 to 2.5 hours each). The first visit will consist of consenting, answering questionnaires, a general explanation of all procedures related to the study, and familiarization with the strength test. The second visit will consist of assessment of cardiovascular variables and completion of the strength tests designed to measure the maximal strength of your legs. In the third visit, you will have 4 DXA scans, a type of x-ray procedure for assessing your body composition and bone mineral density; the same strength test performed at visit 2; and you will be familiarized with both resistance exercise protocols: low and high-load resistance exercise with and without blood flow restriction, respectively. During visits 4 and 5, one of the two exercise protocols will be performed on each day. Three blood samples will be collected at each of visits 4 and 5.

WHY MIGHT I WANT TO PARTICIPATE IN THIS STUDY?

If you agree to take part in this study, the possible benefits are knowing more about the strength levels of your lower body, and your blood pressure. We hope that the information learned from this study will benefit other patients with this disease in the future.

WHY MIGHT I NOT WANT TO PARTICIPATE IN THIS STUDY?

You may decide that you do not want to participate because you will need to lift heavy weights as part of the study procedures, which may make you feel sore for a couple of days. The collection of blood samples from your arms may cause some discomfort locally and cause bruising. As part of this study, restrictive cuffs will be placed around your thighs and inflated as you perform the exercises, which may cause some discomfort during exercise. You should also be aware that this study will require a total time commitment of approximately 12 hours.



WHAT OTHER OPTIONS ARE THERE?
You may choose not to participate in this study.

HOW WILL PARTICIPATING IN THE STUDY AFFECT ME FINANCIALLY?
There is no additional cost to you if you participate in this study. You will be paid \$300.00 for your time if you finish the study. There is no compensation for completing visit 1. You will be paid \$50 if you successfully complete visit 2 (1RM testing). You will then be paid \$50 to successfully complete visit 3 (DXA and 1RM testing). Following successful completion of visit 4 (Pre and post intervention blood draws and exercise intervention) you will be paid an additional \$100. Similar to visit 4, if you successfully complete visit 5 (Pre and post intervention blood draws and exercise intervention) you will receive the last \$100. If all portions of the testing are successfully completed, you would earn a total of \$300. If you do not complete all the test sessions then you will be paid for the visits (2, 3, 4, or 5) that are completed. The final payment for all visits completed will be done at the end of the 5th visit or after the last successfully completed visit if you decide to drop out.

DETAILED INFORMATION ABOUT THE RESEARCH STUDY

The following pages of the consent form will provide you with more information about this study. Please take your time in reviewing this information and ask the investigator and study team any questions you may have.

HOW MANY PEOPLE WILL TAKE PART IN THE STUDY?
About 30 people will take part in this study nationwide; all will participate at this location.

WHAT IS INVOLVED IN THE STUDY?
The first visit (approximately 1.5 hour) will consist of consenting, questionnaires, and familiarization with the strength tests, as follows:

- Physical Activity Readiness Questionnaire (PAR-Q)
- International Physical Activity Questionnaire – it will assess your physical activity status
- Menstrual Questionnaire – will assess the participant's menstrual history.
- Bone-Specific Physical Activity Questionnaire – it will check for activities that you have performed that may affect your bones.
- Self-Administered Kurtzke Questionnaire – it will check the severity of some of the MS related symptoms.
- Modified Fatigue Scale – it consists of a list of statements that describe how fatigue may affect a person.
- MS History Questionnaire – it will check the history of events related to MS.
- Body weight and height assessment.
- Strength test familiarization – You will be familiarized with the strength test protocol that will be used to measure your low-body maximum strength for both two-leg press and knee extension exercises. The familiarization procedure will consist of a thoughtful explanation of the test and then you will be asked to perform a few repetitions with light loads and then the load will be increased to simulate the increments that will be performed during the actual test.

The second visit (approximately 1.5 hour) will consist of assessment of cardiovascular variables and completion of the strength tests as follows:

- Arterial blood pressure – You will rest for 5 minutes and your arterial blood pressure will be measured twice using a non-invasive electronic blood pressure monitor.
- Ankle-brachial Index – It is a non-invasive procedure that will measure the systolic blood pressure in both arms and legs. It is used as an indirect indicator of peripheral artery disease and it will be used as an inclusion criterion.
- Total Occlusion Pressure: It will be used measure the total amount of pressure necessary to completely occlude (block) blood flow to your legs. This will be used to calculate the amount of pressure to be applied during the blood flow restriction exercise protocol. You will be lay on your back on the testing table and the blood flow restriction cuffs will be placed on both legs. The device will be inflated and deflated several times until the occlusion pressure is reached.

The third visit will consist of you completing 4 DXA scans (a pregnancy test will be performed previous to the scans to rule out pregnancy), the maximal strength test for two-leg press and knee extension, and a familiarization of exercising while wearing the blood flow restriction cuffs:

- DXA scans – Four DXA scans will be performed: 1) one total body scan, to measure your total body composition, bone mineral density, and bone mineral content; 2) Two hip scans, to measure bone mineral density and bone mineral content at the hip; and 3) One spine scan; to measure bone mineral density and bone mineral content at the level of the spine.
- Strength test – You will perform two maximal strength testing protocols on both two-leg press and knee extension exercises. You will begin the test at very light loads and progress until your one repetition maximum is reached within.
- You will perform 2 sets of 15-15 repetitions for two exercises (leg press and knee extension) at 20% of your maximal strength wearing the blood flow restriction cuffs.

The fourth and fifth visits (approximately 2 hours each) will be randomized, and you will have at least 2 weeks of rest in between trials. In these sessions, you will perform a bout of two-leg press and knee extension exercises at 2 different conditions. Venous blood samples (7.5 ml each) will be collected at baseline (pre-exercise), 5 min post-exercise, and 1-hr post-exercise. Muscle thickness (using ultrasound) and thigh circumference (using a tape measure) will be measured at baseline (pre-exercise), immediately post-, 15-min post, and 1-hour post-exercise. Muscle activation will be continuously measured using electrodes on your skin during exercise. The two exercise conditions are:

- Condition 1 – Low-load resistance exercise with blood flow restriction: You will perform 4 sets of 30-15-15-15 repetitions of leg press and knee extension at 20% of your maximal strength wearing an inflatable blood pressure cuff (set at 50% of occlusion) with a minute of rest between sets and 3 min between exercises. The cuffs will remain inflated during the rest intervals between sets, but they will be deflated in the interval between exercises.
- Condition 2 – High-load resistance exercise without blood flow restriction: You will perform 4 sets of 8 to 10 repetitions at 70% of your 1RM with a minute of rest between sets. There will be no restriction of blood flow in this testing condition.

CAN I WITHDRAW FROM THE STUDY?
You can stop participating in this study at any time. However, if you decide to stop participating in the study, we encourage you to talk to the researcher. There will be no adverse consequences if you stop participating in this study. There are no procedures for orderly termination of participation, you just need to inform the research about your decision.

There may be circumstances under which your participation may be terminated by the investigator without your consent.

- He feels that it is in your medical best interest.
- Your condition worsens.
- New information becomes available.
- You fail to follow study requirements.
- The study is stopped by the study sponsor.

WHAT ARE THE RISKS OF THE STUDY?
In addition to the risks described in the Key Information section, you may also be at risk for these side effects. You should discuss these with the researcher and/or your regular doctor. Many side effects go away shortly after the exercise interventions are stopped, but in some cases side effects can be serious or long lasting and permanent. The procedures may involve risks that are currently unforeseeable.

Risks and side effects related to the exercise interventions we are studying include:

- Risks and side effects related to the strength tests, low-load resistance exercise with blood flow restriction, and high-load resistance exercise without blood flow restriction we are studying include:
 - Muscle numbness/tingling during or immediately post-exercise;
 - Bruising up to 24h post-exercise;
 - Muscle soreness up to 48h post-exercise;
 - Lightheaded during or immediately post-exercise
 - Nausea during or immediately post-exercise
 - Discomfort during the occlusion of blood flow

Slight itching in the area where the electrodes are placed for measuring muscle activation.

Regarding ultrasound and thigh circumference:
There are no risks associated with these measures.

Regarding DXA scan:
If you participate in this research protocol you will be exposed to radiation from 4 DXA scans. These scans will be performed for research purposes only, and are not required for your medical care. The amount of radiation exposure that you will receive from these four scans is approximately 1% of the amount of radiation that you are exposed to from natural sources in one year. Risk from radiation exposure is cumulative over your lifetime.

For more information about risks and side effects, ask the researcher.

RADIATION RISKS:
In addition to any radiographic procedures that are being done as part of this research, you may also be exposed to radiation from procedures that are part of your normal care. The number and frequency of these procedures are based on standard clinical practices for a person with your condition; however, your doctor may order an additional radiographic test if he/she thinks it is necessary for your care. The risk from radiation exposure increases over your lifetime as you receive additional exposure to radiation.

REPRODUCTIVE RISKS FOR WOMEN:
If you are a female, you must not be and should not become pregnant nor breast-feed an infant while on this study. Undergoing a particular procedure or treatment involved in this study while you are pregnant or breastfeeding may involve risks to an embryo, fetus, or infant, including birth defects which are currently unforeseeable. In order to reduce your risk of pregnancy, you or your partner should use one or

more of the acceptable methods of birth control listed below, regularly and consistently, while you are in this study.

Acceptable methods of birth control (continuing throughout the study) include:

- An approved oral contraceptive (birth control pill)
- Intra-uterine device (IUD)
- Hormone implants
- Contraceptive injection (Depo-Provera)
- Barrier methods (diaphragm with spermicidal gel or condoms)
- Transdermal contraceptives (birth control patch)
- Vaginal contraception ring (birth control ring)
- Sterilization (tubal ligation, hysterectomy or vasectomy)

If you are already using a method of birth control, you should check with the study doctor to make sure it is considered acceptable for this study. Certain drugs may interact with contraceptive agents and reduce their effectiveness; therefore, you should inform the study doctor of all medications (prescription and over-the-counter) that you are currently taking or begin taking during the study.

IN CASE OF PREGNANCY:
If you become pregnant or suspect that you are pregnant, you should immediately inform the study personnel. If you become pregnant or suspect that you are pregnant while on this study, tell the study doctor immediately; the study doctor will perform a pregnancy test. If pregnancy is confirmed, you may be withdrawn from the study.

TO WHAT EXTENT WILL MY INFORMATION BE KEPT CONFIDENTIAL?
Efforts will be made to keep your personal information confidential. You will not be identifiable by name or description in any reports or publications about this study. We cannot guarantee absolute confidentiality. Your personal information may be disclosed if required by law. You will be asked to sign a separate authorization form for use or sharing of your protected health information.

There are organizations outside the OUHSC that may inspect and/or copy your research records for quality assurance and data analysis. These organizations may include the US Food & Drug Administration and other regulatory agencies, and the National Multiple Sclerosis Society. The OUHSC Human Research Participant Program office, the OUHSC Institutional Review Board, OUHSC Office of Compliance, and other University administrative offices may also inspect and/or copy your research records for these purposes.

Identifiable Private Information:
Your information and samples may be used for future studies without your additional consent. We will remove direct identifiers from your information and assign a code. The key to this code will be kept separately and only the researcher for this study will have access to the code. If your [information/sample] is shared with another investigator for research purposes, they will not have access to the key code and will not be able to re-identify you.

WHAT IF I AM INJURED OR BECOME ILL WHILE PARTICIPATING IN THIS STUDY?
In the case of injury or illness results from this study, emergency medical treatment is available. If you need to contact someone regarding an injury related to the study, please, contact Dr. Michael Bembien at 405-325-5211 or mgbembien@ou.edu.

You or your insurance may be charged for this treatment.

Complications arising as a result of the natural progression of an underlying or pre-existing condition will be billed to you or your insurance. Please check with the investigator or with your insurance company if you have questions.

No other funds have been set aside by the University of Oklahoma Health Sciences Center or by the University of Oklahoma - Norman campus to compensate you in the event of injury, illness, or for other damages related to your event of injury or illness.

WHAT ARE MY RIGHTS AS A PARTICIPANT?

Taking part in this study is voluntary. You may choose not to participate. Refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled.

If you agree to participate and then decide against it, you can withdraw for any reason and leave the study at any time. However, please be sure to discuss leaving the study with the principal investigator or your regular doctor. You may discontinue your participation at any time without penalty or loss of benefits to which you are otherwise entitled.

You have the right to access the medical information that has been collected about you as a part of this research study. However, you may not have access to this medical information until the entire research study has completely finished. You consent to this temporary restriction.

Any clinically relevant results with identifiable information will not be disclosed anywhere.

DO I HAVE ANY OTHER RIGHTS OVER MY DATA?

Depending on where the sponsor for your study is located and other factors, you may have additional rights over your personal data collected in this study. For example, the European Union General Data Protection Regulation (GDPR) and some state privacy laws might apply. If the GDPR applies, generally you may have the following rights:

1. The right to request the information collected to be corrected.
2. The right to withdraw your consent for the use of your personal information at any time.
3. The right, in some circumstances, to receive your personal information in a structured, commonly used and machine-readable format and the right to provide your information to a third party.
4. The right to strict confidentiality of your personal data when it is unshared.
5. The right to limit the use/sharing of your personal information in certain circumstances.
6. The right under some circumstances to request the erasure of your personal data.
7. The right to file a complaint with a privacy protection regulator if you believe any of the rights above have been violated.

You can receive more information regarding these rights in the Privacy Notice for Research Participants, located on the OUHSC Office of Human Research Participant Protection website at www.compliance.ouhsc.edu/hrpp/OUHSC/For-Participants/Privacy-Notice.

If you have any questions and requests, please contact the HRRP Office at 405-271-2045.

WHOM DO I CALL IF I HAVE QUESTIONS, SUGGESTIONS, OR CONCERNS?

If you have questions, concerns, or complaints about the study or have a research-related injury, contact Dr. Michael Bemben at 405-325-5211 or mgbemben@ou.edu.

If you cannot reach the Investigator or wish to speak to someone other than the investigator and for questions about your rights as a research participant, contact the OUHSC Director, Office of Human Research Participant Protection, at 405-271-2045.

SIGNATURE:

By signing this form, you are agreeing to participate in this research study under the conditions described. You have not given up any of your legal rights or released any individual or entity from liability for negligence. You have been given an opportunity to ask questions. You will be given a copy of this consent document.

I agree to participate in this study:

PARTICIPANT SIGNATURE (age ≥18)	Printed Name	Date
SIGNATURE OF PERSON OBTAINING CONSENT	Printed Name	Date

International Physical Activity Questionnaire

INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE (October 2002)

LONG LAST 7 DAYS SELF-ADMINISTERED FORMAT

FOR USE WITH YOUNG AND MIDDLE-AGED ADULTS (15-69 years)

The International Physical Activity Questionnaire (IPAQ) comprises a set of 4 questionnaires. Long (5 activity domains asked independently) and short (4 generic items) versions for use by either telephone or self-administered methods are available. The purpose of the questionnaires is to provide common instruments that can be used to obtain internationally comparable data on health-related physical activity.

Background on IPAQ

The development of an international measure for physical activity commenced in Geneva in 1998 and was followed by extensive reliability and validity testing undertaken across 12 countries (14 sites) during 2000. The final results suggest that these measures have acceptable measurement properties for use in many settings and in different languages, and are suitable for national population-based prevalence studies of participation in physical activity.

Using IPAQ

Use of the IPAQ instruments for monitoring and research purposes is encouraged. It is recommended that no changes be made to the order or wording of the questions as this will affect the psychometric properties of the instruments.

Translation from English and Cultural Adaptation

Translation from English is encouraged to facilitate worldwide use of IPAQ. Information on the availability of IPAQ in different languages can be obtained at www.ipaq.ki.se. If a new translation is undertaken we highly recommend using the prescribed back translation methods available on the IPAQ website. If possible please consider making your translated version of IPAQ available to others by contributing it to the IPAQ website. Further details on translation and cultural adaptation can be downloaded from the website.

Further Developments of IPAQ

International collaboration on IPAQ is on-going and an **International Physical Activity Prevalence Study** is in progress. For further information see the IPAQ website.

More Information

More detailed information on the IPAQ process and the research methods used in the development of IPAQ instruments is available at www.ipaq.ki.se and Booth, M.L. (2000). *Assessment of Physical Activity: An International Perspective*. *Research Quarterly for Exercise and Sport*, 71 (2): s114-20. Other scientific publications and presentations on the use of IPAQ are summarized on the website.

INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE

We are interested in finding out about the kinds of physical activities that people do as part of their everyday lives. The questions will ask you about the time you spent being physically active in the **last 7 days**. Please answer each question even if you do not consider yourself to be an active person. Please think about the activities you do at work, as part of your house and yard work, to get from place to place, and in your spare time for recreation, exercise or sport.

Think about all the **vigorous** and **moderate** activities that you did in the **last 7 days**. **Vigorous** physical activities refer to activities that take hard physical effort and make you breathe much harder than normal. **Moderate** activities refer to activities that take moderate physical effort and make you breathe somewhat harder than normal.

PART 1: JOB-RELATED PHYSICAL ACTIVITY

The first section is about your work. This includes paid jobs, farming, volunteer work, course work, and any other unpaid work that you did outside your home. Do not include unpaid work you might do around your home, like housework, yard work, general maintenance, and caring for your family. These are asked in Part 3.

1. Do you currently have a job or do any unpaid work outside your home?

Yes

No →

Skip to PART 2: TRANSPORTATION

The next questions are about all the physical activity you did in the **last 7 days** as part of your paid or unpaid work. This does not include traveling to and from work.

2. During the **last 7 days**, on how many days did you do **vigorous** physical activities like heavy lifting, digging, heavy construction, or climbing up stairs as **part of your work**? Think about only those physical activities that you did for at least 10 minutes at a time.

___ days per week

No vigorous job-related physical activity →

Skip to question 4

3. How much time did you usually spend on one of those days doing **vigorous** physical activities as part of your work?

___ hours per day

___ minutes per day

4. Again, think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do **moderate** physical activities like carrying light loads as **part of your work**? Please do not include walking.

___ days per week

No moderate job-related physical activity →

Skip to question 6

5. How much time did you usually spend on one of those days doing **moderate** physical activities as part of your work?
- ____ hours per day
____ minutes per day
6. During the **last 7 days**, on how many days did you walk for at least 10 minutes at a time as part of your work? Please do not count any walking you did to travel to or from work.
- ____ days per week
- No job-related walking → **Skip to PART 2: TRANSPORTATION**
7. How much time did you usually spend on one of those days walking as part of your work?
- ____ hours per day
____ minutes per day

PART 2: TRANSPORTATION PHYSICAL ACTIVITY

These questions are about how you traveled from place to place, including to places like work, stores, movies, and so on.

8. During the **last 7 days**, on how many days did you travel in a motor vehicle like a train, bus, car, or tram?
- ____ days per week
- No traveling in a motor vehicle → **Skip to question 10**
9. How much time did you usually spend on one of those days traveling in a train, bus, car, tram, or other kind of motor vehicle?
- ____ hours per day
____ minutes per day

Now think only about the **bicycling** and **walking** you might have done to travel to and from work, to do errands, or to go from place to place.

10. During the **last 7 days**, on how many days did you bicycle for at least 10 minutes at a time to go from place to place?
- ____ days per week
- No bicycling from place to place → **Skip to question 12**

LONG LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised October 2002.



11. How much time did you usually spend on one of those days to bicycle from place to place?
- ____ hours per day
____ minutes per day
12. During the **last 7 days**, on how many days did you walk for at least 10 minutes at a time to go from place to place?
- ____ days per week
- No walking from place to place → **Skip to PART 3: HOUSEWORK, HOUSE MAINTENANCE, AND CARING FOR FAMILY**
13. How much time did you usually spend on one of those days walking from place to place?
- ____ hours per day
____ minutes per day

PART 3: HOUSEWORK, HOUSE MAINTENANCE, AND CARING FOR FAMILY

This section is about some of the physical activities you might have done in the **last 7 days** in and around your home, like housework, gardening, yard work, general maintenance work, and caring for your family.

14. Think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do vigorous physical activities like heavy lifting, chopping wood, shoveling snow, or digging in the garden or yard?
- ____ days per week
- No vigorous activity in garden or yard → **Skip to question 16**
15. How much time did you usually spend on one of those days doing vigorous physical activities in the garden or yard?
- ____ hours per day
____ minutes per day
16. Again, think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do moderate activities like carrying light loads, sweeping, washing windows, and raking in the garden or yard?
- ____ days per week
- No moderate activity in garden or yard → **Skip to question 18**

LONG LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised October 2002.



17. How much time did you usually spend on one of those days doing **moderate** physical activities in the garden or yard?
- ____ hours per day
____ minutes per day
18. Once again, think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do moderate activities like carrying light loads, washing windows, scrubbing floors and sweeping inside your home?
- ____ days per week
- No moderate activity inside home → **Skip to PART 4: RECREATION, SPORT AND LEISURE-TIME PHYSICAL ACTIVITY**
19. How much time did you usually spend on one of those days doing moderate physical activities inside your home?
- ____ hours per day
____ minutes per day

PART 4: RECREATION, SPORT, AND LEISURE-TIME PHYSICAL ACTIVITY

This section is about all the physical activities that you did in the **last 7 days** solely for recreation, sport, exercise or leisure. Please do not include any activities you have already mentioned.

20. Not counting any walking you have already mentioned, during the **last 7 days**, on how many days did you walk for at least 10 minutes at a time in your leisure time?
- ____ days per week
- No walking in leisure time → **Skip to question 22**
21. How much time did you usually spend on one of those days walking in your leisure time?
- ____ hours per day
____ minutes per day
22. Think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do vigorous physical activities like aerobics, running, fast bicycling, or fast swimming in your leisure time?
- ____ days per week
- No vigorous activity in leisure time → **Skip to question 24**

LONG LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised October 2002.



23. How much time did you usually spend on one of those days doing vigorous physical activities in your leisure time?
- ____ hours per day
____ minutes per day
24. Again, think about only those physical activities that you did for at least 10 minutes at a time. During the **last 7 days**, on how many days did you do moderate physical activities like bicycling at a regular pace, swimming at a regular pace, and doubles tennis in your leisure time?
- ____ days per week
- No moderate activity in leisure time → **Skip to PART 5: TIME SPENT SITTING**
25. How much time did you usually spend on one of those days doing moderate physical activities in your leisure time?
- ____ hours per day
____ minutes per day

PART 5: TIME SPENT SITTING

The last questions are about the time you spend sitting while at work, at home, while doing course work and during leisure time. This may include time spent sitting at a desk, visiting friends, reading or sitting or lying down to watch television. Do not include any time spent sitting in a motor vehicle that you have already told me about.

26. During the **last 7 days**, how much time did you usually spend sitting on a weekday?
- ____ hours per day
____ minutes per day
27. During the **last 7 days**, how much time did you usually spend sitting on a weekend day?
- ____ hours per day
____ minutes per day

This is the end of the questionnaire, thank you for participating.

LONG LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised October 2002.



HIIPA Form

University of Oklahoma Health Sciences Center Research Privacy Form 1
PHI Research Authorization

**AUTHORIZATION TO USE or SHARE
HEALTH INFORMATION THAT IDENTIFIES YOU FOR RESEARCH**
*An Informed Consent Document for Research Participation may also be required.
Form 2 must be used for research involving psychotherapy notes.*

Title of Research Project: **Acute Physiological Responses to Low-Load Resistance Exercise with Blood Flow Restriction Compared to Traditional High-Load Resistance Exercise in Multiple Sclerosis Patients**

Leader of Research Team: **Michael Bemben, PhD.**

Address: **1401 Asp Avenue, Norman, OK, 73019**

Phone Number: **405-325-2717**

If you decide to sign this document, University of Oklahoma Health Sciences Center (OUHSC) researchers may use or share information that identifies you (protected health information) for their research. Protected health information will be called PHI in this document.

PHI To Be Used or Shared. Federal law requires that researchers get your permission (authorization) to use or share your PHI. If you give permission, the researchers may use or share with the people identified in this Authorization any PHI related to this research from your medical records and from any test results. Information used or shared may include all information relating to any tests, procedures, surveys, or interviews as outlined in the consent form; medical records and charts; name, address, telephone number, date of birth, race, government-issued identification numbers, and blood pressure, height, weight, two-leg press strength test, knee extension strength test, electromyography (EMG), blood lactate, hematocrit, ultrasound measures of thigh muscle, DXA scans and the results of the following questionnaires: International Physical Activity Questionnaire, Physical Activity Readiness Questionnaire, menstrual history questionnaire, Bone-Specific Physical Activity Questionnaire, Kurtzke Questionnaire, Modified Fatigue Impact Scale, and the Medical History Questionnaire.

Purposes for Using or Sharing PHI. If you give permission, the researchers may use your PHI to investigate the physiological responses of a single bout of continuous or intermittent blood flow restriction resistance exercise, as well as to compare the differences between genders.

Other Use and Sharing of PHI. If you give permission, the researchers may also use your PHI to develop new procedures or commercial products. They may share your PHI with other researchers, the research sponsor and its agents, the OUHSC Institutional Review Board, auditors and inspectors

¹ Protected Health Information includes all identifiable information relating to any aspect of an individual's health whether past, present or future, created or maintained by a Covered Entity.

IRB Office Use Only
Version 01/06/2016



IRB NUMBER: 9779
IRB APPROVAL DATE: 10/08/2018

**University of Oklahoma Health Sciences Center Research Privacy Form 1
PHI Research Authorization**

who check the research, and government agencies such as the Food and Drug Administration (FDA) and the Department of Health and Human Services (HHS), and when required by law. The researchers may also share your PHI with no one else.

Confidentiality. Although the researchers may report their findings in scientific journals or meetings, they will not identify you in their reports. The researchers will try to keep your information confidential, but confidentiality is not guaranteed. The law does not require everyone receiving the information covered by this document to keep it confidential, so they could release it to others, and federal law may no longer protect it.

YOU UNDERSTAND THAT YOUR PROTECTED HEALTH INFORMATION MAY INCLUDE INFORMATION REGARDING A COMMUNICABLE OR NONCOMMUNICABLE DISEASE.

Voluntary Choice. The choice to give OUHSC researchers permission to use or share your PHI for their research is voluntary. It is completely up to you. No one can force you to give permission. However, you must give permission for OUHSC researchers to use or share your PHI if you want to participate in the research and, if you cancel your authorization, you can no longer participate in this study.

Refusing to give permission will not affect your ability to get routine treatment or health care unrelated to this study from OUHSC.

Canceling Permission. If you give the OUHSC researchers permission to use or share your PHI, you have a right to cancel your permission whenever you want. However, canceling your permission will not apply to information that the researchers have already used, relied on, or shared or to information necessary to maintain the reliability or integrity of this research.

End of Permission. Unless you cancel it, permission for OUHSC researchers to use or share your PHI for their research will never end.

Contacting OUHSC. You may find out if your PHI has been shared, get a copy of your PHI, or cancel your permission at any time by writing to:

Privacy Official or Privacy Board
University of Oklahoma Health Sciences Center University of Oklahoma Health Sciences Center
PO Box 26901 PO Box 26901
Oklahoma City, OK 73190 Oklahoma City, OK 73190

If you have questions, call: (405) 271-2511 or (405) 271-2045.

Access to Information. You have the right to access the medical information that has been collected about you as a part of this research study. However, you may not have access to this medical information until the entire research study is completely finished. You consent to this temporary restriction.

IRB Office Use Only
Version: 01/06/2016

Page 2 of 3

IRB NUMBER: 9779
IRB APPROVAL DATE: 10/08/2018

IRB Office Use Only
Version: 01/06/2016

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IRB NUMBER: 9779
IRB APPROVAL DATE: 10/08/2018

**University of Oklahoma Health Sciences Center Research Privacy Form 1
PHI Research Authorization**

Giving Permission. By signing this form, you give OUHSC and OUHSC's researchers led by the Research Team Leader permission to share your PHI for the research project listed at the top of this form.

Patient/Participant Name (Print): _____

Signature of Patient-Participant _____ Date _____
or Parent if Participant is a minor

Or

Signature of Legal Representative** _____ Date _____

**If signed by a Legal Representative of the Patient-Participant, provide a description of the relationship to the Patient-Participant and the authority to act as Legal Representative:

OUHSC may ask you to produce evidence of your relationship.

A signed copy of this form must be given to the Patient-Participant or the Legal Representative at the time this signed form is provided to the researcher or his representative.

Medical Clearance



The University of Oklahoma
DEPARTMENT OF HEALTH AND EXERCISE SCIENCE

Multiple Sclerosis Clearance Letter

Date _____

Dear Dr. _____

A patient of yours, _____ would like to participate in a study called, "Acute Physiological Responses to Low-Load Resistance Exercise with Blood Flow Restriction Compared to Traditional High-Load Resistance Exercise in Multiple Sclerosis" which will be conducted at the University of Oklahoma. The goal of this study is to investigate the acute physiological responses of Multiple Sclerosis patients to two different resistance exercise modalities. Each testing/exercise session will be performed at the University of Oklahoma. For the two exercise sessions (visit 3 and visit 4) a nurse will be present to perform the blood draws and will be available if subjects experience light-headedness or begin to feel ill. If this occurs, the subject will then decide if they would like to continue with the study or withdraw from the study.

Your support of our MS research is very much appreciated. To comply with Institutional Review Board policy, we need a letter from you clearing your MS patient for participation in this study. Attached please find a copy of the research protocol and informed consent. Your written approval letter will include the subject's diagnosis, classification of disease and level of disability at study entry, and current medications. This letter indicates that you are aware of the testing procedures and the specific activities this individual will be performing. Participants will be included if they have an expanded disability status score (Ability to Walk section of the Kurtzke questionnaire) less than 6.0 which reflects ambulatory status.

The individual participating in this research study will be advised to contact you if they experience any clinical symptoms between study visits.

Again, we greatly appreciate your support and request, at your earliest convenience, a response to this letter indicating your approval or disapproval of subject participation.

Michael G. Bembem

Michael G. Bembem, Ph.D., Principal Investigator
Department of Health and Exercise Science
College of Arts and Sciences
University of Oklahoma
Norman, OK

Diagnosis: _____ Initials: _____

Disability Status Score: _____ Initials: _____

Current Medications: _____ Medication rationale (e.g., blood pressure): _____

Initials: _____

Additional Comments by Physician:

Please circle as appropriate: **APPROVED**

DISAPPROVED

Name of Physician: _____

Signature of Physician: _____

Date: _____

IRB NUMBER: 9779
IRB APPROVAL DATE: 10/08/2018

IRB NUMBER: 9779
IRB APPROVAL DATE: 10/08/2018

Modified Fatigue Impact Scale

Patient's Code: _____

Date: ____/____/____
month day year

Test#: 1 2 3 4 5 6 7 8

MODIFIED FATIGUE IMPACT SCALE (MFIS)

INSTRUCTIONS

Following is a list of statements that describe how fatigue may affect a person. Fatigue is a feeling of physical tiredness and lack of energy that many people experience from time to time. In medical conditions like MS, feelings of fatigue can occur more often and have a greater impact than usual. Please read each statement carefully, and then **circle the one number** that best indicates how often fatigue has affected you in this way during the **past 4 weeks**. (If you need help in marking your responses, tell the interviewer the number of the best response.) **Please answer every question. You may ask for clarification to explain any words or phrases that you do not understand.**

Because of my fatigue during the past 4 weeks...

	Never	Rarely	Sometimes	Often	Almost Always
1. I have been less alert.	0	1	2	3	4
2. I have had difficulty paying attention for long periods of time.	0	1	2	3	4
3. I have been unable to think clearly.	0	1	2	3	4
4. I have been clumsy and uncoordinated.	0	1	2	3	4
5. I have been forgetful.	0	1	2	3	4
6. I have had to pace myself in my physical activities.	0	1	2	3	4
7. I have been less motivated to do anything that requires physical effort.	0	1	2	3	4
8. I have been less motivated to participate in social activities.	0	1	2	3	4
9. I have been less motivated to do things away from home.	0	1	2	3	4

Date: ____/____/____
Initials: _____
RIS APPROVAL DATE: 10/06/2018

Patient's Code: _____

Date: ____/____/____
month day year

Test#: 1 2 3 4 5 6 7 8

MODIFIED FATIGUE IMPACT SCALE (MFIS)

INSTRUCTIONS

Following is a list of statements that describe how fatigue may affect a person. Fatigue is a feeling of physical tiredness and lack of energy that many people experience from time to time. In medical conditions like MS, feelings of fatigue can occur more often and have a greater impact than usual. Please read each statement carefully, and then **circle the one number** that best indicates how often fatigue has affected you in this way during the **past 4 weeks**. (If you need help in marking your responses, tell the interviewer the number of the best response.) **Please answer every question. You may ask for clarification to explain any words or phrases that you do not understand.**

Because of my fatigue during the past 4 weeks...

	Never	Rarely	Sometimes	Often	Almost Always
1. I have been less alert.	0	1	2	3	4
2. I have had difficulty paying attention for long periods of time.	0	1	2	3	4
3. I have been unable to think clearly.	0	1	2	3	4
4. I have been clumsy and uncoordinated.	0	1	2	3	4
5. I have been forgetful.	0	1	2	3	4
6. I have had to pace myself in my physical activities.	0	1	2	3	4
7. I have been less motivated to do anything that requires physical effort.	0	1	2	3	4
8. I have been less motivated to participate in social activities.	0	1	2	3	4
9. I have been less motivated to do things away from home.	0	1	2	3	4

Date: ____/____/____
Initials: _____
RIS APPROVAL DATE: 10/06/2018

Recruitment Flyer



COLLEGE OF ARTS AND SCIENCES
 DEPARTMENT OF HEALTH
 AND EXERCISE SCIENCE
 The UNIVERSITY of OKLAHOMA

Do you suffer from Multiple Sclerosis?
RESEARCH PARTICIPANTS NEEDED

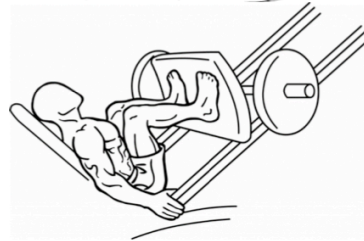
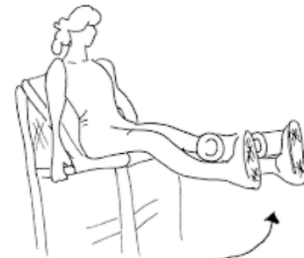
*Acute Physiological Responses to Low-Load Resistance Exercise with Blood Flow Restriction
 Compared to Traditional High-Load Resistance Exercise in Multiple Sclerosis Patients*

What will be measured?

- ✓ Body weight, height, and BMI
- ✓ Body composition: through DXA scans
- ✓ Bone mineral density: through DXA scans
- ✓ Lower body strength: through maximum strength tests
- ✓ Muscle activation: through superficial EMG
- ✓ Blood markers of muscle turnover

Who is eligible to participate?

- ✓ Men and Women diagnosed with Multiple Sclerosis
- ✓ Free from osteomuscular injuries
- ✓ No metal implants at hip or spine



You will be compensated for your time!!!
Principal Investigator: Michael Bemben, PhD

Only 5 visits required

Total time commitment about 9 hours

Tests will take place at the Health and Exercise Science Neuromuscular Lab, University of Oklahoma.

The University of Oklahoma is an equal opportunity institution. IRB 9779

Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804	Name: Eduardo Freitas Email: eduardofreitas@ou.edu Text/Call: (417) 307-8804
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Physical Activity Readiness Questionnaire

Physical Activity Readiness
Questionnaire - PAR-Q
(revised 2002)

PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

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

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

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

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

If
you
answered

YES to one or more questions

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

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

NO to all questions

If you answered NO honestly to all PAR-Q questions, you can be reasonably sure that you can:

- start becoming much more physically active — begin slowly and build up gradually. This is the safest and easiest way to go.
- take part in a fitness appraisal — this is an excellent way to determine your basic fitness so that you can plan the best way for you to live actively. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 144/94, talk with your doctor before you start becoming much more physically active.

DELAY BECOMING MUCH MORE ACTIVE:

- if you are not feeling well because of a temporary illness such as a cold or a fever — wait until you feel better; or
- if you are or may be pregnant — talk to your doctor before you start becoming more active.

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

Informed Use of the PAR-Q: The Canadian Society for Exercise Physiology, Health Canada, and their agents assume no liability for persons who undertake physical activity, and if in doubt after completing this questionnaire, consult your doctor prior to physical activity.

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

NOTE: If the PAR-Q is being given to a person before he or she participates in a physical activity program or a fitness appraisal, this section may be used for legal or administrative purposes.

"I have read, understood and completed this questionnaire. Any questions I had were answered to my full satisfaction."

NAME _____

SIGNATURE _____

DATE _____

SIGNATURE OF PARENT _____

or GUARDIAN (for participants under the age of majority)

WITNESS _____

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



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Supported by:



Health Canada

Santé Canada

continued on other side...

Bone Specific Activity Questionnaire

Bone-Specific Physical Activity Questionnaire (BPAQ)

Sport/Activity	Sport/Activity	Sport/Activity
Aerobics (High Impact)	Resistance Training (Lower body)	*other-Low Impact
Aerobics (Low Impact)	Rollerblading	*other-Moderate Impact
Australian Rules football	Rowing	*other-High Impact
Badminton	Rugby (football)	
Ballet	Running/jogging	
Baseball	Scuba	
Basketball	Shot Put (throwing events)	
Cheerleading	Skate boarding	
Cricket	Skiing	
Cross-country	Soccer (aka football)	
Cycling	Softball	
Dancing	Squash	
Diving	Stairmaster	
Field Hockey	Surfing	
Flag Football	Swimming	
Golf	T-ball	
Gymnastics	Table Tennis	
Horse-riding	Tennis	
Ice Hockey	Touch football	
Ice-skating (Figure/Dance)	Track	
Judo	Triathlon	
Jump rope	Ultimate	
Kung Fu	Volleyball	
Lacrosse	Walking/hiking	
Lawn Bowls	Waterskiing	
Netball	Windsurfing	
Power lifting	Yoga/Pilates	
Racquet ball		

OU Bone Laboratory



IRB NUMBER: 9779
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Self-Administered Kurtzke Scale

TABLE 13-3

Self-Administered Kurtzke

Instructions: Individuals with MS may experience difficulty in a number of different areas. For each of the 8 neurological categories below, please indicate the degree of difficulty (none, minimal, moderate, or severe) that you are experiencing at the present time.

	None	Minimal Difficulty Interferes only Slightly With Function	Moderate Difficulty Interferes Significantly With Function	Severe Difficulty Little or No Function Is Possible
1. Weakness in arm(s) and/or leg(s)	0	1	2	3
2. Tremor, clumsiness, or loss of balance	0	1	2	3
3. Double vision or slurred speech, or difficulty swallowing	0	1	2	3
4. Numbness or difficulty in feeling heat, pain or vibration in any part of the body	0	1	2	3
5. Frequency or urgent urination, awakening to urinate, not emptying the bladder completely, loss of bladder or bowel control, or constipation	0	1	2	3
6. Blurred vision in one or both eyes (even with glasses)	0	1	2	3
7. Difficulty with memory, calculation or reasoning	0	1	2	3
8. Stiffness or jerking of the muscles	0	1	2	3

OVERALL FUNCTION

On the following two pages are a number of statements that might be used to describe the overall function of MS subjects. These statements are arranged in order from least severe (0) to most severe (9.0).

Instructions:

1. First, locate the item that best describes your ability to walk.
 - If you are able to walk without limitations, please choose a statement under the section called "Able to Walk."
 - If you are able to walk only a limited distance, please choose a statement under the section called "Able to Walk Only a Limited Distance."
 - If you require aid(s) or assistance to walk or are unable to walk, please choose a statement under the section called "Aid(s) Required or Unable to Walk."
2. Circle the number of the one statement which best describes your overall condition at the present time.
3. In selecting your answer, refer back to your rating of the 8 neurologic categories listed.

Remember: Choose on *one* of the statements (0-9.0) which follow.

ABLE TO WALK

- 0.0 Essentially normal
- 1.0 Abnormality in *one* of the neurological categories but with no difficulty in function
- 1.5 Abnormality in *more* than one of the neurological categories but with no difficulty in function
- 2.0 Minimal difficulty in one of the neurological categories
- 2.5 Minimal difficulty in two of the neurological categories
- 3.0 Moderate difficulty in one of the neurological categories, able to walk
- 3.5 Moderate difficulty in one of the neurological categories and minimal difficulty in *one or more* of the neurological categories, able to walk



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ABLE TO WALK ONLY A LIMITED DISTANCE	
4.0	Able to walk without aid or rest at least 7 city blocks (500 meters or 1,625 feet) Self-sufficient, up and about some 12 hours a day (Relatively severe difficulty in one neurological category or moderate difficulty in several of the neurological categories)
4.5	Able to walk without aid or rest at least 4 city blocks (300 meters or 975 feet) May need minimal assistance, able to work a full day but may have some limitation of full activity (Relatively severe difficulty in one neurological category or moderate difficulty in several of the neurological categories)
5.0	Able to walk without aid or rest at least 2 ½ city blocks (200 meters or 650 feet) Disability is severe enough to limit full daily activities—for example: to work a full day without job modifications (Very severe difficulty in one of the neurological categories)
5.5	Able to walk without aid or rest at least 1 city block (200 meters or 325 feet) Disability is severe enough to prevent full daily activities (Very severe difficulty in one of the neurological categories or moderate difficulty in several of the neurological categories)
AID(S) REQUIRED OR UNABLE TO WALK	
6.0	Assistance on one side (cane, crutch, brace) is required to walk approximately 1 city block (approximately 100 meters or 325 feet), with or without resting
6.5	Constant assistance on both sides (canes, crutches, braces, walker) is required to walk about 20 meters (65 feet) (Moderate difficulty in more than two neurological categories)
7.0	Unable to walk more than about 5 meters (16 feet) even with aid Essentially restricted to wheelchair Can wheel self in standard wheelchair and can transfer alone Up and about in wheelchair some 12 hours a day (Severe difficulty in more than one neurological category or severe weakness only)
7.5	Unable to take more than a few steps, restricted to wheelchair Can wheel self in standard wheelchair and may need aid to transfer Cannot remain in wheelchair for a full day May require motorized wheelchair (Severe difficulty in more than one neurological category)
8.0	Essentially restricted to bed or chair Propelled by others in wheelchair May be out of bed part of the day Can use arms and able to care for self (Severe difficulty in several neurological categories)
8.5	Essentially restricted to bed much of the day Has limited use of arms Retains some self-care functions (Severe difficulty in several neurological categories)
9.0	Restricted to bed Cannot use arms Can speak, can eat if fed by others (Severe difficulty in several neurological categories)

Source: Scheinberg, I. C. Medical Rehabilitation Research and Training Center for MS, Department of Neurology, Albert Einstein College of Medical, Bronx, New York.

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TABLE 13-4
Self-Administered Kurtzke (French Version)

Symptoms	NONE	MILD	MODERATE	SEVERE
1. Weakness of right arm	0	1	2	3
2. Weakness of left arm	0	1	2	3
3. Weakness of right leg	0	1	2	3
4. Weakness of left leg	0	1	2	3
5. Leg stiffness or deficit at walk	0	1	2	3
6. Tremor	0	1	2	3
7. Clumsiness of arms	0	1	2	3
8. Loss of balance	0	1	2	3
9. Double vision	0	1	2	3
10. Difficulty in speaking and/or swallowing	0	1	2	3
11. Uncontrolled urinary urgency	0	1	2	3
12. Difficulty in urination, incomplete micturition Or bladder emptying	0	1	2	3
13. Constipation	0	1	2	3
14. Loss of control of bladder	0	1	2	3
15. Loss of control of bowel	0	1	2	3
16. Difficulty in feeling a contact	0	1	2	3
17. Difficulty in feeling heat	0	1	2	3
18. Difficulty in feeling pain	0	1	2	3
19. Pain or burning sensation in any part of the body	0	1	2	3
20. Bizarre feeling (pins or needles, constriction) in any part of the body	0	1	2	3
21. Difficulty with memory	0	1	2	3
22. Difficulty with calculations	0	1	2	3
23. Difficulty with reasoning or thinking	0	1	2	3
Level of vision (with glasses)	>7/10 (reading possible)	6/10-4/10 (recognition possible)	3/10 or 2/10 (distinction of forms)	<1/10 (loss of vision)
24. Right eye	0	1	2	3
25. Left eye	0	1	2	3

Source: Verdier-Taillefer MH, Rouillet E, Cesaro P, Alperovitch A. Validation of self-reported neurological disability in multiple sclerosis. *International Journal of Epidemiology* 1994; 23: 148-154.

Menstrual History Questionnaire

<p>Subject ID: _____ Date: _____</p> <p style="text-align: center; font-size: small;">Bone Density Research Laboratory Department of Health and Exercise Science University of Oklahoma</p> <p style="text-align: center; font-size: x-small;">MENSTRUAL HISTORY QUESTIONNAIRE</p> <p style="font-size: x-small;">We are asking you to give us as complete a menstrual history as possible. All information is strictly confidential.</p> <p style="font-size: x-small;">Are you pregnant (circle your response) YES- Do not complete the rest of this form NO- Continue to section A.</p> <p>SECTION A: CURRENT MENSTRUAL STATUS</p> <p>1. Approximately how many menstrual periods have you had during the past 12 months? (please circle what months you have had a period. This means from this time last year to the present month)</p> <p style="text-align: center; font-size: small;">Jan Feb Mar Apr May Jun Jul Aug Sep Oct Nov Dec</p> <p>2. What is the usual length of your menstrual cycle (first day of your period to the next onset of your period)? _____ days. Today is day _____ of your present menstrual cycle.</p> <p>3. What was the date of the onset of your last period?</p> <p>4. When do you expect you next period?</p> <p>5. What is the average length (number of days) of your menstrual flow? _____ days How many of these days do you consider "heavy"? _____ days</p> <p>6. Do you experience cramps during menstruation (dysmenorrhea)? If yes, how many days does this last?</p> <p>7. Do you experience symptoms of premenstrual syndrome (i.e., weight gain, increased eating, depression, headaches, anxiety, breast tenderness)? If yes, please list the symptoms.</p> <div style="text-align: right; font-size: x-small;"> IRB NUMBER: 9779 IRB APPROVAL DATE: 10/08/2018 </div>	<p>Subject ID: _____ Date: _____</p> <p style="text-align: center; font-size: small;">Bone Density Research Laboratory Department of Health and Exercise Science University of Oklahoma</p> <p style="text-align: center; font-size: x-small;">MENSTRUAL HISTORY QUESTIONNAIRE</p> <p style="font-size: x-small;">We are asking you to give us as complete a menstrual history as possible. All information is strictly confidential.</p> <p style="font-size: x-small;">Are you pregnant (circle your response) YES- Do not complete the rest of this form NO- Continue to section A.</p> <p>SECTION A: CURRENT MENSTRUAL STATUS</p> <p>1. Approximately how many menstrual periods have you had during the past 12 months? (please circle what months you have had a period. This means from this time last year to the present month)</p> <p style="text-align: center; font-size: small;">Jan Feb Mar Apr May Jun Jul Aug Sep Oct Nov Dec</p> <p>2. What is the usual length of your menstrual cycle (first day of your period to the next onset of your period)? _____ days. Today is day _____ of your present menstrual cycle.</p> <p>3. What was the date of the onset of your last period?</p> <p>4. When do you expect you next period?</p> <p>5. What is the average length (number of days) of your menstrual flow? _____ days How many of these days do you consider "heavy"? _____ days</p> <p>6. Do you experience cramps during menstruation (dysmenorrhea)? If yes, how many days does this last?</p> <p>7. Do you experience symptoms of premenstrual syndrome (i.e., weight gain, increased eating, depression, headaches, anxiety, breast tenderness)? If yes, please list the symptoms.</p> <div style="text-align: right; font-size: x-small;"> IRB NUMBER: 9779 IRB APPROVAL DATE: 10/08/2018 </div>
-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------	-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------

Medical History Questionnaire

Date: _____	Date: _____
MS-Medical History Participation Information	
Name: _____	Date of Birth: _____
Address: _____	Phone number: (w) _____
	(h) _____
Email: _____	
Blood Pressure: _____ / _____	(cell) _____
Height: _____	Weight: _____
Gender: Male Female (circle)	
Ethnicity: Caucasian African American Hispanic Asian Other	
Emergency contact name and number: _____	
Family Physician name and number: _____	
Please answer the following questions:	
I. GENERAL HEALTH	
1. Have you been diagnosed with diabetes? If "yes", please explain _____	Y N
2. Have you ever had an oral glucose tolerance test? If "yes", please explain _____	Y N
3. Have you ever been told by a physician that you have Osteoporosis/Osteopenia?	Y N
4. Have you ever been told by a physician that you have a heart condition?	Y N
3. Have you or anyone in your immediate family had a heart attack, stroke, or cardiovascular disease before age 50 yrs? If "yes," please explain _____	Y N
5. Have you ever been told by a physician that you have high blood pressure?	Y N
6. Have you ever been told by a physician that you have high cholesterol?	Y N
7. Have you ever been told by a physician that you have thyroid problems?	Y N
If you answered yes, please define (hypothyroidism or hyperthyroidism) _____	
1	2

Date: _____	Date: _____
II. MEDICATION/SUPPLEMENTS	
1. Please list all of the prescription medications you are currently taking.	
Medicine name	Amount taken per day
a. _____	Months/years on the medication
b. _____	Reason
c. _____	_____
d. _____	_____
e. _____	_____
f. _____	_____
2. Any known allergies? Explain _____	
3. Have you been on steroid medication in the past? _____ If so, please explain in detail _____	
4. Please list all of the over-the-counter medicines or supplements (including vitamins that you take regularly)	
Item name	Amount taken per day
a. _____	Months/years on medication
b. _____	Reason
c. _____	_____
d. _____	_____
e. _____	_____
f. _____	_____
3	4

8. Have you ever been told by a physician that you have kidney disease?	Y N
9. Do you feel angina-like symptoms (pain or pressure in your chest, neck, shoulders, or arms) during or after physical activity?	Y N
10. Do you ever lose your balance because of dizziness?	Y N
11. Do you ever lose consciousness?	Y N
12. Do you consider most of your days very stressful?	Y N
13. Do you consider your eating habits healthy overall? (Lower in fats and fried foods, higher in fruits, veggies and grains)	Y N
14. Have you had any major surgeries, or any surgery that required incisions? If "yes", please explain: _____	Y N
15. Do you consider yourself to be generally healthy?	Y N
16. Do you currently smoke cigarettes or cigars or chew tobacco? If "yes", how often and how much: _____	Y N
17. Are you a former smoker? If so, how long has it been since you quit smoking? _____	Y N
18. Has your weight changed more than 5 pounds in the last 6 months?	Y N
EARS:	NOSE:
_____ hearing difficulty	_____ bleeding
_____ ringing	_____ difficulty smelling
_____ pain	_____ nasal congestion
_____ discharge	_____ sinus problems
_____ other	_____ other
Please explain _____	
PULMONARY:	_____ chronic cough
_____ shortness of breath	_____ allergies
_____ wheezing	_____ other
_____ asthma	
Please explain _____	
19. Are there any other health-related issues we should know about? Please explain _____	

III. REPRODUCTIVE STATUS (If male, skip to section IV)	
1. Have you reached menopause? (if NO skip to Section IV)	Y N
2. How long has it been since you reached menopause? _____	Y N
3. Do you still have your ovaries? a. If not, how old were you when they were removed? _____	Y N
4. Have you ever been on hormone replacement therapy? a. If so, are you still taking hormone replacement therapy? b. If you have previously taken hormone replacement therapy, but have since stopped, when did you stop taking hormone replacement therapy? _____	Y N
5. Have you ever taken osteoporosis medications? Which ones and for how long? _____	Y N
IV. OSTEOPOROSIS/FRACTURE/BONE HEALTH SECTION	
1. Have you ever had a bone scan? If so, what year? _____ What was the outcome? _____	Y N
2. Please provide a list of any bone fractures you have had in the past.	
Bone	Cause (fall, accident, etc)
_____	Year
3. Did a doctor tell you that any of these fractures were due to osteoporosis/osteopenia?	Y N
4. Is your diet low in dairy products (≤ 3 servings/day)? Y N	
5. Do you take calcium supplements? If so, how much per day? _____	Y N
6. In a typical week, how many alcoholic drinks do you consume? _____	
7. Do you drink coffee, tea, or cola products routinely? About how much coffee, tea, or cola do you drink on an average day? _____	Y N

Date: _____

8. Do you have a heart valve or implant device such as knee, hip etc.? Y N

FEAR OF FALLING (Falls Efficacy Scale)

On a scale from 1 to 10, with 1 being very confident and 10 being not confident at all, how confident are you that you do the following activities without falling?


Activity	Score 1 very confident 10 not confident at all
Take a bath or shower	
Reach into cabinets or closets	
Walk around the house	
Prepare meals not requiring carrying heavy or hot objects	
Get in and out of bed	
Answer the door or telephone	
Get in and out of a chair	
Getting dressed and undressed	
Personal grooming (e.g., washing your face)	
Getting on and off of the toilet	
Total Score	

V. SUN EXPOSURE

- How many times a week do you spend more than 10 minutes outside? _____
- How much time do you spend outdoors (minutes) per week? _____
- How much of your outdoor time is spent without sunscreen on (minutes)? _____
- How much of your outdoor time is spent "fully exposed" (minutes)? _____
("fully exposed" is defined as uncovered face, arms, and hands)

VI. EXERCISE HABITS

- How many times per week do you generally exercise? _____
 - What type(s) of exercise do you generally perform? (circle all that apply)
 Walking Running Bicycling Swimming

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Date: _____

Weight Lifting Aerobics Spinning Tennis

Other _____

b. In a typical week, how many days do you exercise? (circle)

0-1 time/week 2-3 times/week 4-6 times/week daily

c. How many minutes do you typically exercise per session (circle)

<15 min 15-30 min 30-45 >45

Other _____

d. What is the typical level of exertion during your exercise?

Light Moderate Moderate/Heavy Heavy

e. When you are exercising do you ever feel limited by the following?

	Yes	No	Activity
Breathing	___	___	_____
Chest arm neck pain	___	___	_____
Low back pain	___	___	_____
Side ache	___	___	_____
Leg pain	___	___	_____
Foot drop	___	___	_____


Other? Please explain _____

VII. MULTIPLE SCLEROSIS STATUS

- How long have you been diagnosed with Multiple Sclerosis? _____
- When did you have your first MS symptom? _____
- Has your physician ever discussed what type of MS you have? YES NO

Relapsing remitting Primary progressive Secondary progressive Progressive relapsing

- Briefly described your current MS symptoms _____

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5. Does MS affect your legs? YES NO Does MS affect your arms? YES NO

If yes, which leg is more involved? Right Left Both same
If yes, which arm is more involved? Right Left Both same

6. Do you feel numbness in your legs? YES NO

If yes, which leg is more involved? Right Left Both same

7. Do you feel numbness in your arms? Yes No

If yes, which arm is more involved? Right Left Both same

8. Do you feel tingling in your legs? YES NO

If yes, which leg is more involved? Right Left Both same

9. Do you feel tingling in your arms? YES NO

If yes, which arm is more involved Right Left Both same

10. Do you fatigue easily? YES NO
If yes, what causes it to be worse? _____

11. Do you ever experience worsening of symptoms? YES NO

	Describe	YES	NO	How often?
Bath/shower	_____	___	___	_____
Physical activity	_____	___	___	_____
Hot outside	_____	___	___	_____
Other	_____	___	___	_____
Other	_____	___	___	_____

12. Do you drive yourself independently? YES NO


13. Do you walk (circle) without aid with cane walker wheelchair

14. Has your physician ever recommended that you get a bone scan? _____

15. Has your physician ever recommended that you exercise? _____

Family Practice Physician _____ Phone _____

Neurologist _____ Phone _____

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Date: _____

Other _____ Phone _____

VIII. EMPLOYMENT STATUS

- Full-time employed _____
- Part-time employed _____
- Retired _____
- Not working _____


Please describe employment status _____

IX. EDUCATION

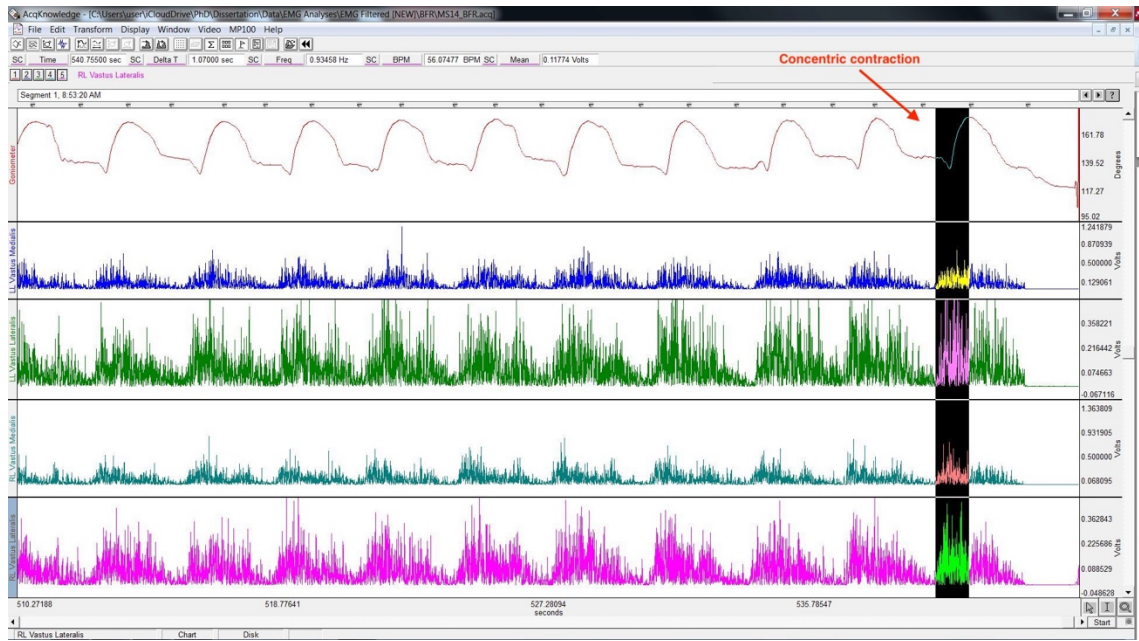
- None _____
- High School _____
- College _____
- Masters _____
- Ph.D. _____
- Other _____

I certify that these answers are accurate and complete

YOUR SIGNATURE _____ DATE _____

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Raw sEMG Signal



Appendix F: Raw Data

Descriptive Data

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
ID	SEX	EDSS	Age	Height	Weight	BMI	Fat_Mass	BFLM	Perc_B_Fat	TB_BMC	TB_BMD	SP_BMD	TH_BMD	FN_BMD	TC_BMD
MS01	1	1.50	38.6	173.5	120.7	40.10	63.11	54.72	53.57	2.88	1.282	1.410	1.107	1.073	0.868
MS03	0	2.00	63	173	119.1	39.79	56.37	59.83	48.50	2.88	1.252	1.178	0.979	0.878	0.842
MS04	1	5.50	53.3	167	101.1	36.25	50.11	48.28	50.94	2.73	1.306	1.405	-	-	-
MS05	0	1.00	48.6	179	101.6	31.71	39.01	59.30	39.70	3.34	1.503	1.365	1.141	1.184	0.944
MS06	1	2.00	53.1	169.5	81.7	28.44	37.89	41.86	47.48	1.90	0.979	1.135	0.778	0.706	0.605
MS07	0	1.00	49.7	171	85.7	29.31	29.42	53.37	35.56	2.95	1.392	1.332	1.026	0.932	0.863
MS08	1	3.50	48.3	173	115.9	38.72	52.51	60.12	46.62	3.28	1.498	1.528	1.197	1.210	0.957
MS09	0	1.00	45.7	171	92.6	31.67	41.41	48.68	45.95	2.47	1.166	1.184	0.913	0.845	0.738
MS10	1	1.00	33.4	174	68.9	22.76	19.50	46.76	29.43	2.65	1.151	1.322	0.890	0.929	0.700
MS12	1	4.00	33.5	182.5	62.7	18.83	18.92	41.26	31.44	2.52	1.179	1.288	0.897	0.913	0.677
MS13	1	2.50	37.5	157	91.2	37.00	39.56	49.24	44.56	2.43	1.244	1.119	1.109	0.931	0.877
MS14	1	0.00	35.3	168.5	61.4	21.63	15.10	43.61	25.71	2.66	1.217	1.010	0.983	1.019	0.795
MS16	1	2.00	41.1	168	107.9	38.23	53.94	51.20	51.31	2.78	1.326	1.366	1.243	1.279	0.938
MS17	1	1.00	60.8	168.5	73	25.71	32.04	38.83	45.18	2.09	1.011	1.040	0.815	0.775	0.679
MS18	1	0.00	43.2	155	92.6	38.54	48.76	41.48	54.00	2.31	1.163	1.229	0.951	0.938	0.720
Ave		1.87	45.67	170.03	91.74	31.91	39.84	49.24	43.33	2.66	1.24	1.26	1.00	0.97	0.80
SD		1.51	9.35	7.06	19.63	7.18	14.64	7.11	8.97	0.39	0.15	0.15	0.14	0.16	0.11
Min		0.00	33.40	155.00	61.40	18.83	15.10	38.83	25.71	1.90	0.98	1.01	0.78	0.71	0.61
Max		5.50	63.00	182.50	120.70	40.10	63.11	60.12	54.00	3.34	1.50	1.53	1.24	1.28	0.96

A	Q	R	S	T	U	V	W	X	Y	Z	AA	AB	AC	AD
ID	TB_Z_Score	SP_Z_Score	TH_Z_Score	FN_Z_Score	TC_Z_Score	TB_T_Score	SP_T_Score	TH_T_Score	FN_T_Score	TC_T_Score	L_Occ	R_OCC	Ave_OCC	50_OCC
MS01	0.50	0.80	0.10	-0.10	-0.50	2.00	1.90	0.80	0.30	0.20	196	171	183.5	92
MS03	-0.20	-0.70	-0.80	-1.00	-1.00	0.50	0.00	-0.20	-1.10	-0.10	214	194	204	102
MS04	1.30	1.40	-	-	-	2.20	1.90	-	-	-	150	160	155	78
MS05	2.00	0.60	0.10	0.90	0.02	3.00	1.50	1.10	1.00	0.80	168	146	157	79
MS06	-1.20	-0.30	-1.60	-1.80	-1.90	-1.00	-0.40	-1.80	-2.40	-2.10	162	156	159	80
MS07	1.60	0.90	-0.30	0.60	-0.60	1.90	1.30	0.10	-0.80	0.10	144	144	144	72
MS08	1.80	1.30	0.10	0.30	-0.10	4.10	2.90	1.50	1.20	0.90	180	180	180	90
MS09	-1.00	-0.70	-1.40	-1.60	-2.00	-0.30	0.00	-0.80	-1.40	-1.00	196	190	193	97
MS10	0.50	1.10	-0.90	-0.60	-1.30	0.70	1.20	-0.90	-0.80	-1.30	140	146	143	72
MS12	1.10	1.00	-0.70	-0.60	-1.30	1.00	0.90	-0.90	-0.90	-1.50	128	123	125.5	63
MS13	0.50	-1.40	0.30	-1.00	-0.30	1.60	1.12	0.80	-0.80	0.20	152	144	148	74
MS14	1.50	-1.30	0.00	-0.20	-0.20	1.40	-1.40	-0.20	-0.50	-0.50	148	152	150	75
MS16	0.90	0.40	1.30	1.40	0.10	2.40	1.50	1.90	1.70	0.80	198	193	195.5	98
MS17	0.00	-0.20	-0.80	-0.80	-0.70	-0.70	-1.20	-1.50	-1.90	-1.50	168	172	170	85
MS18	-0.40	-0.50	-0.80	-0.80	-1.60	0.80	0.40	-0.40	-0.70	-1.10	196	176	186	93
Ave	0.59	0.16	-0.39	-0.38	-0.81	1.31	0.77	-0.04	-0.51	-0.44	169.33	163.13	166.23	83.12
SD	1.00	0.94	0.76	0.92	0.71	1.39	1.20	1.12	1.17	0.98	26.00	21.17	23.06	11.53
Min	-1.20	-1.40	-1.60	-1.80	-2.00	-1.00	-1.40	-1.80	-2.40	-2.10	128.00	123.00	125.50	62.75
Max	2.00	1.40	1.30	1.40	0.10	4.10	2.90	1.90	1.70	0.90	214.00	194.00	204.00	102.00

A	AE	AF	AG	AH	AI	AJ	AK	AL	AM	AN
ID	1RM_LP_1	1RM_LP_2	1RM_LP_MAX	RM_LP_ABS_C	RM_LP_PC	1RM_KE_1	1RM_KE_2	1RM_KE_MAX	RM_KE_ABS_C	RM_KE_PC
MS01	146.08	172.36	172.36	26.28	17.99	75.48	81.19	81.19	5.71	7.56
MS03	117.93	127.01	127.01	9.08	7.70	58.33	58.33	58.33	0	0.00
MS04	54.43	72.57	72.57	18.14	33.33	24.04	29.76	29.76	5.72	23.79
MS05	108.86	108.86	108.86	0	0.00	58.33	58.33	58.33	0	0.00
MS06	81.65	108.86	108.86	27.21	33.33	52.62	52.62	52.62	0	0.00
MS07	164.22	154	164.22	-10.22	-6.22	64.05	58.33	64.05	-5.72	-8.93
MS08	72.57	81.65	81.65	9.08	12.51	41.19	46.9	46.9	5.71	13.86
MS09	91.65	117.93	117.93	26.28	28.67	56.3	46.9	56.3	-9.4	-16.70
MS10	108.86	117.93	117.93	9.07	8.33	52.62	52.62	52.62	0	0.00
MS12	91.86	100.72	100.72	8.86	9.65	46.9	46.9	46.9	0	0.00
MS13	117.93	127.93	127.93	10	8.48	58.33	69.76	69.76	11.43	19.60
MS14	127.01	145.15	145.15	18.14	14.28	58.33	58.33	58.33	0	0.00
MS16	90.72	100.72	100.72	10	11.02	35.47	35.47	35.47	0	0.00
MS17	73.5	99.79	99.79	26.29	35.77	38.37	38.37	38.37	0	0.00
MS18	81.65	81.65	81.65	0	0.00	41.19	41.19	41.19	0	0.00
Ave	101.93	114.48	115.16	12.55	14.32	50.77	51.67	52.67	0.90	2.61
SD	29.46	27.75	28.90	11.15	13.05	12.97	13.30	13.54	4.91	10.20
Min	54.43	72.57	72.57	-10.22	-6.22	24.04	29.76	29.76	-9.40	-16.70
Max	164.22	172.36	172.36	27.21	35.77	75.48	81.19	81.19	11.43	23.79

Muscle Thickness

A	B	C	D	E	F	G	H	I	J	K	L	M
ID	MT_BFR_Pre_RL	MT_BFR_O_RL	MT_BFR_30_RL	MT_BFR_60_RL	MT_HI_Pre_RL	MT_HI_O_RL	MT_HI_30_RL	MT_HI_60_RL	MT_BFR_Pre_LL	MT_BFR_O_LL	MT_BFR_30_LL	MT_BFR_60_LL
MS01												
MS03	4.29	4.41	4.32	4.18	4.35	4.6	4.5	4.55	4.08	4.19	4.12	4.08
MS04												
MS05	4.34	4.71	4.52	4.39	4.52	5.3	4.76	4.4	4.66	4.91	4.58	4.57
MS06	3.01	3.95	4	4	2.99	3.17	2.99	2.89	3.05	3.98	4.38	3.9
MS07	3.97	4.32	4.12	4.08	4.32	3.71	4.5	4.21	3.82	4.46	3.98	3.92
MS08												
MS09	3.68	3.98	3.73	3.64	4.32	4.51	4.42	4.38	3.72	3.81	3.71	3.52
MS10	2.85	3.11	2.97	2.94	2.73	3.2	2.72	2.7	3.14	3.56	3.26	3.21
MS12	2.6	3.1	2.82	2.73	2.75	3.33	2.91	2.76	2.84	3.2	2.99	2.7
MS13	2.81	3.01	2.92	2.9	2.75	2.99	2.81	2.77	2.87	3.09	2.97	2.92
MS14	3.22	3.52	3.47	3.21	2.81	3.18	3.02	2.7	3.15	3.47	3.31	3.19
MS16												
MS17	3.08	3.42	3.04	3.16	3.11	3.68	3.05	3.06	3.04	3.48	2.78	2.86
MS18												

A	N	O	P	Q	R	S	T	U	V	W
ID	MT_HI_Pre_LL	MT_HI_O_LL	MT_HI_30_LL	MT_HI_60_LL	ABS_C_MT_BFR_O_RL	ABS_C_MT_BFR_30_RL	ABS_C_MT_BFR_60_RL	ABS_C_MT_HI_O_RL	ABS_C_MT_HI_30_RL	ABS_C_MT_HI_60_RL
MS01										
MS03	4.08	4.1	4.2	4.25	0.12	0.03	-0.11	0.25	0.15	0.2
MS04										
MS05	4.66	5.43	5	4.74	0.37	0.18	0.05	0.78	0.24	-0.12
MS06	2.96	3.26	2.73	3.05	0.94	0.99	0.99	0.18	0	-0.1
MS07	4.1	4.7	4.42	4.29	0.35	0.15	0.11	-0.61	0.18	-0.11
MS08										
MS09	3.63	3.8	3.71	3.7	0.3	0.05	-0.04	0.19	0.1	0.06
MS10	3.31	3.71	3.25	3.19	0.26	0.12	0.09	0.47	-0.01	-0.03
MS12	2.99	3.52	3.1	3.02	0.5	0.22	0.13	0.58	0.16	0.01
MS13	2.78	3.06	2.9	2.81	0.2	0.11	0.09	0.24	0.06	0.02
MS14	2.91	3.39	3.17	2.88	0.3	0.25	-0.01	0.37	0.21	-0.11
MS16										
MS17	2.94	3.03	2.33	2.34	0.34	-0.04	0.08	0.57	-0.06	-0.05
MS18										

A	X	Y	Z	AA	AB	AC	AD	AE	AF	AG
ID	ABS_C_MT_BFR_O_LL	ABS_C_MT_BFR_30_LL	ABS_C_MT_BFR_60_LL	ABS_C_MT_HI_O_LL	ABS_C_MT_HI_30_LL	ABS_C_MT_HI_60_LL				
MS01										
MS03	0.11	0.04	0	0.02	0.12	0.17				
MS04						0				
MS05	0.25	-0.08	-0.09	0.77	0.34	0.08				
MS06	0.93	1.33	0.85	0.3	-0.23	0.09				
MS07	0.64	0.16	0.1	0.6	0.32	0.19				
MS08										
MS09	0.09	-0.01	-0.2	0.17	0.08	0.07				
MS10	0.42	0.12	0.07	0.4	-0.06	-0.12				
MS12	0.36	0.15	-0.14	0.53	0.11	0.03				
MS13	0.22	0.1	0.05	0.28	0.12	0.03				
MS14	0.32	0.16	0.04	0.48	0.26	-0.03				
MS16										
MS17	0.44	-0.26	-0.18	0.09	-0.61	-0.6				
MS18										

Thigh Circumference

A	B	C	D	E	F	G	H	I	J	K	L	M
ID	CC_BFR_Pre_RL	CC_BFR_0_RL	CC_BFR_30_RL	CC_BFR_60_RL	CC_HI_Pre_RL	CC_HI_0_RL	CC_HI_30_RL	CC_HI_60_RL	CC_BFR_Pre_LL	CC_BFR_0_LL	CC_BFR_30_LL	CC_BFR_60_LL
MS01	75	76.5	76.9	74.6	75.3	75.6	76.8	77.5	76	76.8	76.3	74.5
MS03	64.8	66.1	65.1	64.8	66	66.9	66.3	66.8	63.9	65.1	64.2	63.8
MS04	62	62	62.5	63.5	61.5	62.1	61.8	60.9	63	64.5	63.6	64.5
MS05	61.9	62.9	61.9	61.8	65.4	62.9	64.8	63.3	62.2	63.1	61.8	61.8
MS06	54.5	55	55	55	56.5	57	56.5	55.5	55	56	54	54
MS07	54	55.8	55.6	54.8	56.5	57.5	56.6	55.9	53	53.2	53.8	52.7
MS08	79.2	79.2	79.1	78.4	78.7	78.8	78.9	76.5	78.7	79.3	79.2	78.3
MS09	58.2	58.6	58.4	58.1	58.9	59.7	59.1	58.1	59.7	60.4	58.9	57.9
MS10	51.8	52.1	52.1	51.6	51.6	51.7	51.6	51.5	52.3	52.6	52.6	52.4
MS12	50.5	51.7	50.9	50.1	48.2	49.6	49.4	48.2	49.5	50.7	50.2	49.7
MS13	58.5	61.2	59.2	59	59.3	62	60.1	59.1	58.7	61.7	59.7	59.2
MS14	50	50.6	50.1	49.2	48.1	50.3	50	49.9	49.7	50.2	50.2	49.7
MS16	72	71	71	68	68	70.2	69.8	68	69	70	70.5	67.9
MS17	56.5	56	55.8	55.8	55	55.8	55.5	53	53.9	53.4	53.7	52.7
MS18	74.6	76.9	74.2	73.9	73.1	75.1	74.6	74.7	74.8	76.2	75.1	74.1

A	N	O	P	Q	R	S	T	U	V	W
ID	CC_HI_Pre_LL	CC_HI_0_LL	CC_HI_30_LL	CC_HI_60_LL	ABS_C_CC_BFR_0_RL	ABS_C_CC_BFR_30_RL	ABS_C_CC_BFR_60_RL	ABS_C_CC_HI_0_RL	ABS_C_CC_HI_30_RL	ABS_C_CC_HI_60_RL
MS01	76.2	76.8	77	77.5	1.5	1.9	-0.4	0.3	1.5	2.2
MS03	63	64.9	63.4	65.5	1.3	0.3	0	0.9	0.3	0.8
MS04	61.7	62.6	61.4	60.7	0	0.5	1.5	0.6	0.3	-0.6
MS05	62	63.2	62.7	63.5	1	0	-0.1	-2.5	-0.6	-2.1
MS06	56	58	57	55.5	0.5	0.5	0.5	0.5	0	-1
MS07	54.5	54.5	53.8	53.1	1.8	1.6	0.8	1	0.1	-0.6
MS08	80.4	82	81	76.8	0	-0.1	-0.8	0.1	0.2	-2.2
MS09	60.1	60.2	59.8	57.9	0.4	0.2	-0.1	0.8	0.2	-0.8
MS10	53	53.2	53	52.3	0.3	0.3	-0.2	0.1	0	-0.1
MS12	49	49.5	49.3	48.3	1.2	0.4	-0.4	1.4	1.2	0
MS13	58.4	60.3	59.4	58.7	2.7	0.7	0.5	2.7	0.8	-0.2
MS14	48.4	50.6	50	49.7	0.6	0.1	-0.8	2.2	1.9	1.8
MS16	67	69	68	65.8	-1	-1	-4	2.2	1.8	0
MS17	50.5	51	52.8	51	-0.5	-0.7	-0.7	0.8	0.5	-2
MS18	74.8	75.1	73.3	72.2	2.3	-0.4	-0.7	2	1.5	1.6

A	X	Y	Z	AA	AB	AC
ID	ABS_C_CC_BFR_0_LL	ABS_C_CC_BFR_30_LL	ABS_C_CC_BFR_60_LL	ABS_C_CC_HI_0_LL	ABS_C_CC_HI_30_LL	ABS_C_CC_HI_60_LL
MS01	0.8	0.3	-1.5	0.6	0.8	1.3
MS03	1.2	0.3	-0.1	1.9	0.4	2.5
MS04	1.5	0.6	1.5	0.9	-0.3	-1
MS05	0.9	-0.4	-0.4	1.2	0.7	1.5
MS06	1	-1	-1	2	1	-0.5
MS07	0.2	0.8	-0.3	0	-0.7	-1.4
MS08	0.6	0.5	-0.4	1.6	0.6	-3.6
MS09	0.7	-0.8	-1.8	0.1	-0.3	-2.2
MS10	0.3	0.3	0.1	0.2	0	-0.7
MS12	1.2	0.7	0.2	0.5	0.3	-0.7
MS13	3	1	0.5	1.9	1	0.3
MS14	0.5	0.5	0	2.2	1.6	1.3
MS16	1	1.5	-1.1	2	1	-1.2
MS17	-0.5	-0.2	-1.2	0.5	2.3	0.5
MS18	1.4	0.3	-0.7	0.3	-1.5	-2.6

Interleukin-6

A	B	C	D	E	F	G	H	I	J	K
Uncorrected Interleukin-6										
ID	unc_IL6_BFR_Pre	unc_IL6_BFR_5min	unc_IL6_BFR_1h	unc_IL6_HI_Pre	unc_IL6_HI_5min	unc_IL6_HI_1h	abs_unc_IL6_BFR_5min	abs_unc_IL6_BFR_1h	abs_unc_IL6_HI_5min	abs_unc_IL6_HI_1h
MS01	2.548	2.808	3.112	3.366	3.834	4.506	0.260	0.564	0.468	1.140
MS03	2.532	1.587	2.299	1.764	1.808	1.295	-0.945	-0.233	0.044	-0.469
MS04	2.065	4.442	7.115	3.067	4.449	4.509	2.377	5.050	1.382	1.442
MS05	1.223	1.147	1.197	0.928	0.859	0.565	-0.076	-0.076	-0.069	-0.363
MS06	2.972	1.992	2.562	1.771	2.596	2.781	-0.980	-0.410	0.825	1.010
MS07	1.292	1.141	1.303	2.445	1.685	2.324	-0.91	-0.151	0.011	-0.760
MS08	4.225	4.266	4.742	3.232	3.587	7.314	0.041	0.517	0.355	4.082
MS09	3.625	3.065	3.165	4.133	2.238	2.462	-0.560	-0.460	-1.895	-1.671
MS10	0.847	1.402	1.154	1.328	0.993	0.797	0.555	0.307	-0.335	-0.531
MS12	10.251	10.618	10.659	7.177	7.199	6.071	0.367	0.408	0.022	-1.106
MS13	1.705	1.105	0.873	0.883	0.444	0.612	-0.600	-0.832	-0.439	-0.271
MS14	0.684	0.742	0.536	0.536	0.704	0.771	0.058	0.168	0.168	0.235
MS16	2.424	2.261	2.231	5.316	4.337	3.711	0.47	-0.163	-0.979	-1.605
MS17	0.8075	0.517	0.6545	0.959	0.603	0.367	-0.291	-0.153	-0.356	-0.592
MS18	1.151	0.894	1.266	1.412	1.455	1.834	-0.257	0.115	0.043	0.422

ID	cor_IL6_BFR_Pre	cor_IL6_BFR_5min	cor_IL6_BFR_1h	cor_IL6_HI_Pre	cor_IL6_HI_5min	cor_IL6_HI_1h	abs_c_cor_IL6_BFR_5min	abs_c_cor_IL6_BFR_1h	abs_c_cor_IL6_HI_5min	abs_c_cor_IL6_HI_1h
MS01	2.548	2.504	2.923	3.366	3.757	4.460	-0.04	0.37	0.39	1.09
MS03	2.532	1.620	2.692	1.764	1.771	1.282	-0.91	0.16	0.01	-0.48
MS04	2.065	4.311	7.558	3.067	4.920	5.193	2.25	5.49	1.85	2.13
MS05	1.223	1.159	1.162	0.928	0.762	0.522	-0.06	-0.06	-0.17	-0.41
MS06	2.972	2.012	2.413	1.771	2.649	2.726	-0.96	-0.56	0.88	0.96
MS07	1.292	0.990	1.226	2.445	1.462	2.373	-0.30	-0.07	-0.98	-0.07
MS08	4.225	4.355	4.940	3.232	3.587	7.390	0.13	0.71	0.36	4.16
MS09	3.625	3.065	3.267	4.133	2.192	2.462	-0.56	-0.36	-1.94	-1.67
MS10	0.847	1.317	1.073	1.328	0.983	0.805	0.47	0.23	-0.35	-0.52
MS12	10.251	10.735	11.477	7.177	7.125	5.886	0.48	1.23	-0.05	-1.29
MS13	1.705	1.061	0.891	0.883	0.410	0.618	-0.64	-0.81	-0.47	-0.26
MS14	0.684	0.631	0.536	0.536	0.734	0.787	-0.05	-0.68	0.20	0.25
MS16	2.424	2.404	2.372	5.316	4.337	3.711	-0.02	-0.05	-0.98	-1.61
MS17	0.8075	0.507	0.682	0.959	0.603	0.345	-0.30	-0.13	-0.36	-0.61
MS18	1.151	0.912	1.253	1.412	1.368	1.467	-0.24	0.10	-0.04	0.06

Mammalian Target of Rapamycin

A	B	C	D	E	F	G	H	I	J	K
Uncorrected mTOR										
ID	unc_mTOR_BFR_Pre	unc_mTOR_BFR_5min	unc_mTOR_BFR_1h	unc_mTOR_HI_Pre	unc_mTOR_HI_5min	unc_mTOR_HI_1h	abs_unc_mTOR_BFR_5min	abs_unc_mTOR_BFR_1h	abs_unc_mTOR_HI_5min	abs_unc_mTOR_HI_1h
MS01	3.945	3.975	3.787	3.5135	3.978	3.8495	0.030	-0.158	0.465	0.336
MS03	16.748	15.406	15.469	19.916	18.402	15.085	-1.342	-1.279	-1.514	-4.831
MS04	16.353	18.426	18.188	17.06	17.976	17.387	2.073	1.835	0.916	0.327
MS05	5.539	4.738	5.074	5.32	5.179	5.186	-0.801	-0.465	-0.141	-0.134
MS06	2.19	2.289	2.365	1.97	1.754	1.997	0.099	0.175	-0.216	0.027
MS07	6.337	5.271	7.159	5.88	4.689	5.236	-1.066	0.822	-1.191	-0.644
MS08	18.046	14.247	16.892	15.24	16.386	16.18	-3.799	-1.154	1.146	0.940
MS09	3.884	4.299	4.084	4.549	4.913	5.046	0.415	0.200	0.364	0.497
MS10	8.207	5.525	5.667	5.053	7.029	5.702	-2.682	-2.540	1.976	0.649
MS12	4.873	5.658	5.656	5.4225	5.1455	5.3225	0.785	0.783	-0.277	-0.100
MS13	3.504	3.099	2.856	1.633	5.063	4.396	-0.405	-0.648	3.490	2.765
MS14	12.923	12.878	12.878	14.623	12.833	12.241	-0.045	-0.045	-1.790	-2.382
MS16	5.067	3.955	4.181	4.001	5.32	4.976	-1.072	-0.886	1.319	0.975
MS17	16.354	15.807	14.92	17.815	16.072	15.337	-0.547	-1.434	-1.743	-2.478
MS18	6.603	6.804	7.802	4.508	4.878	4.71	0.201	1.199	0.370	0.202

ID	cor_mTOR_BFR_Pre	cor_mTOR_BFR_5min	cor_mTOR_BFR_1h	cor_mTOR_HI_Pre	cor_mTOR_HI_5min	cor_mTOR_HI_1h	abs_c_cor_mTOR_BFR_5min	abs_c_cor_mTOR_BFR_1h	abs_c_cor_mTOR_HI_5min	abs_c_cor_mTOR_HI_1h
MS01	3.945	3.544	3.557	3.5135	3.898	3.811	-0.40	-0.39	0.33	0.30
MS03	16.748	15.729	18.116	19.916	18.025	14.928	-1.02	1.37	-1.89	-4.99
MS04	16.353	17.882	19.320	17.06	19.878	20.026	1.53	2.97	2.82	2.97
MS05	5.539	4.786	4.924	5.32	4.593	4.787	-0.61	-0.73	-0.61	-0.53
MS06	2.19	2.312	2.227	1.97	1.790	1.958	0.12	0.04	-0.18	-0.01
MS07	6.337	4.574	6.736	5.88	4.067	5.346	-1.76	0.40	-1.81	-0.53
MS08	18.046	14.543	17.596	15.24	16.368	16.348	-3.50	-0.45	1.15	1.11
MS09	3.884	4.299	4.215	4.549	4.813	5.046	0.42	0.33	0.26	0.50
MS10	8.207	5.192	5.270	5.053	6.957	5.762	-3.02	-2.94	1.90	0.71
MS12	4.873	5.720	6.090	5.4225	5.093	5.161	0.85	1.22	-0.33	-2.13
MS13	3.504	2.975	2.916	1.633	4.671	4.443	-0.53	-0.59	3.04	2.81
MS14	12.923	10.945	14.623	13.376	12.494	12.494	-1.98	-1.25	-2.13	-2.13
MS16	5.067	4.248	4.448	4.001	5.320	4.976	-0.82	-0.62	1.32	0.98
MS17	16.354	15.492	15.543	17.815	16.072	14.419	-0.86	-0.81	-1.74	-3.39
MS18	6.603	6.943	7.724	4.508	4.588	3.768	0.34	1.12	0.08	-0.74

Cortisol

A	B	C	D	E	F	G	H	I	J	K
Uncorrected Cortisol										
ID	unc_cort_BFR_Pre	unc_cort_BFR_Smin	unc_cort_BFR_1h	unc_cort_HI_Pre	unc_cort_HI_Smin	unc_cort_HI_1h	abs_unc_cort_BFR_Smin	abs_unc_cort_BFR_1h	abs_unc_cort_HI_Smin	abs_unc_cort_HI_1h
MS01	173.488	169.974	90.137	157.552	117.140	110.474	-3.514	-83.331	-40.412	-47.078
MS03	103.653	113.192	96.354	45.532	127.620	187.273	9.539	-7.299	82.088	141.741
MS04	68.688	39.990	34.500	102.528	124.917	77.421	-28.698	-34.188	22.389	-25.107
MS05	156.221	132.026	109.966	126.089	85.952	78.880	-24.195	-46.255	-40.137	-47.209
MS06	122.887	276.806	140.057	153.581	102.252	192.021	153.919	17.170	-51.329	38.440
MS07	177.208	132.936	133.855	200.840	147.422	145.926	-44.272	-43.353	-53.418	-54.914
MS08	127.448	199.054	134.226	108.606	163.332	105.957	71.606	6.778	54.726	-2.649
MS09	132.389	72.774	86.244	143.625	95.227	79.754	-59.615	-46.145	-48.398	-63.871
MS10	312.446	216.374	132.108	306.544	247.470	177.050	-96.072	-180.338	-59.074	-129.894
MS12	173.488	132.073	95.466	175.596	110.910	87.392	-41.415	-78.022	-64.686	-88.204
MS13	114.955	190.611	117.458	173.488	167.698	132.263	75.656	2.503	-5.790	-41.225
MS14	178.683	135.773	135.773	245.448	183.492	124.434	-42.910	-178.683	-61.956	-121.014
MS16	125.581	133.277	86.532	117.638	53.621	73.086	7.696	-39.049	-64.017	-44.552
MS17	192.558	202.162	175.440	1369.650	2184.382	3841.921	9.604	-17.119	814.732	2472.271
MS18	405.196	290.778	238.053	403.317	316.968	310.937	-114.418	-167.143	-86.349	-92.380
Corrected Cortisol										
ID	cor_cort_BFR_Pre	cor_cort_BFR_Smin	cor_cort_BFR_1h	cor_cort_HI_Pre	cor_cort_HI_Smin	cor_cort_HI_1h	abs_c_cor_cort_BFR_Smin	abs_c_cor_cort_BFR_1h	abs_c_cor_cort_HI_Smin	abs_c_cor_cort_HI_1h
MS01	173.488	151.55	84.67	157.552	114.79	109.36	-21.94	-88.81	-42.76	-48.20
MS03	103.653	115.57	112.84	45.532	125.00	185.33	11.91	9.19	79.47	139.80
MS04	68.688	38.81	36.65	102.528	138.14	89.17	-29.88	-32.04	35.61	-13.36
MS05	156.221	133.316	106.72	126.089	76.23	72.81	-22.86	-49.50	-49.86	-53.27
MS06	122.887	279.61	131.91	153.581	104.35	188.24	156.73	9.02	-49.23	34.66
MS07	177.208	115.36	125.96	200.84	127.87	148.99	-61.84	-51.25	-72.97	-51.85
MS08	127.448	203.19	139.82	108.606	163.33	107.06	75.74	12.37	54.73	-1.55
MS09	132.389	72.77	89.02	143.625	93.28	79.75	-59.62	-43.37	-50.34	-63.87
MS10	312.4455	203.32	122.85	306.5435	244.93	178.91	-109.12	-189.60	-61.61	-127.63
MS12	173.488	133.52	102.80	175.596	109.77	84.73	-39.56	-70.69	-65.82	-90.86
MS13	114.955	183.01	119.91	173.488	154.72	133.61	68.06	4.96	-18.76	-39.87
MS14	178.683	115.39	135.773	245.448	191.25	127.00	-43.29	-178.68	-54.20	-118.44
MS16	125.5805	141.73	92.02	117.638	53.62	73.09	16.15	-33.56	-64.02	-44.55
MS17	192.558	198.13	182.77	1369.650	2184.38	3614.35	5.57	-9.79	814.73	2244.70
MS18	405.196	296.72	235.67	403.317	298.12	248.78	-108.47	-169.53	-105.20	-154.53

Myostatin

A	B	C	D	E	F	G	H	I	J	K
Uncorrected Myostatin										
ID	unc_MSTN_BFR_Pre	unc_MSTN_BFR_Smin	unc_MSTN_BFR_1h	unc_MSTN_HI_Pre	unc_MSTN_HI_Smin	unc_MSTN_HI_1h	abs_unc_MSTN_BFR_Smin	abs_unc_MSTN_BFR_1h	abs_unc_MSTN_HI_Smin	abs_unc_MSTN_HI_1h
MS01	1.546	1.23	1.428	1.508	1.731	1.436	0.316	0.118	-0.223	0.072
MS03	1.039	0.818	0.873	1.094	1.107	1.021	0.221	0.166	-0.013	0.073
MS04	0.994	1.104	1.135	1.072	1.016	1.314	-0.11	-0.141	0.056	-0.242
MS05	1.175	1.195	1.084	1.445	1.281	1.124	0.02	0.091	0.164	0.321
MS06	2.983	2.313	2.104	2.093	2.861	2.194	0.65	0.859	-0.768	-0.101
MS07	1.09	1.351	1.29	1.112	1.164	1.275	-0.261	-0.2	-0.052	-0.163
MS08	1.366	1.348	1.273	1.428	1.378	1.303	0.018	0.093	0.05	0.125
MS09	1.912	2.132	2.148	1.86	1.975	2.303	-0.22	-0.236	-0.115	-0.443
MS10	1.379	1.208	1.193	1.072	1.282	1.05	0.171	0.186	-0.21	0.022
MS12	3.483	2.844	2.94	3.093	3.929	3.036	0.639	0.543	-0.836	0.057
MS13	2.392	2.697	3.404	2.686	2.483	2.793	-0.305	-1.012	0.203	-0.107
MS14	2.991	3.11	3.567	2.839	2.601	2.601	-0.119	-0.991	0.728	0.966
MS16	1.751	1.9	1.86	0.819	1.13	1.017	-0.149	-0.109	-0.311	-0.198
MS17	1.451	1.36	1.751	1.531	1.542	1.573	0.091	-0.3	-0.011	-0.042
MS18	6.964	1.708	1.786	1.983	1.892	1.782	5.256	5.178	0.091	0.201
Corrected Myostatin										
ID	cor_MSTN_BFR_Pre	cor_MSTN_BFR_Smin	cor_MSTN_BFR_1h	cor_MSTN_HI_Pre	cor_MSTN_HI_Smin	cor_MSTN_HI_1h	abs_c_cor_MSTN_BFR_Smin	abs_c_cor_MSTN_BFR_1h	abs_c_cor_MSTN_HI_Smin	abs_c_cor_MSTN_HI_1h
MS01	1.546	1.097	1.341	1.508	1.696	1.421	0.45	0.20	-0.19	0.09
MS03	1.039	0.835	1.022	1.094	1.084	1.010	0.20	0.02	0.01	0.08
MS04	0.994	1.071	1.206	1.072	1.124	1.513	-0.08	-0.21	-0.05	-0.44
MS05	1.175	1.167	1.052	1.445	1.136	1.038	0.01	0.12	0.31	0.41
MS06	2.983	2.336	1.982	2.093	2.920	2.151	0.63	0.98	-0.83	-0.06
MS07	1.09	1.172	1.214	1.112	1.010	1.302	-0.08	-0.12	0.10	-0.19
MS08	1.366	1.376	1.326	1.428	1.378	1.317	-0.01	0.04	0.05	0.11
MS09	1.912	2.132	2.217	1.86	1.935	2.303	-0.22	-0.31	-0.07	-0.44
MS10	1.379	1.135	1.109	1.072	1.269	1.061	0.24	0.27	-0.20	0.01
MS12	3.483	2.875	3.166	3.093	3.889	2.944	0.61	0.32	-0.80	0.15
MS13	2.392	2.589	3.475	2.686	2.291	2.822	-0.20	-1.08	0.40	-0.14
MS14	2.991	2.643	3.567	2.711	2.655	2.655	0.35	0.86	0.86	0.91
MS16	1.751	2.020	1.978	0.819	1.130	1.017	-0.27	-0.23	-0.31	-0.20
MS17	1.451	1.51	0.783	1.531	1.137	0.778	0.30	0.67	0.39	0.75
MS18	6.964	1.743	1.768	1.983	1.779	1.426	5.22	5.20	0.20	0.56

Whole-Blood Lactate

A	B	C	D	E	F	G	H	I	J
Uncorrected Lactate									
ID	La_BFR_Pre_1	La_BFR_Pre_2	La_BFR_Pre_Ave	La_BFR_5min_1	La_BFR_5min_2	La_BFR_5min_Ave	La_BFR_1h_1	La_BFR_1h_2	La_BFR_1h_Ave
MS01	0.8	0.8	0.8	2.3	2.2	2.25	0.5	0.5	0.5
MS03	1.6	1.5	1.55	2.6	2.6	2.6	0.9	1.2	1.05
MS04	2	2.3	2.15	2	2.1	2.05	0.9	0.9	0.9
MS05	0.5	0.4	0.45	2.5	2.9	2.7	0.9	1	0.95
MS06	0.7	0.8	0.75	2.5	2.4	2.45	0.7	0.7	0.7
MS07	0.9	0.9	0.9	3.5	3.4	3.45	1.1	1.2	1.15
MS08	0.7	0.7	0.7	1.1	1.1	1.1	0.3	0.5	0.4
MS09	1.5	1.2	1.35	3.4	3.3	3.35	2	2.1	2.05
MS10	0.7	0.8	0.75	2.2	2.2	2.2	0.7	0.7	0.7
MS12	0.4	0.4	0.4	2.8	2.8	2.8	0.7	0.5	0.6
MS13	0.8	0.8	0.8	2.2	2.3	2.25	0.9	0.9	0.9
MS14	1.6	1.8	1.7	2.5	2.5	2.5	1.6	2.1	1.85
MS16	0.7	0.7	0.7	1.8	1.8	1.8	0.8	0.8	0.8
MS17	0.4	0.4	0.4	0.8	0.8	0.8	0.5	0.8	0.65
MS18	0.7	0.7	0.7	1.8	1.9	1.85	1.1	1	1.05

A	K	L	M	N	O	P	Q	R	S
Corrected Lactate									
ID	La_HI_Pre_1	La_HI_Pre_2	La_HI_Pre_Ave	La_HI_5min_1	La_HI_5min_2	La_HI_5min_Ave	La_HI_1h_1	La_HI_1h_2	La_HI_1h_Ave
MS01	0.7	0.7	0.7	5.1	4.9	5	0.8	0.9	0.85
MS03	2.6	2.4	2.5	2.1	2.1	2.1	1.1	1	1.05
MS04	2.3	2.4	2.35	3	3	3	1.5	1.5	1.5
MS05	1.2	1.3	1.25	3.9	4.1	4	1.3	1.2	1.25
MS06	1.1	1.2	1.15	3.5	3.4	3.45	0.9	0.8	0.85
MS07	0.9	0.9	0.9	8.4	8.1	8.25	1.4	1.4	1.4
MS08	0.7	0.7	0.7	1.7	1.7	1.7	0.3	0.4	0.35
MS09	2.4	2.5	2.45	4.1	4	4.05	1.9	2.2	2.05
MS10	0.5	0.5	0.5	3.2	3.6	3.4	0.6	0.7	0.65
MS12	0.6	0.7	0.65	5.8	5.7	5.75	1.2	1.1	1.15
MS13	0.7	0.7	0.7	4.9	4.7	4.8	1.2	1.1	1.15
MS14	0.6	0.6	0.6	3.1	3.1	3.1	0.7	0.8	0.75
MS16	0.9	0.9	0.9	2.8	2.7	2.75	0.8	0.9	0.85
MS17	0.9	0.9	0.9	2.3	2.5	2.4	1.1	1	1.05
MS18	1.6	1.6	1.6	3.9	4.2	4.05	1.5	1.6	1.55

A	B	C	D	E	F	G	H	I	J	K
Corrected Lactate										
ID	La_BFR_Pre	La_BFR_5min	La_BFR_1h	La_HI_Pre	La_HI_5min	La_HI_1h	abs_c_cor_la_BFR_5min	abs_c_cor_la_BFR_1h	abs_c_cor_la_HI_5min	abs_c_cor_la_HI_1h
MS01	0.80	2.01	0.47	0.70	4.90	0.84	-1.21	0.33	-4.20	-0.14
MS03	1.55	2.65	1.23	2.50	2.06	1.04	-1.10	0.32	0.44	1.46
MS04	2.15	1.99	0.96	2.35	3.32	1.73	0.16	1.19	-0.97	0.62
MS05	0.45	2.73	0.92	1.25	3.55	1.15	-2.28	-0.47	-2.30	0.10
MS06	0.75	2.47	0.66	1.15	3.52	0.83	-1.72	0.09	-2.37	0.32
MS07	0.90	2.99	1.08	0.90	7.16	1.43	-2.09	-0.18	-6.26	-0.53
MS08	0.70	1.12	0.42	0.70	1.70	0.35	-0.42	0.28	-1.00	0.35
MS09	1.35	3.35	2.12	2.45	3.97	2.05	-2.00	-0.77	-1.52	0.40
MS10	0.75	2.07	0.65	0.50	3.37	0.66	-1.32	0.10	-2.87	-0.16
MS12	0.40	2.83	0.65	0.65	5.69	1.12	-2.43	-0.25	-5.04	-0.47
MS13	0.80	2.16	0.92	0.70	4.43	1.16	-1.36	-0.12	-3.73	-0.46
MS14	1.70	2.12	1.19	0.60	3.23	0.77	-0.42	0.51	-2.63	-0.17
MS16	0.70	1.91	0.85	0.90	2.75	0.85	-1.21	-0.15	-1.85	0.05
MS17	0.40	0.78	0.68	0.90	2.40	0.99	-0.38	-0.28	-1.50	-0.09
MS18	0.70	1.89	1.04	1.60	3.81	1.24	-1.19	-0.34	-2.21	0.36

Hematocrit

A	B	C	D	E	F	G	H	I	J
ID	Ht_BFR_Pre_1	Ht_BFR_Pre_2	Ht_BFR_Pre_AVG	Ht_BFR_5min_1	Ht_BFR_5min_2	Ht_BFR_5min_AVG	Ht_BFR_1h_1	Ht_BFR_1h_2	Ht_BFR_1h_AVG
MS01	38	39	38.5	41	41.5	41.25	40	40	40
MS03	40.5	41	40.75	40	40.5	40.25	37	37	37
MS04	48	48	48	49	48.5	48.75	46	47	46.5
MS05	48	47.5	47.75	47.5	47.5	47.5	48.5	48.5	48.5
MS06	47	47	47	47	46.5	46.75	48	49	48.5
MS07	43	42.5	42.75	46.5	46	46.25	44	44.5	44.25
MS08	43	44	43.5	43	43	43	42	43	42.5
MS09	41	41	41	41	41	41	39.5	41	40.25
MS10	39	40	39.5	41	41	41	41	41.5	41.25
MS12	40	39	39.5	39	39.5	39.25	38.5	37	37.75
MS13	43	43	43	44	44	44	43	42	42.5
MS14	41.5	41.5	41.5	46	45	45.5	51	54	52.5
MS16	43	44	43.5	42	42	42	42	42	42
MS17	43	43	43	43	44	43.5	42	42	42
MS18	44.5	45	44.75	44	44.5	44.25	45	45	45

A	K	L	M	N	O	P	Q	R	S
ID	Ht_HI_Pre_1	Ht_HI_Pre_2	Ht_HI_Pre_AVG	Ht_HI_5min_1	Ht_HI_5min_2	Ht_HI_5min_AVG	Ht_HI_1h_1	Ht_HI_1h_2	Ht_HI_1h_AVG
MS01	42	42.5	42.25	43	42.5	42.75	42	43	42.5
MS03	39	39	39	39	40	39.5	39	39.5	39.25
MS04	47.5	47.5	47.5	45	45	45	44.5	43.5	44
MS05	49	49	49	52	52	52	51	51	51
MS06	47	46.5	46.75	46	46.5	46.25	47	47.5	47.25
MS07	42	41.5	41.75	45	45.5	45.25	41	41.5	41.25
MS08	42	42	42	42	42	42	41.5	42	41.75
MS09	41	41	41	41.5	41.5	41.5	41	41	41
MS10	41	40.5	40.75	41	41	41	41	40	40.5
MS12	40.5	41	40.75	41	41	41	41	42	41.5
MS13	45	45	45	47	47	47	45	44.5	44.75
MS14	45	44	44.5	43	44	43.5	44	44	44
MS16	43	43	43	43	43	43	43	43	43
MS17	42	42	42	42	42	42	44	43	43.5
MS18	41	42	41.5	43	43	43	47	47	47

Plasma Volume Change

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
ID	PV_BFR_Pre_1	PV_BFR_Pre_2	PV_BFR_Pre_AVG	PV_BFR_5min_1	PV_BFR_5min_2	PV_BFR_5min_AVG	PV_BFR_1h_1	PV_BFR_1h_2	PV_BFR_1h_AVG	PV_HI_5min_1	PV_HI_5min_2	PV_HI_5min_AVG	PV_HI_1h_1	PV_HI_1h_2	PV_HI_1h_AVG
MS01	-	-	-	-11.80	-9.88	-10.84	-8.06	-4.10	-6.08	-4.01	0.00	-2.00	0.00	-2.02	-1.01
MS03	-	-	-	2.10	2.09	2.10	15.90	18.32	17.11	0.00	-4.10	-2.05	0.00	-2.08	-1.04
MS04	-	-	-	-3.92	-1.98	-2.95	8.36	4.09	6.23	10.58	10.58	10.58	12.84	17.52	15.18
MS05	-	-	-	2.02	0.00	1.01	-1.98	-3.93	-2.95	-11.31	-11.31	-11.31	-7.69	-7.69	-7.69
MS06	-	-	-	0.00	2.03	1.01	-3.93	-7.70	-5.82	4.10	0.00	2.05	0.00	-3.94	-1.97
MS07	-	-	-	-13.21	-13.23	-13.22	-3.99	-7.82	-5.90	-11.49	-15.03	-13.26	4.21	0.00	2.10
MS08	-	-	-	0.00	4.15	2.08	4.18	4.15	4.16	0.00	0.00	0.00	2.08	0.00	1.04
MS09	-	-	-	0.00	0.00	0.00	6.44	0.00	3.22	-2.04	-2.04	-2.04	0.00	0.00	0.00
MS10	-	-	-	-8.00	-4.07	-6.03	-8.00	-6.02	-7.01	0.00	-2.05	-1.02	0.00	2.10	1.05
MS12	-	-	-	4.27	-2.08	1.10	6.49	8.86	7.68	-2.05	0.00	-1.02	-2.05	-4.04	-3.04
MS13	-	-	-	-3.99	-3.99	-3.99	0.00	4.18	2.09	-7.74	-7.74	-7.74	0.00	2.04	1.02
MS14	-	-	-	-16.72	-13.30	-15.01	-31.84	-39.57	-35.71	8.46	0.00	4.23	4.13	0.00	2.07
MS16	-	-	-	4.18	8.50	6.34	4.18	8.50	6.34	0.00	0.00	0.00	0.00	0.00	0.00
MS17	-	-	-	0.00	-3.99	-1.99	4.18	4.18	4.18	0.00	0.00	0.00	-7.84	-4.01	-5.92
MS18	-	-	-	2.05	2.04	2.05	-2.00	0.00	-1.00	-7.88	-4.01	-5.95	-21.64	-18.34	-19.99

Ratings of Pain

A	B	C	D	E	F	G	H	I
ID	DISC_BFR_PRE_LP	DISC_BFR_POST_S1_LP	DISC_BFR_PRE_S2_LP	DISC_BFR_POST_S2_LP	DISC_BFR_PRE_S3_LP	DISC_BFR_POST_S3_LP	DISC_BFR_PRE_S4_LP	DISC_BFR_POST_S4_LP
MS01	0	2	2	3	3	3	4	4
MS03	0	0	0	1	1	1	1	1
MS04	0	5	4	3	7	5	5	8
MS05	0	4	4	5	4	5	6	6
MS06	0	6	7	8	7	8	9	9
MS07	0	1	0.5	1	1	1	1	2
MS08	0	5	6	7	8	9	10	10
MS09	0	2	3	4	4	4	5	5
MS10	0	3	3	3	3	4	4	5
MS12	0	1	2	2	3	2	2	2
MS13	0	1	0	1	0.5	2	0.5	2
MS14	0	3	3	3	3	4	1	3
MS16	0	2	2	1	2	2	2	3
MS17	0	2	3	4	1	3	3	3
MS18	0	2	2	2	3	3	3	3

A	J	K	L	M	N	O	P	Q
ID	DISC_HI_PRE_LP	DISC_HI_POST_S1_LP	DISC_HI_PRE_S2_LP	DISC_HI_POST_S2_LP	DISC_HI_PRE_S3_LP	DISC_HI_POST_S3_LP	DISC_HI_PRE_S4_LP	DISC_HI_POST_S4_LP
MS01	0	0.5	0	0.5	0	1	0.5	0.5
MS03	0	4	1	3	3	5	2	5
MS04	0	5	9	10	7	10	7	10
MS05	0	2	0	2	0.5	3	1	3
MS06	0	0	0	2	0	2	0	2
MS07	0	3	1	3	1	4	2	5
MS08	0	0.5	0	0.5	0.5	3	2	3
MS09	0	2	0	3	0	4	0	5
MS10	0	0.5	0	0.5	0	0.5	0.5	0.5
MS12	0	0	0	0	0	0.5	0.5	0.5
MS13	0	1	0	1	0	1	0	0.5
MS14	0	0	0	0	0	0	0	0
MS16	0	1	0	0.5	0	0.5	0	2
MS17	0	2	0	0.5	0.5	1	0.5	0.5
MS18	0	1	2	3	2	3	4	3

A	R	S	T	U	V	W	X	Y
ID	DISC_BFR_PRE_KE	DISC_BFR_POST_S1_KE	DISC_BFR_PRE_S2_KE	DISC_BFR_POST_S2_KE	DISC_BFR_PRE_S3_KE	DISC_BFR_POST_S3_KE	DISC_BFR_PRE_S4_KE	DISC_BFR_POST_S4_KE
MS01	2	7	6	5	5	5	5	5
MS03	1	4	3	3	2	3	4	4
MS04	5	10	10	5	5	10	8	8
MS05	0	2	1	3	2	4	2	5
MS06	0	9	6	9	6	10	7	9
MS07	0	7	2	6	2	4	2	6
MS08	3	8	8	10	9	10	9	10
MS09	0	4	3	4	5	5	5	5
MS10	2	4	3	4	4	4	3	4
MS12	0	3	2	2	1	2	1	1
MS13	0	3	1	2	0.5	2	0.5	1
MS14	0	3	2	2	0.5	2	0	0
MS16	0.5	4	3	3	2	2	1	3
MS17	0	3	0	3	0.5	4	2	3
MS18	0	2	3	2	2	0.5	0.5	1

A	Z	AA	AB	AC	AD	AE	AF	AG
ID	DISC_HI_PRE_KE	DISC_HI_POST_S1_KE	DISC_HI_PRE_S2_KE	DISC_HI_POST_S2_KE	DISC_HI_PRE_S3_KE	DISC_HI_POST_S3_KE	DISC_HI_PRE_S4_KE	DISC_HI_POST_S4_KE
MS01	0	0.5	0.5	1	1	2	1	2
MS03	2	0.5	3	5	2	6	3	6
MS04	5	10	7	8	8	9	9	10
MS05	0	2	0.5	3	0.5	3	2	4
MS06	0	3	0	2	1	3	1	2
MS07	1	7	3	6	1	8	3	8
MS08	0.5	5	3	5	4	5	5	6
MS09	0	4	2	5	0.5	2	0.5	3
MS10	0	0.5	0.5	1	0.5	1	0.5	1
MS12	0	0.5	0	0.5	0	0.5	0	0.5
MS13	0	3	0	3	0	4	0	2
MS14	0	0	0	0	0	0	0	0
MS16	0	2	0	4	0.5	5	0.5	7
MS17	0	0	0	1	0.5	1	0.5	1
MS18	2	3	2	3	2	3	3	3

Ratings of Soreness

A	B	C	D	E	F	G	H	I	J	K	L	M
ID	SORE_BFR_PRE	SORE_BFR_0	SORE_BFR_5	SORE_BFR_30	SORE_BFR_60	SORE_BFR_24	SORE_HI_PRE	SORE_HI_0	SORE_HI_5	SORE_HI_30	SORE_HI_60	SORE_HI_24
MS01	0	5	1	0.5	0	2	0	2	0	0	0	0
MS03	0	4	1	0	0	3	0	5	5	0	0	1
MS04	0	8	3	0	0	0	0	10	9	3	0	1
MS05	0	5	1	0.5	0.5	1	0	4	1	1	2	3
MS06	0	9	4	0	0	2	0	2	0	0	0	0
MS07	0	6	1	0	0	0.5	0	8	1	0	0	3
MS08	0	10	3	0.5	0	0	0	6	2	0.5	0	0
MS09	0	5	0	0	0	2	0	3	2	0	0.5	3
MS10	0	4	2	1	0.5	0.5	0	1	0.5	0	0	3
MS12	0	1	0	0	0	0	0	0.5	0	0	0	1
MS13	0	1	0	0	0	0	0	2	0	0	0	0
MS14	0	2	0	0	0	0	0	0	0	0	0	1
MS16	0	3	0.5	0	0	0	0	7	1	0	0	1
MS17	0	3	0	0	0	0	0	1	0	0	0	0
MS18	0	1	0.5	0	0	3	0	3	3	0.5	1	6

Ratings of Perceived Exertion

A	R	S	T	U	V	W	X	Y
ID	RPE_BFR_S1_LP	RPE_BFR_S2_LP	RPE_BFR_S3_LP	RPE_BFR_S4_LP	RPE_HI_S1_LP	RPE_HI_S2_LP	RPE_HI_S3_LP	RPE_HI_S4_LP
MS01	4	3	3	4	4	2	3	2
MS03	7	6	5	5	8	8	9	10
MS04	8	2	5	5	9	10	10	10
MS05	5	5	6	6	6	7	8	9
MS06	3	4	6	7	5	3	4	3
MS07	4	3	5	5	6	7	8	8
MS08	5	8	9	10	6	7	10	10
MS09	3	2	3	3	7	8	9	9
MS10	3	3	3	4	9	9	9	9
MS12	8	9	10	10	6	5	7	8
MS13	2	1	2	2	7	8	8	8
MS14	5	5	5	4	8	8	8	8
MS16	2	0	2	2	6	5	5	5
MS17	3	4	6	3	8	5	7	5
MS18	1	2	2	2	6	6	6	7

A	Z	AA	AB	AC	AD	AE	AF	AG
ID	RPE_BFR_S1_KE	RPE_BFR_S2_KE	RPE_BFR_S3_KE	RPE_BFR_S4_KE	RPE_HI_S1_KE	RPE_HI_S2_KE	RPE_HI_S3_KE	RPE_HI_S4_KE
MS01	7	6	5	6	3	4	4	3
MS03	8	9	9	10	10	10	10	10
MS04	5	7	8	7	8	8	9	10
MS05	5	5	6	7	5	7	8	10
MS06	9	9	10	9	8	9	9	8
MS07	9	8	7	7	9	10	10	10
MS08	8	10	10	9	10	10	10	10
MS09	2	2	3	2	8	9	8	9
MS10	6	7	7	8	10	10	10	10
MS12	10	8	9	10	8	10	10	10
MS13	9	8	7	6	9	9	9	8
MS14	7	6	6	6	9	9	9	9
MS16	2	2	4	4	8	5	5	6
MS17	6	7	9	8	4	8	8	10
MS18	4	2	1	2	7	8	7	7

BI	BK	BL	BM	BN	BO	BP	BQ	BR	BS
EMG_LP_BP_S4_R1_LL_VLM	EMG_LP_BP_S4_R2_LL_VLM	EMG_LP_BP_S4_R3_LL_VLM	EMG_LP_BP_S4_R4_LL_VLM	EMG_LP_BP_S4_R5_LL_VLM	EMG_LP_BP_S4_R6_LL_VLM	EMG_LP_BP_S4_R7_LL_VLM	EMG_LP_BP_S4_R8_LL_VLM	EMG_LP_BP_S4_R9_LL_VLM	EMG_LP_BP_S4_R10_LL_VLM
6.02	11.81	11.81	11.81	11.81	11.81	11.81	11.81	11.81	11.81
27.82	27.84	20.68	22.00	21.41	21.41	20.70	22.14	22.14	18.50
26.31	29.08	30.20	31.24	31.11	30.20	30.14	30.81	29.04	31.48
9.87	17.06	21.40	17.40	20.48	17.40	21.52	21.09	20.15	21.83
43.10	30.31	33.03	30.07	31.87	29.20	32.27	30.03	31.88	43.41
27.28	27.10	28.10	28.18	28.18	28.18	28.18	28.18	28.18	27.74
67.89	68.57	71.17	71.58	71.58	71.58	71.58	71.58	71.58	69.83
16.50	22.13	30.70	30.26	25.48	31.17	31.38	31.24	27.18	26.81
12.26	16.45	17.62	17.62	17.62	17.62	17.62	17.62	17.62	12.26
22.27	21.45	28.09	28.09	28.09	28.09	28.09	28.09	28.09	22.27
22.78	27.81	20.10	24.71	24.26	24.57	22.38	22.11	22.83	22.11
23.54	23.85	30.45	30.45	30.45	30.45	30.45	30.45	30.45	23.54
32.71	30.18	5.55	13.50	13.40	13.40	13.50	13.40	13.50	34.53
27.28	27.10	28.10	28.18	28.18	28.18	28.18	28.18	28.18	27.74
22.65	18.61	18.61	18.61	18.61	18.61	18.61	18.61	18.61	22.65
EMG_LP_BP_S4_R1_LL_VLM	EMG_LP_BP_S4_R2_LL_VLM	EMG_LP_BP_S4_R3_LL_VLM	EMG_LP_BP_S4_R4_LL_VLM	EMG_LP_BP_S4_R5_LL_VLM	EMG_LP_BP_S4_R6_LL_VLM	EMG_LP_BP_S4_R7_LL_VLM	EMG_LP_BP_S4_R8_LL_VLM	EMG_LP_BP_S4_R9_LL_VLM	EMG_LP_BP_S4_R10_LL_VLM
25.94	17.83	16.70	16.62	15.68	17.85	17.80	14.10	17.46	15.96
20.17	29.18	31.64	31.05	24.81	31.23	30.70	29.11	27.71	26.50
10.17	15.16	17.70	17.41	17.41	17.41	17.41	17.41	17.41	10.17
48.42	38.18	30.06	31.22	22.60	31.36	31.31	30.39	28.62	30.50
28.09	28.10	28.10	28.10	28.10	28.10	28.10	28.10	28.10	28.09
21.80	24.80	24.80	24.80	24.80	24.80	24.80	24.80	24.80	21.80
20.12	29.43	42.44	39.92	30.68	44.93	43.92	46.12	30.41	34.86
17.60	18.43	23.96	21.48	22.77	18.91	21.13	21.70	21.96	24.14
28.13	29.00	29.71	29.71	29.71	29.71	29.71	29.71	29.71	28.13
16.57	23.52	21.33	19.97	17.09	19.81	21.87	21.41	22.09	20.19
22.61	17.57	17.85	16.30	15.70	17.43	17.43	17.43	17.43	18.72
29.27	41.46	21.59	21.59	21.59	21.59	21.59	21.59	21.59	29.27
31.26	23.24	20.25	20.25	20.25	20.25	20.25	20.25	20.25	31.26
EMG_LP_BP_S4_R1_LL_VLM	EMG_LP_BP_S4_R2_LL_VLM	EMG_LP_BP_S4_R3_LL_VLM	EMG_LP_BP_S4_R4_LL_VLM	EMG_LP_BP_S4_R5_LL_VLM	EMG_LP_BP_S4_R6_LL_VLM	EMG_LP_BP_S4_R7_LL_VLM	EMG_LP_BP_S4_R8_LL_VLM	EMG_LP_BP_S4_R9_LL_VLM	EMG_LP_BP_S4_R10_LL_VLM
11.17	11.17	11.17	11.17	11.17	11.17	11.17	11.17	11.17	11.17
38.87	39.45	39.34	37.84	35.86	38.07	38.93	38.97	38.97	38.76
11.00	14.55	13.34	15.09	17.47	14.20	14.48	14.55	14.50	15.85
13.00	17.49	17.49	17.49	17.49	17.49	17.49	17.49	17.49	13.00
54.58	45.05	39.50	41.49	40.84	42.23	41.79	41.17	40.72	41.19
48.17	48.17	48.17	48.17	48.17	48.17	48.17	48.17	48.17	48.17
13.00	19.51	18.02	18.02	18.02	18.02	18.02	18.02	18.02	13.00
32.51	30.12	27.02	31.86	31.60	34.84	36.55	36.25	30.41	31.86
25.35	22.86	20.81	20.81	20.81	20.81	20.81	20.81	20.81	25.35
17.79	20.99	18.79	22.62	18.16	21.91	22.87	22.20	20.29	22.13
15.10	11.96	11.96	11.96	11.96	11.96	11.96	11.96	11.96	15.10
27.74	20.33	19.97	21.49	21.49	21.49	21.49	21.49	21.49	27.74
12.37	14.06	14.34	10.07	14.87	16.79	16.38	15.86	15.86	17.09
16.26	15.56	15.56	15.56	15.56	15.56	15.56	15.56	15.56	16.26
26.57	20.25	17.97	17.97	17.97	17.97	17.97	17.97	17.97	26.57
EMG_LP_BP_S4_R1_LL_VLM	EMG_LP_BP_S4_R2_LL_VLM	EMG_LP_BP_S4_R3_LL_VLM	EMG_LP_BP_S4_R4_LL_VLM	EMG_LP_BP_S4_R5_LL_VLM	EMG_LP_BP_S4_R6_LL_VLM	EMG_LP_BP_S4_R7_LL_VLM	EMG_LP_BP_S4_R8_LL_VLM	EMG_LP_BP_S4_R9_LL_VLM	EMG_LP_BP_S4_R10_LL_VLM
11.03	11.03	11.03	11.03	11.03	11.03	11.03	11.03	11.03	11.03
48.43	51.80	46.47	46.79	45.04	42.02	43.46	43.28	43.28	47.25
12.11	12.09	12.58	13.38	15.27	12.89	13.50	12.33	12.86	14.33
8.64	11.99	11.72	12.40	11.79	12.40	11.72	14.14	14.06	8.64
40.54	47.82	38.70	40.02	42.69	41.20	39.91	40.59	36.44	39.35
17.35	14.59	15.09	15.09	15.09	15.09	15.09	15.09	15.09	17.35
19.19	25.29	25.36	25.61	28.85	25.59	27.76	26.87	26.34	30.55
27.64	29.18	29.67	31.77	31.78	31.56	28.82	31.21	34.15	29.23
23.86	24.86	24.86	24.86	24.86	24.86	24.86	24.86	24.86	23.86
18.86	21.10	22.48	21.84	21.84	24.96	24.12	25.18	25.18	20.69
19.11	17.69	17.15	17.31	18.14	19.09	20.64	21.10	18.12	22.45
23.15	24.13	24.56	24.56	24.56	24.56	24.56	24.56	24.56	23.15
12.01	11.44	10.29	11.71	13.61	14.59	17.77	19.49	15.51	13.78
24.84	20.49	18.46	21.58	21.57	15.99	43.91	25.97	21.75	31.88
30.33	22.72	24.42	23.45	23.45	23.45	23.45	23.45	23.45	34.77

BT	BV	BW	BX	BY	BZ	CA	CB	CC
EMG_LP_BP_S4_R11_LL_VLM	EMG_LP_BP_S4_R12_LL_VLM	EMG_LP_BP_S4_R13_LL_VLM	EMG_LP_BP_S4_R14_LL_VLM	EMG_LP_BP_S4_R15_LL_VLM	EMG_LP_BP_S4_R16_LL_VLM	EMG_LP_BP_S4_R17_LL_VLM	EMG_LP_BP_S4_R18_LL_VLM	EMG_LP_BP_S4_R19_LL_VLM
13.26	14.26	14.26	14.26	14.26	14.26	14.26	14.26	14.26
38.48	38.48	38.48	38.48	38.48	38.48	38.48	38.48	38.48
24.50	18.45	21.79	20.11	22.28	18.44	18.78	17.32	17.32
36.30	36.30	36.30	36.30	36.30	36.30	36.30	36.30	36.30
29.25	30.28	32.93	36.22	29.98	37.25	38.17	34.51	39.28
71.88	71.88	71.88	71.88	71.88	71.88	71.88	71.88	71.88
22.19	84.43	88.01	92.29	96.99	104.10	71.93	81.57	81.36
24.66	27.57	24.00	26.07	27.25	26.20	28.26	28.09	28.53
33.27	27.22	29.84	29.87	29.76	49.33	74.93	79.72	74.14
39.09	25.33	24.70	22.28	21.50	40.80	103.00	89.09	89.09
36.48	36.48	36.48	36.48	36.48	36.48	36.48	36.48	36.48
14.27	14.25	15.08	15.13	15.08	15.07	15.08	15.08	15.14
11.66	15.29	25.10	19.74	26.83	82.32	76.11	61.20	69.83
25.42	23.22	23.48	23.48	23.48	23.48	23.48	23.48	23.48
EMG_LP_BP_S4_R11_LL_VLM	EMG_LP_BP_S4_R12_LL_VLM	EMG_LP_BP_S4_R13_LL_VLM	EMG_LP_BP_S4_R14_LL_VLM	EMG_LP_BP_S4_R15_LL_VLM	EMG_LP_BP_S4_R16_LL_VLM	EMG_LP_BP_S4_R17_LL_VLM	EMG_LP_BP_S4_R18_LL_VLM	EMG_LP_BP_S4_R19_LL_VLM
26.36	25.20	14.60	16.10	16.10	21.14	21.14	21.14	21.14
16.50	13.37	12.87	12.87	12.87	15.78	16.46	16.51	16.51
27.46	26.21	30.14	27.39	27.68	40.43	48.00	44.31	41.58
11.58	16.06	18.55	15.81	19.34	44.58	15.80	14.03	16.41
39.15	29.82	29.86	32.17	39.51	71.57	40.82	30.30	30.20
34.21	34.21	34.21	34.21	34.21	34.21	34.21	34.21	34.21
26.24	26.21	27.77	27.77	27.77	27.77	27.77	27.77	27.77
34.73	47.46	41.46	46.15	40.17	48.16	40.17	40.17	40.17
33.81	33.81	33.81	33.81	33.81	33.81	33.81	33.81	33.81
36.09	18.37	24.98	23.91	21.31	43.12	61.12	105.76	112.28
27.69	25.13	29.28	29.28	29.28	33.49	139.20	86.17	104.68
18.17	20.00	19.14	19.14	19.14	23.17	33.03	13.47	45.09
30.03	21.84	19.89	18.40	17.58	44.90	92.86	86.72	86.32
20.53	27.77	31.40	30.02	37.80	91.07	91.79	75.99	75.37
30.50	28.89	34.80	34.80	34.80	34.80	34.80	34.80	34.80
EMG_LP_BP_S4_R11_LL_VLM	EMG_LP_BP_S4_R12_LL_VLM	EMG_LP_BP_S4_R13_LL_VLM	EMG_LP_BP_S4_R14_LL_VLM	EMG_LP_BP_S4_R15_LL_VLM	EMG_LP_BP_S4_R16_LL_VLM	EMG_LP_BP_S4_R17_LL_VLM	EMG_LP_BP_S4_R18_LL_VLM	EMG_LP_BP_S4_R19_LL_VLM
34.58	36.27	36.62	41.46	41.46	41.46	41.46	41.46	41.46
16.01	16.09	17.23	14.11	11.30	26.13	25.24	22.05	24.22
20.89	20.97	26.10	19.24	20.34	49.76	75.50	47.56	73.23
35.52	41.86	46.70	46.70	46.70	46.70	46.70	46.70	46.70
16.03	16.16	16.16	16.16	16.16	16.16	16.16	16.16	16.16
17.89	17.72	15.18	15.35	13.79	43.79	46.95	44.05	41.32
25.55	31.77	32.13	36.20	37.80	43.49	40.61	44.41	44.23
30.96	37.08	33.62	33.62	33.62	43.05	74.60	74.60	74.60
27.78	19.43	21.24	22.08	21.06	22.26	40.47	40.47	40.47
13.21	14.40	12.81	14.03	14.03	14.17	16.18	16.18	16.18
32.58	34.37	35.36	32.56	33.23	45.24	45.50	40.84	43.10
16.87	18.61	18.46	18.46	18.46	18.47	18.46	18.46	18.46
15.20	14.76	18.86	19.91	21.56	43.91	41.91	44.86	44.86
50.81	29.31	34.44	39.43	34.23	78.13	84.08	78.18	80.07
EMG_LP_BP_S4_R11_LL_VLM	EMG_LP_BP_S4_R12_LL_VLM	EMG_LP_BP_S4_R13_LL_VLM	EMG_LP_BP_S4_R14_LL_VLM	EMG_LP_BP_S4_R15_LL_VLM	EMG_LP_BP_S4_R16_LL_VLM	EMG_LP_BP_S4_R17_LL_VLM	EMG_LP_BP_S4_R18_LL_VLM	EMG_LP_BP_S4_R19_LL_VLM
35.85	23.51	27.65	27.65	27.65	27.65	27.65	27.65	27.65
43.32	48.15	48.12	50.74	42.75	47.00	43.47	48.92	49.27
24.27	22.97	22.46	22.46	22.46	22.46	22.46	22.46	22.46
11.02	12.08	11.03	11.03	11.03	11.03	11.03	11.03	11.03
34.96	36.18	31.46	47.74	46.55	54.83	74.16	43.25	73.81
15.84	18.13	16.59	24.51	18.71	43.03	71.49	72.09	82.81

CD	CE	CF	CG	CH	CI	CJ	CK	CL	CM
EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM	EMG_UP_H1_S1_R1_UL_VM
55.84	59.75	62.52	67.70	69.45	71.18	73.29	74.58	75.70	76.83
51.58	55.79	58.72	64.53	65.46	79.40	83.55	87.65	91.58	94.13
75.81	74.63	71.29	69.08	69.08	69.08	69.08	69.08	69.08	69.08
56.57	62.29	72.17	65.57	70.63	60.27	72.82	68.25	58.58	57.92
53.52	53.05	52.43	51.13	57.52	63.20	51.14	55.46	51.67	51.89
81.61	82.61	82.61	82.61	82.61	82.61	82.61	82.61	82.61	82.61
75.49	81.10	76.83	64.83	72.24	84.12	63.07	71.96	58.36	70.49
74.99	74.99	66.29	70.00	84.25	43.14	52.02	57.08	69.67	63.96
78.86	85.20	85.20	85.20	84.57	84.57	84.56	84.56	84.56	84.56
90.74	88.98	88.98	84.80	83.53	63.76	91.40	134.98	108.52	91.93
50.11	69.92	66.86	66.86	60.35	66.26	65.49	59.46	62.53	70.51
75.96	70.51	69.29	70.75	80.52	82.38	82.38	82.38	82.38	82.38
73.58	68.20	66.53	59.57	75.91	79.38	64.55	76.32	66.66	52.84
81.61	81.61	81.61	81.61	81.61	81.61	81.61	81.61	81.61	81.61
EMG_UP_H1_S1_R5_UL_VM	EMG_UP_H1_S1_R7_UL_VM	EMG_UP_H1_S1_R9_UL_VM	EMG_UP_H1_S1_R11_UL_VM	EMG_UP_H1_S1_R13_UL_VM	EMG_UP_H1_S1_R15_UL_VM	EMG_UP_H1_S1_R17_UL_VM	EMG_UP_H1_S1_R19_UL_VM	EMG_UP_H1_S1_R21_UL_VM	EMG_UP_H1_S1_R23_UL_VM
76.55	74.05	74.48	77.47	77.09	81.80	83.10	89.31	83.30	75.30
53.76	67.47	65.81	67.19	73.47	56.60	56.60	62.66	55.18	59.18
90.50	90.51	89.53	89.53	89.53	89.53	89.53	89.53	89.53	89.53
82.46	82.21	87.07	58.37	77.78	68.94	82.33	81.77	82.58	83.72
60.81	61.81	61.81	61.81	61.81	61.81	61.81	61.81	61.81	61.81
55.09	59.25	55.11	69.47	58.15	65.09	60.10	64.80	59.48	53.68
92.10	93.60	83.49	80.96	87.69	88.52	89.33	89.64	87.02	80.99
113.74	118.93	118.93	118.93	118.93	118.93	118.93	118.93	118.93	118.93
69.01	82.85	91.51	75.80	86.97	50.34	64.40	71.52	78.99	73.15
99.34	116.73	128.35	99.25	105.73	47.55	78.05	87.90	88.24	91.60
96.28	100.53	100.53	100.53	100.53	100.53	100.53	100.53	100.53	100.53
55.30	75.35	63.40	76.17	67.04	66.96	54.98	52.63	50.35	58.86
102.32	95.57	97.69	98.43	98.53	85.48	102.52	84.59	107.16	94.43
84.49	78.23	73.88	73.88	73.88	73.88	73.88	73.88	73.88	73.88
91.84	100.74	113.94	110.48	109.18	69.71	82.10	81.36	81.36	81.36
EMG_UP_H1_S1_R25_UL_VM	EMG_UP_H1_S1_R27_UL_VM	EMG_UP_H1_S1_R29_UL_VM	EMG_UP_H1_S1_R31_UL_VM	EMG_UP_H1_S1_R33_UL_VM	EMG_UP_H1_S1_R35_UL_VM	EMG_UP_H1_S1_R37_UL_VM	EMG_UP_H1_S1_R39_UL_VM	EMG_UP_H1_S1_R41_UL_VM	EMG_UP_H1_S1_R43_UL_VM
69.84	65.53	71.47	71.51	71.51	62.61	62.61	62.61	62.61	62.61
76.97	80.01	91.73	81.14	81.90	67.55	64.42	66.97	68.25	81.73
85.90	87.92	87.71	80.86	81.00	84.83	78.14	85.73	82.36	85.26
77.78	77.84	62.55	71.12	63.55	74.49	63.11	75.17	76.48	70.65
63.02	62.33	62.73	65.88	66.21	56.63	61.85	56.25	61.31	56.08
68.10	71.97	68.97	71.95	68.97	71.95	70.99	75.41	68.31	75.24
77.04	72.38	69.84	69.84	69.84	69.84	69.84	69.84	69.84	69.84
95.64	91.05	90.05	90.05	89.23	73.13	71.78	80.83	81.47	79.05
75.86	75.86	75.86	75.86	75.86	75.86	75.86	75.86	75.86	75.86
74.84	74.77	76.00	76.17	75.33	57.78	70.22	72.73	65.54	62.16
57.19	81.85	63.04	68.07	68.99	55.17	63.17	89.92	82.16	78.05
102.52	80.13	67.44	60.28	67.44	69.43	64.68	64.68	64.68	64.68
77.66	80.06	80.06	80.06	80.06	80.06	80.06	80.06	80.06	80.06
77.16	68.59	61.13	67.13	88.80	69.98	59.17	72.03	49.02	67.61
102.71	95.93	105.93	105.93	105.93	105.93	105.93	105.93	105.93	105.93
EMG_UP_H1_S1_R45_UL_VM	EMG_UP_H1_S1_R47_UL_VM	EMG_UP_H1_S1_R49_UL_VM	EMG_UP_H1_S1_R51_UL_VM	EMG_UP_H1_S1_R53_UL_VM	EMG_UP_H1_S1_R55_UL_VM	EMG_UP_H1_S1_R57_UL_VM	EMG_UP_H1_S1_R59_UL_VM	EMG_UP_H1_S1_R61_UL_VM	EMG_UP_H1_S1_R63_UL_VM
91.88	90.08	89.83	89.83	90.03	58.18	76.33	75.92	88.24	75.80
91.77	89.81	89.82	89.76	89.76	89.76	89.76	89.76	89.76	89.76
81.58	79.04	81.46	84.40	82.87	72.83	84.47	77.03	80.60	81.84
71.64	68.10	69.41	57.55	64.87	56.58	68.66	64.20	66.52	64.27
73.52	73.52	73.52	73.52	73.52	73.52	73.52	73.52	73.52	73.52
73.35	69.57	81.75	68.75	68.56	60.69	61.94	67.98	69.31	67.76
82.83	89.46	77.09	87.29	104.62	71.23	79.20	87.83	78.66	92.68
96.07	96.07	96.07	96.07	96.07	96.07	96.07	96.07	96.07	96.07
76.92	77.25	74.96	74.96	73.98	63.79	63.79	64.12	65.01	65.65
84.69	84.69	84.69	84.69	84.69	84.69	84.69	84.69	84.69	84.69
68.19	76.83	95.54	81.77	64.81	57.80	90.90	116.89	122.12	96.15
62.97	57.61	50.71	53.22	54.95	51.96	45.31	39.72	51.19	51.84
102.81	102.81	102.81	102.81	102.81	102.81	102.81	102.81	102.81	102.81
104.75	75.60	76.69	90.83	116.40	68.76	58.45	79.94	71.72	65.66
98.91	95.69	101.16	96.97	84.14	49.22	60.21	75.31	89.45	83.19

CN	CO	CP	CQ	CR	CS	CT	CU	CV	CW
EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM	EMG_UP_H2_S1_R1_UL_VM
80.75	59.05	56.37	51.09	30.90	66.12	81.24	54.81	50.94	50.94
44.86	67.11	67.14	73.01	80.95	69.00	67.11	69.09	69.09	69.09
59.97	63.40	63.40	68.78	77.45	81.29	100.43	81.29	118.06	106.62
76.15	81.98	81.98	89.23	83.78	58.17	79.40	61.82	76.72	73.60
65.82	60.83	73.69	64.79	63.48	66.30	59.02	61.93	61.93	61.93
50.73	46.47	47.91	53.39	47.30	48.30	49.55	48.30	45.16	52.06
78.90	77.20	78.01	57.81	77.22	68.47	66.18	75.90	68.10	63.79
64.10	65.35	65.22	65.53	62.79	71.53	69.79	59.38	64.50	64.50
75.84	81.93	81.93	81.93	81.93	81.93	81.93	81.93	81.93	81.93
50.43	69.59	86.41	78.47	88.52	61.45	80.68	87.99	85.15	73.18
80.05	81.81	84.87	82.87	80.87	80.87	80.87	80.87	80.87	80.87
61.10	54.52	74.16	69.56	69.20	62.54	64.12	62.54	67.74	58.37
73.58	77.04	78.19	81.77	88.00	80.13	74.18	65.09	58.99	70.14
93.03	78.55	71.26	67.56	64.46	66.95	75.71	79.84	71.99	69.19
96.65	81.30	90.49	113.62	113.60	67.13	88.35	78.78	74.82	84.25
EMG_UP_H2_S1_R5_UL_VM	EMG_UP_H2_S1_R7_UL_VM	EMG_UP_H2_S1_R9_UL_VM	EMG_UP_H2_S1_R11_UL_VM	EMG_UP_H2_S1_R13_UL_VM	EMG_UP_H2_S1_R15_UL_VM	EMG_UP_H2_S1_R17_UL_VM	EMG_UP_H2_S1_R19_UL_VM	EMG_UP_H2_S1_R21_UL_VM	EMG_UP_H2_S1_R23_UL_VM
65.44	61.77	66.77	66.82	66.82	66.82	66.82	66.82	66.82	66.82
95.31	91.92	90.97	92.92	94.05	73.70	78.30	76.52	75.85	76.36
84.96	79.32	82.61	90.09	88.74	59.96	76.60	67.48	74.54	73.49
67.03	70.36	68.83	69.84	69.84	81.32	84.06	61.13	63.83	53.48
56.72	49.50	54.82	59.46	54.21	47.08	55.00	61.11	43.50	52.99
76.79	71.03	76.10	81.77	80.11	63.15	66.88	77.58	67.08	73.23
95.64	81.65	91.74	85.92	87.51	59.69	71.27	74.69	78.69	88.73
92.98	100.70	95.33	82.19	91.70	72.19	91.66	101.12	66.86	66.60
106.17	109.18	112.09	109.97	114.99	73.92	90.12	103.12	116.44	103.57
94.63	94.68	103.13	100.79	117.43	86.57	111.83	90.03	90.03	94.29
63.63	59.10	49.25	72.93	73.33	52.06	64.60	51.36	57.48	46.34
98.55	98.67	106.34	107.73	110.18	69.67	112.66	69.09	69.09	114.41
64.32	81.24	80.44	80.60	80.60	95.48	90.33	90.33	91.41	91.41
106.81	83.56	88.64	119.02	124.99	63.06	70.98	67.00	67.48	74.80
EMG_UP_H2_S1_R25_UL_VM	EMG_UP_H2_S1_R27_UL_VM	EMG_UP_H2_S1_R29_UL_VM	EMG_UP_H2_S1_R31_UL_VM	EMG_UP_H2_S1_R33_UL_VM	EMG_UP_H2_S1_R35_UL_VM	EMG_UP_H2_S1_R37_UL_VM	EMG_UP_H2_S1_R39_UL_VM	EMG_UP_H2_S1_R41_UL_VM	EMG_UP_H2_S1_R43_UL_VM
67.77	77.36	65.11	58.15	66.15	44.18	57.67	62.27	61.39	65.54
74.84	71.65	84.91	90.29	96.35	68.84	86.57	70.28	72.15	81.30
105.16	100.89	113.01	105.65	113.71	80.01	86.56	80.12	69.06	69.52
71.08	77.86	74.91	81.18	67.85	69.11	57.03	69.04	79.04	71.54
60.94	63.38	65.12	62.78	63.88	66.36	67.31	58.31	55.47	53.09
65.17	65.28	64.50	64.74	66.58	52.03	68.04	68.40	52.08	63.73
85.27	77.54	85.85	81.86	100.71	71.90	78.62	81.58	87.56	73.57
88.13	87.42	79.78	90.32	112.78	79.89	89.73	88.96	76.54	81.35
87.63	78.37	68.54	62.92	72.92	62.92	62.92	62.92	62.92	62.92
73.22	69.82	71.88	80.17	94.78	55.21	80.78	62.94	72.52	76.75
67.60	75.03	69.14	58.86	56.14	53.27	64.68	69.43	59.87	75.09
65.37	71.55	58.30	64.60	71.30	74.88	60.98	73.57	69.21	63.20
73.88	92.54	103.85	106.32	86.20	86.20	86.20	79.61	86.95	80.51
64.81	79.84	63.03	74.28	68.65	83.10	65.29	62.39	62.39	82.95
97.66	83.33	105.14	105.63	114.66	68.81	62.54	53.17	61.58	50.54
EMG_UP_H2_S1_R45_UL_VM									

CG	CY	CZ	DA	DB	DC	DD	DE	DF	DG
EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM
59.07	59.08	54.98	52.04	72.29	53.75	52.22	53.23	55.18	67.52
71.58	71.57	80.77	80.11	84.84	70.13	70.19	61.69	72.74	65.29
134.80	106.13	110.26	110.26	112.98	72.63	72.63	72.63	82.27	57.47
85.94	86.03	74.14	82.98	69.44	64.67	63.75	70.47	67.76	74.42
72.71	86.02	87.60	87.34	74.77	72.00	64.03	61.15	61.00	61.00
45.99	46.67	46.60	46.55	46.33	58.99	61.43	50.17	53.15	74.36
69.27	59.43	69.08	78.07	67.17	50.43	62.97	70.19	66.96	72.37
75.25	74.84	65.53	63.66	65.97	63.96	63.96	63.11	69.56	66.54
71.51	61.59	65.98	73.89	73.36	56.49	76.24	76.24	67.63	68.95
80.15	73.36	78.16	93.56	88.68	65.99	78.35	76.48	84.99	76.76
103.94	102.09	102.24	85.04	78.40	74.85	65.00	63.00	69.50	81.28
70.53	55.79	59.27	56.28	59.68	43.77	47.64	47.64	54.84	58.64
79.10	67.73	103.00	85.24	94.02	69.84	62.28	72.88	80.07	72.35
52.80	51.06	68.01	73.59	81.77	93.40	65.94	83.14	82.15	65.14
85.10	81.78	91.81	92.57	86.14	80.31	79.22	80.31	82.15	72.95
EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM
65.00	63.79	69.05	82.22	74.04	65.66	68.37	72.32	68.08	67.62
77.64	73.23	84.30	79.50	89.32	71.98	86.22	78.52	80.31	85.76
73.75	69.13	81.65	73.82	70.09	55.90	73.11	68.19	69.86	70.90
68.06	72.96	76.38	74.42	69.58	86.54	65.63	71.96	66.51	63.80
45.67	47.19	46.99	48.63	49.12	50.31	53.22	85.99	59.38	58.81
68.86	74.05	67.24	69.20	71.75	63.54	64.07	68.68	67.76	69.79
97.14	88.67	80.54	80.46	82.66	66.97	64.29	82.74	81.81	85.90
79.10	79.30	80.15	98.08	83.95	78.24	68.19	87.97	75.07	82.52
112.55	108.65	122.39	122.01	123.89	123.89	126.31	126.34	58.69	102.74
112.43	93.09	109.94	93.50	80.62	100.22	78.88	100.22	118.20	84.93
63.75	52.19	58.96	53.75	66.10	60.05	52.32	89.93	56.35	58.08
86.48	100.13	118.37	118.54	113.28	85.26	76.34	80.42	89.85	103.67
63.02	71.59	62.36	85.30	80.31	92.06	78.78	84.10	76.01	77.15
89.54	108.75	81.61	80.89	88.66	52.99	77.05	81.67	67.90	135.62
EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM
69.17	55.77	65.35	61.93	52.37	51.69	51.69	67.93	67.93	67.93
74.95	88.67	74.58	87.39	93.17	66.11	78.36	73.65	71.33	80.94
84.12	86.26	92.26	86.71	83.86	80.00	81.80	81.80	81.80	81.80
73.87	72.51	81.25	71.09	81.02	70.72	75.24	72.20	79.21	79.21
67.24	69.48	61.02	59.51	65.07	65.00	65.41	61.02	66.78	57.64
47.60	61.60	71.87	59.56	70.14	75.86	62.65	70.14	69.46	69.46
64.15	68.01	89.58	83.12	85.72	77.81	78.59	92.67	81.70	74.15
72.26	80.85	76.03	88.45	78.30	78.57	69.13	82.32	76.65	89.90
65.80	64.86	69.61	69.61	74.64	74.64	66.57	66.56	70.66	69.80
61.81	67.52	80.41	77.47	89.49	59.28	65.14	75.77	61.92	61.92
71.57	67.34	67.10	51.49	61.25	47.98	52.11	87.66	61.28	58.90
65.50	64.26	66.43	75.41	75.16	75.16	76.40	59.87	61.56	61.56
76.35	58.55	77.21	70.52	76.43	75.03	64.21	84.21	89.80	81.01
63.10	85.45	54.47	78.38	77.61	85.34	76.83	74.64	74.52	54.68
70.18	80.06	80.36	80.36	88.51	68.01	64.01	71.78	68.01	68.01
EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM	EMG_UP_H3_R3_LL_VLM
76.04	74.35	87.72	78.11	78.88	54.94	55.88	73.00	75.16	86.11
82.25	87.41	101.94	87.41	84.64	77.78	81.17	81.43	79.07	81.29
86.64	85.66	84.56	93.34	83.06	80.45	80.39	87.37	80.45	87.21
77.12	59.21	73.71	61.90	64.10	53.86	54.00	71.57	57.61	63.62
66.86	66.84	66.89	66.89	66.83	71.00	68.77	68.89	68.89	68.89
46.52	59.02	53.16	42.68	51.94	51.94	54.36	54.36	56.50	53.44
85.56	86.73	92.29	84.12	85.45	73.72	83.43	104.11	86.60	81.08
49.69	67.69	80.14	74.25	74.25	72.16	71.42	77.59	72.07	83.89
77.31	91.31	66.81	78.76	76.09	56.93	63.90	73.07	69.47	69.47
100.94	79.52	88.26	94.26	113.51	58.15	85.16	88.52	83.93	78.48
109.97	83.87	83.87	83.87	83.87	80.80	80.80	102.52	102.52	102.52
63.68	58.86	47.88	58.83	64.24	53.49	43.57	46.23	45.95	45.90
86.43	110.84	118.19	107.20	100.96	84.84	94.61	97.34	113.80	95.95
71.24	71.16	62.42	71.14	66.21	81.17	74.22	74.22	74.87	61.12
70.10	86.06	79.49	78.87	45.31	60.57	60.50	84.84	84.84	76.40

DI	DI	DI	DI	DI	DI	DI	DI	DI	DI
EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM
87.11	87.07	53.15	58.89	60.01	60.01	60.01	60.01	60.01	60.01
71.25	53.19	47.63	47.08	60.29	60.29	60.29	60.29	60.29	60.29
50.48	76.77	76.35	75.99	83.93	83.93	83.93	83.93	83.93	83.93
66.86	47.06	81.04	72.99	72.94	72.94	72.94	72.94	72.94	72.94
49.37	48.24	50.04	60.94	51.78	51.78	51.78	51.78	51.78	51.78
68.08	68.08	68.08	68.08	68.08	68.08	68.08	68.08	68.08	68.08
72.85	69.45	71.01	67.84	70.52	70.52	70.52	70.52	70.52	70.52
61.51	61.51	61.51	61.51	61.51	61.51	61.51	61.51	61.51	61.51
89.10	87.42	86.45	80.37	86.65	86.65	86.65	86.65	86.65	86.65
61.86	61.82	55.24	60.45	61.84	61.84	61.84	61.84	61.84	61.84
81.07	81.07	81.07	81.07	81.07	81.07	81.07	81.07	81.07	81.07
68.41	70.10	66.30	94.78	73.01	73.01	73.01	73.01	73.01	73.01
69.88	69.88	69.88	69.88	69.88	69.88	69.88	69.88	69.88	69.88
EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM
61.72	76.20	62.99	72.11	75.81	75.81	75.81	75.81	75.81	75.81
51.86	67.61	54.77	71.59	74.16	74.16	74.16	74.16	74.16	74.16
84.40	78.53	71.38	70.89	76.89	76.89	76.89	76.89	76.89	76.89
71.29	83.26	71.24	83.87	65.80	65.80	65.80	65.80	65.80	65.80
64.00	65.91	62.50	81.28	75.22	75.22	75.22	75.22	75.22	75.22
52.14	58.18	54.10	67.10	64.32	64.32	64.32	64.32	64.32	64.32
74.84	67.76	69.09	74.00	69.61	69.61	69.61	69.61	69.61	69.61
88.87	86.46	97.34	94.45	81.43	81.43	81.43	81.43	81.43	81.43
100.96	100.96	100.96	100.96	100.96	100.96	100.96	100.96	100.96	100.96
88.77	100.89	111.18	120.74	120.47	120.47	120.47	120.47	120.47	120.47
83.66	89.51	89.51	84.02	76.91	76.91	76.91	76.91	76.91	76.91
52.47	60.34	60.38	63.51	70.31	70.31	70.31	70.31	70.31	70.31
85.12	101.09	77.71	95.18	83.45	83.45	83.45	83.45	83.45	83.45
51.14	65.50	66.82	66.82	65.58	65.58	65.58	65.58	65.58	65.58
69.46	69.46	69.46	69.46	69.46	69.46	69.46	69.46	69.46	69.46
EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM	EMG_UP_H4_R4_LL_VLM
61.00	61.00	61.00	61.00	61.00	61.00	61.00	61.00	61.00	61.00
100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
76.38	90.07	61.19	88.00	74.48	74.48	74.48	74.48	74.48	74.48
51.87	59.50	56.22	42.30	71.74	71.74	71.74	71.74	71.74	71.74
72.24	60.24	62.43	68.13	63.44	63.44	63.44	63.44	63.44	63.44
84.66	92.89	79.84	81.54	91.86	91.86	91.86	91.86	91.86	91.86
6									

Surface Electromyography: Knee Extension

A	B	C	D	E	F	G	H	I	J	K
MR01	25.66	25.44	25.44	25.44	25.44	25.44	25.44	25.44	25.44	25.44
MR03	27.77	28.43	29.38	29.38	27.15	24.03	25.07	24.43	28.32	25.06
MR04	25.92	43.39	44.73	57.67	56.82	55.65	43.75	53.07	50.59	51.21
MR05	51.41	46.61	60.28	53.19	50.11	41.11	46.90	56.59	56.07	51.20
MR06	25.02	30.60	24.37	37.30	36.79	34.23	37.81	34.23	37.81	44.23
MR07	40.76	46.88	46.57	46.47	41.45	46.29	54.43	44.84	52.51	58.99
MR08	37.88	34.58	32.39	32.39	32.39	32.39	32.39	32.39	32.39	32.39
MR09	38.62	41.20	41.80	44.80	43.30	44.34	44.34	44.34	44.34	44.34
MR10	37.80	46.43	42.80	47.04	54.17	53.57	53.22	53.15	55.83	57.33
MR11	23.77	43.23	43.69	43.69	43.69	43.69	43.69	43.69	43.69	43.69
MR12	33.01	34.88	38.94	43.30	41.19	41.68	45.27	45.84	48.22	47.62
MR14	34.72	38.15	39.28	32.39	32.39	41.28	48.11	43.74	45.75	46.11
MR15	33.94	43.21	40.40	40.40	40.40	40.40	40.40	40.40	40.40	40.40
MR17	36.66	39.03	35.10	44.13	40.60	36.43	40.21	43.96	46.33	47.82
MR18	32.58	34.35	35.93	36.36	43.87	38.64	38.29	45.87	44.76	50.73

L	M	N	O	P	Q	R	S	T	U
29.90	27.06	28.06	25.80	26.67	31.86	24.20	38.39	35.16	35.78
25.11	27.40	28.88	31.66	28.98	28.72	28.54	28.51	31.91	31.91
35.09	37.88	47.64	53.51	57.58	56.88	60.51	60.51	61.77	61.88
40.59	63.89	57.67	63.59	60.82	69.60	62.58	62.58	76.77	64.55
40.69	47.67	37.51	38.30	37.05	36.87	43.72	35.75	48.49	35.07
46.87	57.59	61.59	68.44	66.12	69.69	61.62	61.62	61.78	61.78
60.09	77.59	74.80	82.02	81.23	86.07	87.03	87.03	76.66	80.80
50.09	48.10	51.54	58.12	53.02	56.93	56.47	59.72	53.12	55.85
57.21	54.86	52.00	56.04	56.04	59.47	61.56	61.56	61.78	61.78
48.80	50.88	49.14	52.82	54.86	53.15	57.25	54.37	66.45	50.95
48.40	48.89	47.77	43.58	42.64	39.33	44.36	43.66	50.56	41.11
53.88	53.01	62.85	58.65	62.41	61.10	65.26	64.38	60.81	65.12
43.84	46.14	33.02	44.49	44.49	48.71	48.07	48.07	41.00	43.80
48.78	45.09	43.53	45.31	45.31	52.08	46.50	46.50	47.90	47.90
54.21	43.69	55.36	55.33	53.71	55.43	53.81	57.73	51.84	55.25

V	W	X	Y	Z	AA	AB	AC	AD	AE
31.02	40.03	34.76	29.88	33.71	38.21	38.10	28.92	27.84	36.80
32.55	31.51	37.51	34.13	34.53	33.14	39.14	33.51	40.54	36.80
71.39	66.19	58.87	63.22	57.36	64.72	61.23	64.18	67.38	70.88
73.30	73.51	75.06	70.14	75.97	61.66	74.84	68.00	75.49	70.88
52.80	40.88	44.88	44.88	53.88	48.88	48.88	48.88	48.88	48.88
55.00	65.40	66.72	75.93	59.97	72.87	79.40	62.03	82.96	76.72
75.24	80.13	75.40	68.12	72.35	89.56	70.06	75.55	78.90	77.76
44.82	24.92	61.75	55.36	50.68	45.99	44.32	54.13	54.04	57.01
71.96	72.77	63.51	75.00	78.73	79.45	67.95	69.20	70.20	76.61
60.02	64.18	72.97	71.22	73.31	73.11	69.14	73.86	64.57	68.09
47.88	40.52	44.75	46.80	44.15	44.87	48.29	45.09	40.92	46.19
56.01	61.65	54.57	52.12	53.55	48.29	54.65	51.66	53.06	55.70
51.07	58.71	52.23	43.52	45.31	45.31	53.42	55.51	48.31	55.09
49.38	53.06	44.19	48.44	56.45	52.70	55.14	55.51	49.81	48.50
53.58	48.18	48.89	48.52	58.18	61.13	61.81	54.61	63.03	62.05
EMG_RL_RFR_S1_R21_LL_VL	EMG_RL_RFR_S1_R22_LL_VL	EMG_RL_RFR_S1_R23_LL_VL	EMG_RL_RFR_S1_R24_LL_VL	EMG_RL_RFR_S1_R25_LL_VL	EMG_RL_RFR_S1_R26_LL_VL	EMG_RL_RFR_S1_R27_LL_VL	EMG_RL_RFR_S1_R28_LL_VL	EMG_RL_RFR_S1_R29_LL_VL	EMG_RL_RFR_S1_R30_LL_VL
58.13	58.77	60.86	51.87	50.95	48.87	48.87	35.51	59.83	74.83
36.25	37.31	38.75	34.90	35.89	36.68	44.24	40.51	42.08	44.97
78.08	73.89	57.00	59.96	63.03	69.48	68.29	68.22	69.85	73.87
8.00	80.79	76.37	73.41	86.98	81.24	79.01	85.17	80.99	82.69
66.83	72.04	66.14	66.74	72.53	68.29	75.59	73.79	73.80	66.01
73.73	90.11	87.46	101.85	81.17	88.37	111.89	81.75	115.87	104.39
57.13	60.81	60.34	62.86	60.03	65.37	65.11	62.58	66.84	65.82
92.44	66.12	71.75	73.57	63.09	53.55	57.97	63.17	71.90	69.72
79.94	81.61	78.56	78.56	73.58	73.09	81.11	74.29	74.29	73.27
57.79	56.59	72.14	79.71	77.45	93.69	80.61	83.96	71.36	71.36
32.73	30.64	26.07	28.87	24.33	24.84	23.43	28.18	27.61	27.61
30.41	33.55	37.75	41.30	52.51	53.63	58.82	53.28	57.78	58.86
48.55	43.65	43.14	45.48	45.48	38.17	38.17	41.88	40.80	43.19
47.49	41.83	44.47	43.51	46.96	43.51	46.96	43.51	46.96	43.51
74.43	68.64	72.07	78.48	68.60	73.05	87.47	75.77	84.36	70.07
EMG_RL_RFR_S1_R31_LL_VL	EMG_RL_RFR_S1_R32_LL_VL	EMG_RL_RFR_S1_R33_LL_VL	EMG_RL_RFR_S1_R34_LL_VL	EMG_RL_RFR_S1_R35_LL_VL	EMG_RL_RFR_S1_R36_LL_VL	EMG_RL_RFR_S1_R37_LL_VL	EMG_RL_RFR_S1_R38_LL_VL	EMG_RL_RFR_S1_R39_LL_VL	EMG_RL_RFR_S1_R40_LL_VL
47.81	41.80	43.43	52.09	56.47	46.18	44.47	55.01	49.35	72.81
62.22	65.17	70.18	65.57	68.58	59.30	64.47	70.42	51.92	64.45
59.84	60.20	53.11	54.47	50.17	59.80	59.17	60.39	61.31	61.31
49.74	47.86	61.80	50.86	59.58	53.43	52.46	59.38	51.15	56.83
78.76	88.27	72.48	76.19	85.93	82.67	79.66	80.12	79.84	73.27
51.03	51.85	60.80	52.04	53.31	59.01	54.05	53.93	58.77	68.79
49.62	49.62	47.70	52.17	52.76	52.76	52.76	52.76	52.76	52.76
55.40	29.18	35.59	46.07	45.94	37.25	37.25	41.41	35.51	36.00
61.80	78.02	76.03	75.05	77.15	77.80	74.48	72.07	76.32	73.02
56.64	28.00	48.57	48.54	61.75	61.75	61.75	61.75	69.80	69.80
54.53	37.50	40.87	44.58	49.29	46.01	43.74	43.74	39.81	43.28
61.62	68.44	58.77	60.40	53.97	52.39	60.32	55.11	67.47	60.70
60.68	60.29	61.18	59.26	60.60	60.60	60.60	60.60	60.60	60.60
56.70	76.26	53.30	55.38	60.25	55.58	66.00	52.29	57.47	61.65
61.55	58.82	71.82	59.22	59.22	65.22	67.85	66.84	60.74	73.39
EMG_RL_RFR_S1_R41_LL_VL	EMG_RL_RFR_S1_R42_LL_VL	EMG_RL_RFR_S1_R43_LL_VL	EMG_RL_RFR_S1_R44_LL_VL	EMG_RL_RFR_S1_R45_LL_VL	EMG_RL_RFR_S1_R46_LL_VL	EMG_RL_RFR_S1_R47_LL_VL	EMG_RL_RFR_S1_R48_LL_VL	EMG_RL_RFR_S1_R49_LL_VL	EMG_RL_RFR_S1_R50_LL_VL
302.23	41.81	41.81	71.61	41.81	41.81	71.61	41.81	41.81	41.81
52.62	49.89	52.63	46.99	53.22	45.48	51.15	46.62	53.73	51.87
52.86	51.48	48.30	51.22	40.83	46.43	45.57	51.86	51.02	58.37
58.03	42.43	62.77	62.02	62.02	59.69	61.64	61.04	56.17	61.04
57.19	60.84	51.90	55.82	54.63	54.24	57.79	56.89	53.05	51.15
57.28	65.08	68.72	59.87	68.24	78.49	58.32	64.67	67.62	77.06
63.75	57.60	64.19	66.90	63.57	60.67	71.79	49.12	59.87	63.96
33.68	24.57	27.06	27.04	29.52	29.52	29.52	29.52	29.52	29.52
67.90	70.48	66.30	81.04	72.11	78.00	76.19	75.67	70.00	79.90
51.54	26.45	46.78	50.73	59.14	62.76	63.75	61.33	61.59	61.59
42.83	31.83	36.39	36.07	34.23	34.23	34.23	34.23	34.23	34.23
62.00	60.44	52.26	61.91	46.51	57.39	48.14	53.72	58.77	54.65
45.62	52.89	42.10	40.97	51.46	54.49	49.57	60.63	51.07	46.52
50.60	46.40	56.50	60.50	60.48	60.48	60.48	60.48	60.48	60.48
53.01	57.11	55.27	60.70	57.98	64.73	58.79	68.84	70.05	68.84
EMG_RL_RFR_S2_R1_LL_VL	EMG_RL_RFR_S2_R2_LL_VL	EMG_RL_RFR_S2_R3_LL_VL	EMG_RL_RFR_S2_R4_LL_VL	EMG_RL_RFR_S2_R5_LL_VL	EMG_RL_RFR_S2_R6_LL_VL	EMG_RL_RFR_S2_R7_LL_VL	EMG_RL_RFR_S2_R8_LL_VL	EMG_RL_RFR_S2_R9_LL_VL	EMG_RL_RFR_S2_R10_LL_VL
35.93	29.52	52.09	42.61	85.50	36.31	38.84	34.17	34.90	30.81
36.92	29.37	29.50	28.79	25.57	36.44	32.83	35.11	45.00	38.12
60.48	63.82	63.97	59.20	63.68	58.41	55.40	65.90	69.24	68.86
72.72	60.58	60.58	59.14	59.13	57.81	57.84	54.25	54.25	54.25
40.44	34.03	42.03	42.03	42.44	47.12	42.03	40.53	40.54	41.18
43.32	32.88	40.55	38.07	46.87	49.43	47.06	49.12	51.38	37.77
61.50	60.63	60.60	60.60	60.60	60.60	60.60	60.60	60.60	60.60
55.47	63.95	58.81	48.63	47.73	47.73	47.73	64.03	55.81	61.03
45.36	53.51	59.86	57.95	57.92	57.92	62.11	59.83	63.59	61.26
44.09	44.23	45.86	54.19	49.16	52.53	57.50	51.43	61.59	62.54
33.76	33.96	33.96	33.96	33.96	33.96	33.96	33.96	33.96	33.96
30.27	44.27	46.86	44.11	50.42	50.09	49.57	44.80	49.86	51.70
31.13	33.45	33.45	34.28	31.85	35.12	38.33	44.62	40.22	38.81
52.79	53.21	47.90	44.48	45.59	44.58	44.58	44.58	44.58	44.58
42.28	40.86	40.87	39.88	42.87	40.49	39.88	33.52	46.62	40.00
EMG_RL_RFR_S2_R11_LL_VL	EMG_RL_RFR_S2_R12_LL_VL	EMG_RL_RFR_S2_R13_LL_VL	EMG_RL_RFR_S2_R14_LL_VL	EMG_RL_RFR_S2_R15_LL_VL	EMG_RL_RFR_S2_R16_LL_VL	EMG_RL_RFR_S2_R17_LL_VL	EMG_RL_RFR_S2_R18_LL_VL	EMG_RL_RFR_S2_R19_LL_VL	EMG_RL_RFR_S2_R20_LL_VL
66.44	62.88	62.09	73.23	68.44	58.60	56.70	60.40	66.20	63.33
31.58	35.24	34.40	31.42	29.25	44.75	36.24	36.24	46.95	35.12
64.97	52.13	58.87	56.71	55.73	65.00	69.09	69.09	73.28	65.74
71.79	65.93	59.48	59.37	64.77	57.69	67.35	62.96	62.67	65.73
58.96	66.78	66.78	66.78	66.78	66.78	66.78	66.78	66.78	66.78
57.67	52.97	54.39	52.15	50.61	65.65	65.65	65.65	73.94	80.30
48.18	48.07	49.65	48.89	50.71	51.70	49.37	55.35	54.23	56.89
43.80	55.99	53.12	54.65	50.29	55.11	48.86	74.55	58.28	62.36
43.88	50.48	46.58	52.53	56.78	60.90	61.98	61.98	61.98	61.98
44.13	51.43	47.22	48.55	60.26	54.73	61.10	60.26	59.10	65.90
24.04	23.84	25.15	25.58	23.60	21.26	21.26	21.26	24.57	25.73
31.18	38.63	42.61	42.61	42.61	39.31	45.46	43.16	48.13	48.13
43.08	34.98	36.68	34.96	33.80	33.80	33.80	40.83	38.13	45.13
43.18	45.10	40.83	39.09	41.53	38.88	40.46	41.58	41.58	41.58
48.20	53.39	51.72	54.17	52.97	52.15	47.15	64.49	62.95	61.77
EMG_RL_RFR_S2_R21_LL_VL	EMG_RL_RFR_S2_R22_LL_VL	EMG_RL_RFR_S2_R23_LL_VL	EMG_RL_RFR_S2_R24_LL_VL	EMG_RL_RFR_S2_R25_LL_VL	EMG_RL_RFR_S2_R26_LL_VL	EMG_RL_RFR_S2_R27_LL_VL	EMG_RL_RFR_S2_R28_LL_VL	EMG_RL_RFR_S2_R29_LL_VL	EMG_RL_RFR_S2_R30_LL_VL
25.04	24.09	24.09	24.09	25.23	24.54	24.54	24.54	24.54	24.54
56.66	54.14	47.83	46.38	48.18	53.43	55.99	58.13	56.84	56.84
60.62	55.45	66.96	59.47	53.88	50.55	59.94	59.15	66.78	63.94
47.61	47.46	47.46	47.46	47.46	47.46	47.46	47.46	47.46	47.46
69.89	75.98	76.09	56.61	58.06	60.71	59.86	64.98	73.90	73.90
43.09	43.09	44.23	38.99	41.14	39.84	42.95	41.64	44.77	44.27
60.01	43.00	46.36	43.00	37.13	37.13	37.13	37.13	42.54	42.54
55.05	40.80	47.46	40.49	41.03	47.36	39.51	41.05	50.87	45.73
51.18	52.01	64.37	61.13	60.41	60.41	59.29	55.82	64.52	64.52
44.73	44.85	50.47	44.41	50.15	51.34	59.31	61.32	58.27	60.81
30.22	30.35	29.43	31.06	31.21	27.84	30.95	31.84	30.98	29.70
43.39	44.15	57.88	56.20	52.86	56.47	51.78	51.78	48.13	58.74
50.48	66.81	66.71	77.31	68.94	69.05	72.79	75.56	79.95	79.91
42.14	59.88	51.69	53.68	46.09	49.13	48.19	49.34	52.31	51.83
39.87	48.10	50.93	44.71	61.36	52.06	65.38	63.25	65.15	55.15
EMG_RL_RFR_S2_R31_LL_VL	EMG_RL_RFR_S2_R32_LL_VL	EMG_RL_RFR_S2_R33_LL_VL	EMG_RL_RFR_S2_R34_LL_VL	EMG_R					

AP	AQ	AR	AS	AT	AU	AV	AW	AX	AY
EMG_KF_RFR_S2_R11_UL_VM	EMG_KF_RFR_S2_R12_UL_VM	EMG_KF_RFR_S2_R13_UL_VM	EMG_KF_RFR_S2_R14_UL_VM	EMG_KF_RFR_S2_R15_UL_VM	EMG_KF_RFR_S2_R16_UL_VM	EMG_KF_RFR_S2_R17_UL_VM	EMG_KF_RFR_S2_R18_UL_VM	EMG_KF_RFR_S2_R19_UL_VM	EMG_KF_RFR_S2_R20_UL_VM
20.69	43.29	28.19	28.27	40.37	30.47	28.08	33.08	33.60	29.60
39.56	39.54	44.80	38.02	41.47	31.10	31.49	31.49	31.26	37.71
69.76	58.57	60.91	71.79	67.67	58.95	61.27	58.38	65.54	62.57
71.12	64.64	75.40	63.63	72.34	62.43	55.34	56.95	56.43	59.23
39.84	43.27	43.60	40.77	42.43	37.48	37.48	37.79	37.79	41.78
63.45	71.26	62.93	77.10	76.46	38.08	48.83	43.13	50.44	59.23
67.32	73.69	63.42	70.02	68.81	61.89	63.30	77.92	75.46	72.86
53.19	51.09	58.41	54.93	53.82	53.91	50.70	59.52	48.18	53.79
65.99	65.81	78.75	63.41	70.43	51.60	50.28	54.88	62.49	62.49
63.04	62.91	68.90	70.75	65.31	39.00	48.63	44.78	51.05	64.42
36.60	41.04	33.18	39.91	41.49	32.11	41.68	38.64	29.52	35.09
48.41	56.79	52.80	47.95	51.54	44.58	44.58	45.99	40.89	48.62
42.46	44.48	46.41	44.41	38.83	35.45	37.97	33.80	37.69	35.75
50.64	61.31	69.01	58.99	62.09	58.44	58.35	44.28	44.72	44.91
54.34	63.08	67.08	65.47	65.31	65.31	65.31	61.79	61.79	61.79
EMG_KF_RFR_S2_R11_LL_VM	EMG_KF_RFR_S2_R12_LL_VM	EMG_KF_RFR_S2_R13_LL_VM	EMG_KF_RFR_S2_R14_LL_VM	EMG_KF_RFR_S2_R15_LL_VM	EMG_KF_RFR_S2_R16_LL_VM	EMG_KF_RFR_S2_R17_LL_VM	EMG_KF_RFR_S2_R18_LL_VM	EMG_KF_RFR_S2_R19_LL_VM	EMG_KF_RFR_S2_R20_LL_VM
42.15	59.91	52.73	63.22	60.61	60.61	64.64	62.70	62.70	62.70
43.75	42.27	46.11	43.04	42.34	37.57	38.21	43.58	40.51	38.09
58.80	66.14	73.50	62.99	69.79	61.75	60.60	63.89	60.45	64.84
67.66	72.65	85.99	69.84	84.65	63.05	61.65	68.17	59.86	68.10
55.94	73.73	84.03	87.06	69.50	51.08	57.13	66.74	65.92	60.43
91.89	95.85	95.03	113.02	110.10	48.34	62.17	66.72	68.35	74.61
55.79	60.55	62.49	64.63	65.04	47.22	50.90	52.69	52.90	49.17
49.37	52.21	55.91	62.31	59.92	44.38	49.22	56.56	52.74	56.27
63.59	65.84	65.01	71.71	74.09	45.61	44.72	51.95	51.50	59.48
73.69	78.50	78.50	73.40	69.32	39.56	52.43	47.53	53.52	50.79
37.38	29.77	22.85	28.38	33.29	28.89	31.78	28.04	23.85	26.28
44.57	61.21	57.68	53.85	56.76	42.49	44.46	47.20	39.55	49.39
42.64	46.12	44.26	43.26	41.66	40.78	40.78	42.58	37.46	37.46
37.67	44.81	57.61	60.99	46.26	46.33	47.12	38.01	38.51	38.88
70.70	73.84	76.72	70.34	76.07	47.09	54.95	48.78	51.70	59.55
EMG_KF_RFR_S2_R11_LL_VL	EMG_KF_RFR_S2_R12_LL_VL	EMG_KF_RFR_S2_R13_LL_VL	EMG_KF_RFR_S2_R14_LL_VL	EMG_KF_RFR_S2_R15_LL_VL	EMG_KF_RFR_S2_R16_LL_VL	EMG_KF_RFR_S2_R17_LL_VL	EMG_KF_RFR_S2_R18_LL_VL	EMG_KF_RFR_S2_R19_LL_VL	EMG_KF_RFR_S2_R20_LL_VL
47.07	38.17	37.61	37.61	45.25	31.86	31.86	30.44	34.65	28.54
57.91	55.43	60.83	62.23	52.28	58.01	54.26	54.44	55.44	48.63
57.67	64.12	66.50	73.98	81.60	67.12	63.52	60.16	56.59	62.12
41.15	47.11	48.20	49.08	52.46	44.32	37.03	31.48	34.98	35.72
80.41	74.62	78.45	74.45	64.23	74.01	61.61	65.11	68.34	68.34
47.83	49.99	47.11	52.82	55.13	40.77	51.17	50.40	44.93	46.38
58.83	50.86	50.04	57.77	43.23	43.78	43.78	40.88	40.88	40.88
38.13	58.25	56.24	58.29	50.84	70.19	50.28	38.07	46.80	42.46
24.53	75.70	73.13	58.22	66.22	61.22	56.61	64.60	65.17	55.17
66.38	69.33	69.08	66.52	63.04	42.73	47.77	45.33	47.19	46.10
37.09	35.03	33.67	40.61	39.17	39.44	29.69	29.52	29.52	30.67
49.99	59.44	64.16	55.53	64.87	55.60	61.50	53.15	48.10	47.10
82.80	75.30	77.50	88.29	86.57	77.63	78.13	74.60	74.60	74.60
51.47	60.31	60.31	62.45	62.45	62.45	62.45	61.17	61.17	61.17
68.70	70.39	67.28	69.04	71.17	37.77	44.28	60.97	60.97	47.52
EMG_KF_RFR_S2_R11_RL_VL	EMG_KF_RFR_S2_R12_RL_VL	EMG_KF_RFR_S2_R13_RL_VL	EMG_KF_RFR_S2_R14_RL_VL	EMG_KF_RFR_S2_R15_RL_VL	EMG_KF_RFR_S2_R16_RL_VL	EMG_KF_RFR_S2_R17_RL_VL	EMG_KF_RFR_S2_R18_RL_VL	EMG_KF_RFR_S2_R19_RL_VL	EMG_KF_RFR_S2_R20_RL_VL
50.17	72.46	78.16	81.04	81.04	81.04	81.04	81.04	76.81	76.81
48.83	51.17	30.05	33.74	48.93	38.13	44.18	47.05	49.47	38.29
54.71	55.13	54.25	58.37	68.87	54.77	53.55	54.87	44.87	51.34
45.70	45.18	50.92	44.89	54.85	47.25	39.32	43.79	40.80	43.84
51.61	52.81	51.20	49.60	48.26	41.20	47.54	44.27	46.29	46.29
52.00	50.42	56.18	64.30	41.31	46.95	47.63	48.06	48.06	48.06
37.72	46.34	50.26	45.03	49.53	44.47	49.35	45.58	43.10	38.66
23.40	30.45	30.45	31.25	31.25	31.25	31.25	31.25	27.89	27.89
71.80	69.50	68.79	71.96	60.94	58.33	58.33	60.54	56.20	56.20
61.83	67.52	58.84	56.98	64.26	39.03	40.74	48.61	42.10	43.76
20.96	29.20	27.77	37.58	38.30	28.94	28.94	26.76	28.84	20.40
45.79	51.89	51.89	53.77	60.08	58.78	53.40	46.18	41.88	41.88
47.50	46.01	41.82	53.52	53.30	27.73	33.87	44.74	44.30	43.22
40.16	51.93	50.07	54.92	39.43	41.50	43.84	41.88	39.50	31.96
58.77	58.65	54.67	58.86	59.83	33.47	42.77	44.66	52.83	48.21

AZ	BA	BB	BC	BD	BE	BF	BG	BH	BI
EMG_KF_RFR_S3_R1_UL_VM	EMG_KF_RFR_S3_R2_UL_VM	EMG_KF_RFR_S3_R3_UL_VM	EMG_KF_RFR_S3_R4_UL_VM	EMG_KF_RFR_S3_R5_UL_VM	EMG_KF_RFR_S3_R6_UL_VM	EMG_KF_RFR_S3_R7_UL_VM	EMG_KF_RFR_S3_R8_UL_VM	EMG_KF_RFR_S3_R9_UL_VM	EMG_KF_RFR_S3_R10_UL_VM
51.51	28.51	37.34	37.34	34.66	34.66	34.66	37.28	30.24	40.01
89.90	42.09	35.20	39.28	43.68	40.37	37.27	34.21	30.79	39.45
63.61	63.32	58.74	72.12	63.55	57.47	66.25	72.95	81.72	79.69
55.25	63.73	65.29	61.22	61.22	61.22	61.22	61.63	61.63	70.32
37.85	39.47	43.98	44.41	44.17	39.94	45.86	47.71	44.22	44.22
58.59	61.74	56.49	58.61	62.39	69.81	61.64	68.95	83.67	78.60
68.66	69.75	68.78	67.92	66.75	69.91	83.67	66.57	74.94	86.26
55.88	54.32	51.64	54.12	54.65	53.17	53.17	53.17	53.48	50.79
58.84	61.10	61.72	67.41	66.61	67.86	72.79	72.00	70.94	67.12
52.46	51.98	57.43	61.41	61.23	61.43	68.07	67.17	73.27	71.01
41.83	39.88	37.58	37.22	37.22	37.22	37.22	37.22	45.80	44.49
46.72	53.19	49.46	51.44	49.09	56.83	53.63	49.51	59.66	51.75
39.56	40.36	48.34	46.53	41.16	50.01	51.57	43.55	51.92	45.05
43.69	44.21	43.10	45.87	49.15	59.11	59.13	46.93	51.78	53.78
48.37	46.02	44.07	43.17	44.07	44.07	44.07	44.07	60.77	54.86
EMG_KF_RFR_S3_R1_LL_VL	EMG_KF_RFR_S3_R2_LL_VL	EMG_KF_RFR_S3_R3_LL_VL	EMG_KF_RFR_S3_R4_LL_VL	EMG_KF_RFR_S3_R5_LL_VL	EMG_KF_RFR_S3_R6_LL_VL	EMG_KF_RFR_S3_R7_LL_VL	EMG_KF_RFR_S3_R8_LL_VL	EMG_KF_RFR_S3_R9_LL_VL	EMG_KF_RFR_S3_R10_LL_VL
65.14	58.28	74.82	66.97	60.57	44.54	64.49	73.06	67.56	61.34
43.30	41.98	41.98	41.98	41.98	41.98	41.98	41.98	41.98	41.98
71.60	74.78	74.78	81.41	64.05	73.13	74.63	86.76	96.79	86.79
65.08	61.71	79.75	62.88	61.88	66.10	75.26	72.45	67.54	76.10
69.72	60.07	69.40	63.54	65.33	66.86	64.22	64.98	76.22	60.90
79.13	86.08	78.32	78.48	89.09	92.06	91.64	90.45	115.48	105.47
56.66	56.38	58.50	58.73	59.31	58.12	60.66	62.79	66.61	64.92
67.83	53.09	51.49	52.31	51.03	56.98	52.29	56.22	54.47	50.52
65.84	72.08	76.26	68.09	71.46	72.34	67.80	65.15	70.84	72.83
57.19	61.82	66.35	60.23	69.30	61.32	70.14	75.13	75.29	72.87
29.37	27.18	28.50	23.19	33.45	24.98	32.32	28.99	31.43	28.07
46.16	46.27	51.20	49.30	55.97	49.59	54.86	51.64	61.38	55.83
35.39	37.80	40.49	40.82	40.82	40.82	39.93	41.81	44.85	51.72
38.51	40.18	41.48	41.15	45.17	48.85	47.92	42.26	53.02	53.02
58.07	58.80	59.00	64.01	70.05	65.51	72.42	71.95	71.88	61.70
EMG_KF_RFR_S3_R1_LL_VL	EMG_KF_RFR_S3_R2_LL_VL	EMG_KF_RFR_S3_R3_LL_VL	EMG_KF_RFR_S3_R4_LL_VL	EMG_KF_RFR_S3_R5_LL_VL	EMG_KF_RFR_S3_R6_LL_VL	EMG_KF_RFR_S3_R7_LL_VL	EMG_KF_RFR_S3_R8_LL_VL	EMG_KF_RFR_S3_R9_LL_VL	EMG_KF_RFR_S3_R10_LL_VL
33.31	32.74	48.58	41.20	44.86	44.86	44.15	56.69	45.24	45.24
56.92	56.27	61.71	60.91						

CD	CE	CF	CG	CH	CI	CJ	CK	CL	CM
80.82	88.73	99.85	104.30	93.65	56.73	73.46	74.67	74.53	80.17
81.37	105.09	98.11	116.73	105.06	104.95	65.55	82.37	82.37	82.37
92.87	85.37	83.31	93.67	90.40	73.43	76.02	78.91	76.90	89.98
123.92	140.10	150.15	140.89	151.79	135.78	123.26	127.05	145.39	141.58
84.92	93.34	116.97	154.41	145.28	73.78	76.58	88.61	76.40	69.87
113.23	121.81	136.97	135.19	124.03	119.77	80.50	94.15	107.43	123.18
101.86	100.28	103.78	108.48	108.23	81.37	91.77	92.14	82.90	100.32
82.25	93.89	101.30	89.04	72.62	68.12	68.12	79.05	85.88	95.61
117.80	116.61	110.49	102.99	110.05	69.67	99.90	104.87	101.98	102.01
120.47	114.97	116.97	115.19	124.03	119.77	67.54	109.87	105.87	120.61
94.02	104.41	103.90	103.90	111.43	66.98	86.42	83.23	79.56	104.76
75.51	77.55	82.84	82.84	103.28	76.15	67.71	73.04	72.31	76.94
99.80	114.11	110.55	116.02	122.23	108.00	108.99	99.40	113.28	130.34
87.77	91.98	89.98	86.84	86.10	67.44	107.21	86.70	94.35	106.85
100.51	129.38	137.39	117.42	126.13	81.50	92.91	100.34	98.69	123.21
EMG_RE_H1_S1_R1_U1_V1	EMG_RE_H1_S1_R7_U1_V1	EMG_RE_H1_S1_R8_U1_V1	EMG_RE_H1_S1_R9_U1_V1	EMG_RE_H1_S1_R10_U1_V1	EMG_RE_H1_S2_R1_U1_V1	EMG_RE_H1_S2_R2_U1_V1	EMG_RE_H1_S2_R3_U1_V1	EMG_RE_H1_S2_R4_U1_V1	EMG_RE_H1_S2_R5_U1_V1
112.57	132.92	157.03	123.20	139.08	75.93	91.87	117.26	106.43	128.13
86.74	106.63	103.50	110.26	103.82	99.64	91.84	99.01		
82.90	78.28	85.11	83.98	83.98	66.00	74.40	63.22	76.19	89.40
123.08	136.12	136.56	124.78	130.17	122.71	116.61	109.23	128.81	126.40
82.43	91.34				69.76	81.86	82.06	71.91	76.06
123.00	124.21	139.45	153.30	148.60	95.74	90.14	92.82	108.79	133.54
109.01	87.80	101.48	110.34	86.92	79.20	79.36	86.88	82.35	95.86
100.11	106.55	113.76	115.53	120.72	76.70	80.90	96.87	101.16	101.27
148.01	145.44	140.69	123.94	126.25	121.09	108.98	121.82	142.88	132.41
114.56	97.67	116.74	102.55	114.50	84.84	97.83	103.75	114.33	105.14
80.99	88.27	98.27	98.86	47.33	62.42	68.80	74.33	74.33	78.57
80.96	73.65	76.59	87.72	84.91	58.76	51.15	59.24	61.23	77.17
144.54	123.06	126.28	115.87	120.78	79.83	80.58	108.33	104.14	115.86
87.18	104.03	113.50	117.37	116.55	90.68	105.74	98.12	104.82	105.67
134.11	142.08	137.71	127.01	150.50	83.68	97.61	134.42	119.78	123.55
EMG_RE_H1_S1_R6_U1_V1	EMG_RE_H1_S1_R7_U1_V1	EMG_RE_H1_S1_R8_U1_V1	EMG_RE_H1_S1_R9_U1_V1	EMG_RE_H1_S1_R10_U1_V1	EMG_RE_H1_S2_R1_U1_V1	EMG_RE_H1_S2_R2_U1_V1	EMG_RE_H1_S2_R3_U1_V1	EMG_RE_H1_S2_R4_U1_V1	EMG_RE_H1_S2_R5_U1_V1
81.16	91.64	91.47	93.61	84.81	65.59	64.41	64.41	71.38	74.37
123.37	125.38	118.87	127.24	129.97	112.88	117.11	102.83		
103.25	117.92	120.05	113.54	103.22	80.47	107.73	90.93	102.47	105.79
121.28	128.86	141.07	139.24	124.92	119.31	101.71	110.85	110.17	116.63
109.49	105.72				73.38	81.99	82.30	90.98	97.81
88.03	88.61	96.56	96.89	97.26	73.51	73.89	77.99	95.43	97.93
76.38	63.00	67.39	77.79	78.58	69.84	67.25	72.33	58.78	68.44
76.52	69.21	89.73	86.45	95.84	70.22	81.17	90.78	75.64	91.20
110.62	120.30	108.84	115.47	110.24	75.90	96.36	86.01	94.76	113.49
115.87	127.43	123.11	133.99	145.71	91.21	112.54	130.52	133.61	136.13
83.51	95.58	98.68	93.69	99.14	57.52	70.41	82.55	65.51	84.00
90.34	86.86	97.11	87.47	110.88	84.74	91.27	78.98	86.56	96.76
132.42	130.09	125.48	134.54	126.42	126.42	126.42	143.65	138.30	134.76
86.65	97.81	96.11	103.92	118.83	92.58	99.49	93.46	93.46	105.74
140.79	145.73	137.82	129.28	147.76	74.83	100.59	102.12	110.98	117.01
EMG_RE_H1_S1_R1_U1_V1	EMG_RE_H1_S1_R2_U1_V1	EMG_RE_H1_S1_R3_U1_V1	EMG_RE_H1_S1_R4_U1_V1	EMG_RE_H1_S1_R5_U1_V1	EMG_RE_H1_S2_R1_U1_V1	EMG_RE_H1_S2_R2_U1_V1	EMG_RE_H1_S2_R3_U1_V1	EMG_RE_H1_S2_R4_U1_V1	EMG_RE_H1_S2_R5_U1_V1
129.38	114.72	114.72	114.72	114.72	93.18	114.72	93.18	114.72	93.18
98.51	90.42	94.38	84.08	90.31	74.52	84.38	75.42	85.82	90.98
115.75	114.65	112.13	122.06	130.40	115.95	109.86	109.86	110.73	113.73
91.65	94.35				70.31	78.27	70.42	81.69	80.21
128.56	135.61	124.28	144.68	134.43	90.41	88.67	84.52	109.44	122.66
90.49	77.28	80.16	98.26	98.49	69.26	67.78	78.98	77.22	76.91
75.34	80.94	75.60	75.46	75.46	68.07	72.31	72.18	72.18	72.18
150.04	133.82	127.60	120.23	121.98	77.12	103.38	103.38	113.48	110.38
107.92	109.41	103.93	103.93	121.70	82.23	99.00	102.07	114.32	112.29
80.57	87.03	95.99	85.68	91.33	50.33	64.80	75.42	65.42	84.20
88.44	93.87	83.38	82.69	82.69	78.69	78.54	78.54	79.53	82.89
146.62	135.93	138.78	151.77	140.68	105.54	112.70	131.30	142.71	153.33
95.78	100.82	101.23	120.54	128.50	69.46	85.11	81.93	98.16	84.37
136.68	137.85	142.05	136.23	142.20	82.68	96.70	107.75	120.55	122.10
ON	CO	CP	CQ	CR	CS	CT	CU	CV	CW
EMG_RE_H1_S1_R6_U1_V1	EMG_RE_H1_S1_R7_U1_V1	EMG_RE_H1_S1_R8_U1_V1	EMG_RE_H1_S1_R9_U1_V1	EMG_RE_H1_S1_R10_U1_V1	EMG_RE_H1_S2_R1_U1_V1	EMG_RE_H1_S2_R2_U1_V1	EMG_RE_H1_S2_R3_U1_V1	EMG_RE_H1_S2_R4_U1_V1	EMG_RE_H1_S2_R5_U1_V1
92.22	93.00	101.61	102.72	92.79	79.42	86.41	93.11	80.74	85.11
81.71	87.20	82.18	97.36	81.67	84.90	95.76	88.72	99.24	95.54
141.37	164.96	161.70	153.19	183.03	143.89	132.66	159.60	157.66	172.12
81.83	84.05	74.90			85.62	70.07	85.96	76.04	87.81
128.28	131.43	153.71	155.68	168.47	108.33	125.09	120.46	112.27	128.56
103.80	101.84	106.81	107.94	115.98	81.58	85.33	91.98	85.33	91.98
103.72	102.36	104.05	116.02	94.38	66.01	64.15	73.09	74.18	90.36
113.79	116.22	117.36	112.78	112.78	81.18	78.95	82.56	83.17	94.95
124.69	126.82	131.17	143.66	131.09	92.68	115.48	119.82	147.80	144.12
106.78	104.23	104.23	111.21	106.79	106.79	107.86	107.86	107.86	110.04
76.67	99.88	119.57	83.11	90.45	69.40	72.57	70.15	86.45	70.40
112.84	134.65	148.28	148.37	152.94	113.20	103.38	110.96	122.01	133.51
102.01	94.46	106.13	96.95	107.49	91.38	101.54	93.52	95.69	90.63
124.57	124.15	125.46	125.93	125.03	125.03	125.03	125.03	125.03	125.03
EMG_RE_H1_S2_R6_U1_V1	EMG_RE_H1_S2_R7_U1_V1	EMG_RE_H1_S2_R8_U1_V1	EMG_RE_H1_S2_R9_U1_V1	EMG_RE_H1_S2_R10_U1_V1	EMG_RE_H1_S3_R1_U1_V1	EMG_RE_H1_S3_R2_U1_V1	EMG_RE_H1_S3_R3_U1_V1	EMG_RE_H1_S3_R4_U1_V1	EMG_RE_H1_S3_R5_U1_V1
121.81	116.04	128.69	121.15	129.24	105.20	112.46	120.54	120.54	120.54
74.51	85.30	74.82	79.32	77.38	58.83	76.25	72.22	84.62	75.00
125.48	127.85	151.57	140.43	164.80	113.46	114.62	127.48	127.48	119.92
72.71	83.53	75.78			84.57	70.44	69.42	74.66	87.36
134.17	127.00	148.05	154.49	168.10	92.47	119.54	123.44	120.75	136.75
88.66	93.77	85.72	96.77	103.64	62.01	78.64	80.50	76.90	86.32
114.83	105.96	121.18	122.27	113.45	71.76	73.91	87.97	93.98	110.92
116.55	128.74	60.80	121.00	116.79	62.50	86.72	96.68	104.27	118.71
111.58	119.71	118.41	117.37	112.68	84.69	85.98	104.29	104.53	109.29
90.16	84.75	80.81	84.77	91.56	67.51	83.97	90.44	87.04	97.50
78.54	86.89	92.86	88.54	81.51	68.51	69.65	65.13	65.13	77.79
108.38	132.37	151.28	120.00	140.02	97.07	96.33	110.48	105.10	128.56
99.45	102.11	131.26	117.23	138.58	91.52	91.52	94.18	95.70	86.78
145.18	145.21	161.06	144.77	178.12	94.37	101.84	101.85	122.28	136.77
EMG_RE_H1_S3_R6_U1_V1	EMG_RE_H1_S3_R7_U1_V1	EMG_RE_H1_S3_R8_U1_V1	EMG_RE_H1_S3_R9_U1_V1	EMG_RE_H1_S3_R10_U1_V1	EMG_RE_H1_S4_R1_U1_V1	EMG_RE_H1_S4_R2_U1_V1	EMG_RE_H1_S4_R3_U1_V1	EMG_RE_H1_S4_R4_U1_V1	EMG_RE_H1_S4_R5_U1_V1
93.64	86.52	92.38	93.00	90.66	66.27	80.66	80.66	80.66	80.66
113.77	113.24	113.02	122.58	113.91	96.29	107.69	105.69	122.58	122.58
146.44	149.26	150.67	180.15	191.21	117.08	123.93	144.98	144.64	140.09
98.27	99.64	75.34			88.69	80.08	91.82	75.69	75.69
98.46									

CX	CY	CZ	DA	DB	DC	DD	DE	DF	DG
EMG_KE_HI_S1_R6_IL_VLM	EMG_KE_HI_S1_R7_IL_VLM	EMG_KE_HI_S1_R8_IL_VLM	EMG_KE_HI_S1_R9_IL_VLM	EMG_KE_HI_S1_R10_IL_VLM	EMG_KE_HI_S1_R11_IL_VLM	EMG_KE_HI_S1_R12_IL_VLM	EMG_KE_HI_S1_R13_IL_VLM	EMG_KE_HI_S1_R14_IL_VLM	EMG_KE_HI_S1_R15_IL_VLM
102.26	102.88	104.88	86.72	92.32	57.51	86.48	96.35	92.78	88.02
86.72	83.68	107.99	123.29	103.05	104.93	91.62	100.69	88.48	103.28
170.75	172.01	170.17	165.81	204.06	175.45	142.46	160.90	156.31	172.96
65.63	86.77	73.04	91.79	81.94	79.52	75.66	79.52	66.85	84.49
133.94	142.90	147.90			85.53	101.62	81.78	129.46	126.98
87.83					71.96	82.31			
98.54	80.71	88.92	87.43	92.10	73.98	69.23	81.45	86.38	83.98
97.38	98.84	106.46	97.73	102.95	75.14	79.91	83.05	83.84	96.45
144.68	143.81	135.91	137.94	138.53	102.36	117.50	132.60	137.78	130.91
104.92	108.75	112.18			81.28	93.89	88.86	96.07	85.85
79.07	88.97	83.99	85.42	105.49	75.91	77.45	69.15	73.55	75.90
127.03	146.61	155.07	185.34	149.25	127.73	131.96	146.18	176.41	164.35
93.44	94.65	102.40	88.28	98.46	103.78	88.17	110.66	96.56	89.50
96.49	96.49	132.47	127.86	115.70	91.91	77.07	114.90	100.49	107.82
EMG_KE_HI_S1_R6_IL_VL	EMG_KE_HI_S1_R7_IL_VL	EMG_KE_HI_S1_R8_IL_VL	EMG_KE_HI_S1_R9_IL_VL	EMG_KE_HI_S1_R10_IL_VL	EMG_KE_HI_S1_R11_IL_VL	EMG_KE_HI_S1_R12_IL_VL	EMG_KE_HI_S1_R13_IL_VL	EMG_KE_HI_S1_R14_IL_VL	EMG_KE_HI_S1_R15_IL_VL
135.28	130.97	126.81	129.65	125.05	75.54	107.61	121.34	119.70	111.66
					91.09	92.58	84.64	102.23	105.89
77.48	79.11	81.01	100.16	94.48	78.56	74.64	76.34	75.27	79.18
64.71	85.67	77.37	65.61	83.99	88.05	85.31	85.11	59.16	76.48
143.82	142.92	150.90			85.53	102.86	92.23	102.86	145.70
73.69					87.64	80.24	70.58		
109.74	102.25	106.16	119.71	102.07	77.75	83.75	90.19	103.34	105.34
139.40	116.97	123.15	117.54	125.71	72.13	103.40	122.33	109.35	139.29
111.32	127.04	104.06	110.20	110.20	93.55	100.96	92.75	112.38	112.38
95.19	96.57	88.11			61.31	77.97	83.80	68.87	69.84
70.78	86.77	86.77	94.66	94.23	62.57	72.20	70.99	63.50	80.54
135.83	131.80	134.98	165.00	139.41	89.27	117.56	143.46	133.80	144.90
97.96	91.60	102.05	94.21	110.07	110.33	92.10	123.63	110.97	113.84
108.27	132.24	134.13	135.50	154.32	108.75	108.75	134.54	134.54	123.41
EMG_KE_HI_S1_R6_IL_VL	EMG_KE_HI_S1_R7_IL_VL	EMG_KE_HI_S1_R8_IL_VL	EMG_KE_HI_S1_R9_IL_VL	EMG_KE_HI_S1_R10_IL_VL	EMG_KE_HI_S1_R11_IL_VL	EMG_KE_HI_S1_R12_IL_VL	EMG_KE_HI_S1_R13_IL_VL	EMG_KE_HI_S1_R14_IL_VL	EMG_KE_HI_S1_R15_IL_VL
85.01	88.63	81.69	97.88	103.76	65.54	83.87	101.73	95.64	85.65
122.58	123.70	129.29	154.83	122.78	113.94	119.01	101.28	127.98	115.38
177.52	177.97	179.58	194.96	198.91	175.30	154.61	146.55	182.69	181.78
58.65	86.62	86.62	96.53	100.06	92.67	89.85	85.11	79.39	89.39
88.60	105.80	114.26			78.89	72.37	71.48	94.24	90.31
62.25					58.13	51.88	54.90		
94.42	94.06	78.51	102.27	94.73	69.63	78.71	91.91	81.30	85.17
102.62	102.36	102.36	115.76	113.83	70.32	96.54	90.27	91.69	94.93
142.91	140.93	151.41	153.44	162.59	113.15	140.46	135.60	151.22	126.09
101.30	106.74	99.87			74.91	79.35	79.07	84.55	82.41
107.70	105.99	115.56	108.76	100.19	83.01	93.00	87.15	100.04	91.60
194.82	194.82	200.00			180.45	173.80	150.83	205.08	210.90
108.06	101.44	82.06	89.47	89.21	93.21	96.06	104.26	85.71	107.08
107.82	135.57	134.29	136.41	141.96	88.97	87.65	98.37	110.57	124.40
EMG_KE_HI_S1_R6_IL_VL	EMG_KE_HI_S1_R7_IL_VL	EMG_KE_HI_S1_R8_IL_VL	EMG_KE_HI_S1_R9_IL_VL	EMG_KE_HI_S1_R10_IL_VL	EMG_KE_HI_S1_R11_IL_VL	EMG_KE_HI_S1_R12_IL_VL	EMG_KE_HI_S1_R13_IL_VL	EMG_KE_HI_S1_R14_IL_VL	EMG_KE_HI_S1_R15_IL_VL
141.00	145.80	143.92	126.82	133.24	113.24	88.85	106.75	124.53	141.77
103.00	97.44	107.86	108.26	102.08	88.09	93.19	85.72	101.64	93.77
164.86	188.99	165.19	171.54	186.49	147.48	148.93	127.74	166.14	163.54
71.80	77.86	84.72	84.24	84.10	102.27	82.33	82.92	74.95	79.98
127.84	147.62	148.81							
74.02					65.83	64.20	65.12		111.95
89.16	78.25	71.42	93.80	87.97	61.76	60.78	76.04	66.70	75.56
105.01	99.08	131.08	111.45	120.21	69.73	110.62	102.24	91.46	107.67
110.08	113.86	118.19	120.83	128.79	71.88	82.87	90.45	90.83	86.74
78.85	81.45	76.49			57.89	65.87	68.11	75.33	67.83
78.31	91.06	82.18	89.70	94.34	81.04	73.99	69.00	82.12	78.27
170.64	186.15	198.29	193.83	184.54	108.10	115.97	159.76	196.99	186.24
96.72	95.86	99.17	101.54	113.22	95.80	82.23	111.47	94.58	117.64
135.95	147.42	135.72	138.74	161.29	82.67	97.29	99.21	115.21	130.72

DH	DI	DJ	DK	DL	DM	DN	DO	DP	DQ	DR
EMG_KE_HI_S4_R6_RL_VLM	EMG_KE_HI_S4_R7_RL_VLM	EMG_KE_HI_S4_R8_RL_VLM	EMG_KE_HI_S4_R9_RL_VLM	EMG_KE_HI_S4_R10_RL_VLM						
87.66	87.66	106.30	101.27	87.61						
101.88	101.88	90.81	102.00	108.91						
174.69	169.33	190.93	198.00	205.74						
80.25	79.71	80.84	75.32	82.26						
151.15	141.69	147.20	160.24	165.57						
99.54	91.77	90.01	85.38	93.90						
97.20	103.52	109.68	109.60	117.08						
118.63	146.06	150.54	141.46	145.58						
114.43	98.23	100.48	99.29	103.92						
78.20	86.75	95.00	98.48	103.33						
108.88	123.68	123.68	123.68	202.89						
93.47	93.90	93.83	110.53	86.65						
114.50	118.97	112.08	132.08	127.15						
EMG_KE_HI_S4_R6_RL_VL	EMG_KE_HI_S4_R7_RL_VL	EMG_KE_HI_S4_R8_RL_VL	EMG_KE_HI_S4_R9_RL_VL	EMG_KE_HI_S4_R10_RL_VL						
100.00	106.35	115.25	110.11	117.09						
78.43	88.05	85.15	86.10	96.54						
141.08	144.08	160.09	166.48	168.82						
88.70	85.83	87.89	78.05	76.96						
130.44	134.73	147.32	148.19	167.48						
114.03	98.14	106.22	109.91	110.08						
110.52	116.38	126.47	121.01	99.48						
103.99	108.01	105.35	117.44	84.53						
86.63	82.69	79.98	87.01	85.88						
80.63	79.75	92.15	89.89	95.13						
145.83	154.26	162.72	200.73	210.72						
117.88	102.63	127.19	99.69	114.27						
135.32	140.87	151.05	133.03	161.80						
EMG_KE_HI_S4_R6_RL_VL	EMG_KE_HI_S4_R7_RL_VL	EMG_KE_HI_S4_R8_RL_VL	EMG_KE_HI_S4_R9_RL_VL	EMG_KE_HI_S4_R10_RL_VL						
82.64	83.75	101.61	100.00	105.91						
121.37	113.25	136.49	132.58	127.12						
119.95	108.75	213.48	200.09	214.48						
95.46	90.28	86.82	193.83	103.09						
88.95	89.91	111.80	99.30	118.52						
88.51	82.04	86.15	86.61	107.99						
105.93	109.06	108.03	112.25	111.18						
151.79	151.02	152.40	160.54	159.54						
110.60	100.82	102.40	91.78	101.91						
86.77	88.36	117.20	118.22	111.53						
244.00	178.17	223.98	272.68	298.21						
107.29	99.89	100.86	91.87	86.54						
153.84	140.50	136.40	164.46	154.40						
EMG_KE_HI_S4_R6_RL_VL	EMG_KE_HI_S4_R7_RL_VL	EMG_KE_HI_S4_R8_RL_VL	EMG_KE_HI_S4_R9_RL_VL	EMG_KE_HI_S4_R10_RL_VL						
130.51	136.84	123.53	118.70	135.65						
89.62	94.32	98.36	101.22	103.42						
181.07	187.88	198.09	222.31	189.80						
85.55	91.03	89.78	89.66	99.98						
127.48	135.23	148.41	154.78	135.20						
95.01</										