ACUTE EFFECTS OF HIGH-INTENSITY INTERVAL EXERCISE VS. CONCURRENT EXERCISE ON FLOW-MEDIATED DILATION IN COLLEGE-AGED WOMEN

A THESIS

SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS

FOR THE DEGREE OF MASTERS OF SCIENCE

IN THE GRADUATE SCHOOL OF THE

TEXAS WOMAN'S UNIVERSITY

SCHOOL OF HEALTH PROMOTION AND KINESIOLOGY

COLLEGE OF HEALTH SCIENCES

BY

ASHLEY ALLEN, B.S

DENTON, TEXAS

DECEMBER, 2019

DEDICATION

For my parents, Leslie and Leon, who told me I could do anything and never stopped pushing me. To my friends who patiently sat through the ups and downs to reach this point, without you all I wouldn't have made it, thank you.

ACKNOWLEDGEMENTS

I would like to express my gratitude to my project advisor Dr. Rigby for his patience and advice during the planning and development of this research project. I am also grateful to the research team, Matt Sokoloski, Nick Levine, and Chris Irvine, all of who gave me countless hours of their time either helping run the study or troubleshooting data. Finally, I would like to thank my family for being there for advice and late-night phone calls to help me get through school.

ABSTRACT

ASHLEY ALLEN

ACUTE EFFECTS OF HIGH INTENSITY INTERVAL EXERCISE VS
CONCURRENT EXERCISE ON FLOW MEDIATED DILATION
IN COLLEGE-AGED WOMEN

DECEMBER 2019

Heart disease is the leading cause of female mortality worldwide. As a treatment, exercise can improve cardiac and endothelial function, increase bone mineral density, promote lean muscle mass, and improve pulmonary function. The purpose of this study was to compare the effect of two different modes of exercise on flow mediated dilation in the brachial artery in college-aged women. Ten recreationally active women were assigned to either a high intensity interval group (HT) or a group who completed strength exercises and high-intensity interval exercise, called the concurrent exercise group (CT). Measurements of flow mediated dilation (i.e., the hyperemic response of a blood vessel post occlusion) were taken at baseline, after one workout session, and after three workout sessions. There were no differences for FMD% (flow-mediated dilation) or brachial artery diameter across all time points for either training protocol. Neither a combination of high-intensity interval exercise and resistance exercise nor high-intensity interval exercise alone acutely led to structural or functional changes in the brachial artery in recreationally active healthy young women.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Heart disease is the leading cause of female mortality worldwide, according to the World Health Organization. Metabolic diseases also contribute to a significant number of deaths. According to the Centers for Disease Control (CDC), over two-thirds of the U.S population is overweight or obese (CDC, 2017). Physical activity is a common method prescribed to prevent excessive weight gain and to promote weight loss. Physical fitness (e.g., muscular strength and aerobic exercise) can be a way to increase bone density, improve cardiac and endothelial function, promote lean muscle mass, and improve pulmonary function. Moderate-intensity exercise (i.e., 45 to 59% $\dot{V}O_2R$ for 150-300 min/week), or vigorous-intensity exercise (i.e., 60 to 89% $\dot{V}O_2R$ for 75 to 150 min/week) is the current recommendation by the American College of Sports Medicine for weight loss (American College of Sports Medicine, 2018; Donnelly et al., 2009). Therefore, exercise programs typically include 30 to 60 min of steady-state exercise at a moderate intensity on most days of the week.

Time commitment is often cited as a reason people do not exercise. Interval training may be the solution for populations seeking general health benefits, but who are not able to take time for long duration exercise bouts. High-intensity interval training (HIIT) is a form of interval training that alternates periods of high-intensity (i.e., ~80% $\dot{V}O_{2max}$ to supramaximal exertion) exercise with less intense rest periods (Laursen & Jenkins, 2002). High-intensity interval exercise appears to produce general health adaptations, including structural changes of peripheral arteries (Rakobowchuk et al., 2008), and improve sport performance in a variety of populations (Rakobowchuk et al., 2005; Sperlich et al., 2011). There is evidence that HIIT protocols also have similar, if not greater, improvements in aerobic and anaerobic performance ($\dot{V}$O2max, peak power output) versus steady-state exercise (Gibala et al., 2006; Gibala et al., 2012; Helgerud et al., 2007). Though there is not a set standard for intensity, time, or volume of HIIT, these adaptations have been observed across participants (Laursen & Jenkins, 2002).

Training for muscular strength concurrently with HIIT has only been recently introduced as a training option. The concurrent protocols include both split (specific muscle groups) and whole-body strength exercise routines mixed with a wide variety of sprint or cycle HIIT. Split strength routines are being utilized as a time effective way to achieve similar strength gains versus whole-body routines (Calder, Chilibeck, Webber, & Sale, 1994). Flow-mediated dilation is the hyperemic response of a blood vessel following occlusion, and it can be quantified as the amount of expansion of an artery's diameter (hyperemic response) after blood flow returns (Celermajer et. al., 1992). The peak brachial artery vasodilation (the largest diameter of an artery following return of blood flow) and the shear stress in response to occlusion is indicative of an individual's endothelial function (Pyke, Dwyer, & Tschakovsky, 2004; Pyke, & Tschakovsky, 2005). The FMD technique is utilized primarily in populations with diagnosed heart disease (Meyer et al., 2005), or as a predictor of a cardiac event (Shechter, Schechter, Koren-Morag, Feinberg, & Hiersch, 2014), because it is a non-invasive way to assess endothelial function that correlates to the distensibility or stiffness of central vasculature.

FMD is a significant independent predictor of a cardiac event dependent of cardiovascular disease in people who do not currently have the disease. Participants who have FMD values below a certain cut point for FMD% consistently exhibit a greater risk for an adverse cardiac event (Gori et al., 2011; Schechter et al., 2009). Because there is a significant gender difference in FMD%, data recorded using the FMD technique in men might not apply to females (Patel et al., 2005). When considering the differences in FMD data in women during the menstrual phases, women's FMD data during their menses correlate most closely with men. Hormones such as estrogen, progesterone, and luteinizing hormone may be the reason women have greater artery dilation than men during all other phases of the menstrual cycle (Hashimoto et al., 1995).

Problem Statement

While brachial artery structure has been well characterized using the FMD technique in men, few studies exist that include the measurement in women. Even fewer studies exist that include the measurement following acute exercise in women. The purpose of this study was to compare the effect of two different modes of exercise on FMD in the brachial artery in recreationally active, college-aged women.

Hypotheses

H_o: There will be no difference in FMD response post exercise in the high-intensity interval exercise group as compared to pre-exercise FMD in recreationally active college-aged women.

H_o: There will be no difference in FMD response post exercise in the concurrent exercise group as compared to pre-exercise FMD in recreationally active college-aged women.

H_o: There will be no differences in FMD response post exercise between the high-intensity interval exercise group and concurrent exercise group in recreationally active college-aged women.

H_A: There will be an increase in FMD response post exercise in the high-intensity training group compared to pre-exercise FMD in recreationally active college-aged women.

H_A: There will be an increase in FMD response post exercise in the concurrent exercise group compared to pre-exercise FMD in recreationally active college-aged women.

H_A: There will be a greater increase in FMD response post exercise for the concurrent exercise group than the high-intensity interval exercise group in recreationally active college-aged women.

Assumptions

1. The participants will cooperate and provide reliable responses to health questionnaires for study inclusion.
2. The participants will follow the guidelines prior to testing.
3. The participants will exert themselves to reach maximal exertion during maximal tests.
4. The participants recruited will be representative of the population targeted.

Limitations

1. The study is limited to healthy female participants, so the findings may not translate to males.
2. The study was limited to one college.

Delimitations

1. Female participants were chosen exclusively due to sex differences found in the literature.

Definitions

1. Cardiorespiratory fitness- the capacity of respiratory and cardiovascular systems to provide muscles with oxygen during sustained exercise
2. Endothelial function- the ability of the endothelium found in the heart and blood vessels to vasoconstrict and vasodilate
3. Flow-mediated dilation- the hyperemic response of a blood vessel following occlusion

4. High-intensity interval exercise- a form of interval exercise that alternates periods of high-intensity exercise with less intense rest periods
5. Hyperemic response- measurement of an artery vasodilation

Significance of Study

There are few studies that include the effect of a HIIT protocol on healthy and clinical populations. Concurrent protocols of HIIT and resistance exercise have been investigated almost exclusively in competitive athletic populations. There is little literature on HIIT and concurrent protocols and the effect on FMD in healthy female populations. Female-specific FMD research needs to be implemented due to the high prevalence of heart disease in women. Therefore, there is a need to find exercise modalities that could mitigate factors that possibly contribute to a future cardiac event. The purpose of this study was to compare the effect of two different modes of exercise on FMD in the brachial artery in recreationally active, college-aged women. The exercise protocols could have an impact on how exercise is prescribed as a method for disease prevention for women.

CHAPTER II

LITERATURE REVIEW

High-Intensity Interval Training

Physical activity is an important part of maintaining health and well-being. Cardiorespiratory fitness is especially important as a leading cause for all-cause mortality is heart disease. HIIT is becoming a popular way to achieve similar physiological adaptations as long distance aerobic exercise. Helgerud et al. (2007) conducted a study that investigated the benefits of HIIT on maximal oxygen consumption ($\dot{V}O_{2max}$) in comparison to continuous, moderate-intensity aerobic training. In this study, healthy, non-smoking, moderately-trained males were enrolled in an 8-week training program. Only 40 participants completed 90% of the training sessions to be included in the results, and four people from each group were not able to perform the tests measuring cardiac output, stroke volume, and $\dot{V}O_{2max}$. Only six participants from each group were reflected in those results. Groups consisted of long slow distance running (LSD), lactate threshold running (LT), 15/15 interval running (15/15), and 4 x 4-min interval running (4 x 4-min). All training groups completed the training three days a week. The LSD group performed a continuous run at 70% HR_{max} for 45 min. The LT group performed a continuous run at the lactate threshold (85% of HR_{max}) for 24.25 min. The 15/15 interval group performed 47 repetitions of 15 s intervals at 90-95% HR_{max} with 15 s of active rest (70% HR_{max}). The 4 x 4 min interval group completed four repetitions of 4 min of training at 90-95% HR_{max} with 3 min active rest at 70% HR_{max} between each interval. The authors found that the groups utilizing HIIT had significant increases in $\dot{V}O_{2max}$ when compared to the LSD running group and the LT running group. Percentage increases for the 15/15 and 4 x 4-min group were 5.5 and 7.2%, reflecting $\dot{V}O_{2max}$ increases from 60.5 to 64.4 $ml \cdot kg^{-1} \cdot min^{-1}$. Stroke volume increased by approximately 10% after interval training. The authors of the study concluded that HIIT is significantly more effective than continuous, moderate-intensity aerobic training at 70% HR_{max}, or at the lactate threshold, in improving $\dot{V}O_{2max}$ (Helgerud et al., 2007).

Farzad et al. (2011) conducted a study that investigated the physiological and performance changes from the addition of a sprint interval training program to wrestling. In this study, 14 trained male freestyle wrestlers with 6 to 7 years of wrestling training experience were randomly assigned to one of two groups. The first group was the experimental group (EXP), and the second was a control group (CON). Both groups underwent a traditional preparation phase that consisted of learning a drilling technique, live wrestling, and weight training for four weeks. The EXP group performed a running-based sprint interval training (SIT) protocol in addition to the traditional preparation. The SIT protocol consisted of six, 35-m sprints at maximum effort with 10 s recovery between bouts, performed twice a week for four weeks. Pre-and post-testing for the groups consisted of a graded exercise treadmill test and four successive Wingate tests. The authors found the EXP group had significant improvements in $\dot{V}O_{2max}$ (+5.4%), peak oxygen pulse (+7.7%) and time to exhaustion (+32.2%) compared with pretesting. The EXP group produced significant increases in peak power output and mean power output during the Wingate testing compared with pretesting. After the 4-week training program, total testosterone and the total testosterone-cortisol ratio increased significantly in the EXP group. The authors concluded that the addition of a SIT program with short recovery can improve both aerobic and anaerobic performance in wrestlers, and the training induced anabolic adaptations (Farzad et al., 2011).

HIIT for health was investigated by Gibala et al. (2012). In this review, physiological adaptations to low-volume HIIT with regard to health and disease were investigated. Some of the possible mechanisms that cause improved skeletal muscle function, metabolic control, and changes in cardiorespiratory function in response to low-volume HIIT were also investigated. Low-volume HIIT (using Wingate tests) elicits physiological adaptations in skeletal muscle and the cardiorespiratory system similar to that of endurance trained participants. Wingate tests are very demanding cycle tests that might not be tolerated by a wide range of participants. With a decreased absolute intensity and increased duration (20 min), skeletal muscle and cardiorespiratory training adaptations were comparable to endurance training and other low-volume HIIT protocols. Low-volume HIIT may therefore be an effective way to potentially mitigate factors that could cause cardiovascular disease. Gibala et al. (2012) concluded that there was considerable evidence to support low-volume HIIT as a time-effective training method for causing improvements in skeletal muscle and the cardiorespiratory system. There is limited research on this training protocol in populations with, or at risk for, cardiometabolic disorders. The majority of the potential benefits of low-volume HIIT are unclear as the majority of low-volume HIIT protocols in diseased populations have only utilized relatively short intervention periods. Finally, the authors suggest future research involving long-term (i.e., months to years) interventions for a variety of clinical populations may be needed to better understand how exercise affects chronic cardiorespiratory and skeletal muscle responses in those populations (Gibala et al., 2012)

High-Intensity Interval Training and Resistance Training

Improvements in cardiorespiratory fitness can be obtained through aerobic exercise. However, cardiorespiratory fitness is not the only factor in overall fitness that leads to the prevention or mitigation of cardiovascular and metabolic diseases. Regimented strength training, used in combination with aerobic exercise, may elicit physiological adaptations that are greater in magnitude versus either regimen by itself. Botonis et al. (2015) investigated the effects of concurrent strength and interval endurance training in water polo players. Fourteen elite male water polo players were divided into two training groups (HIIT 4 x 4 min or HIIT 16 x 100 m), both of which incorporated strength training and HIIT. This study used water polo players, so the high-intensity interval bouts were swimming sprints. The strength training portion of the study incorporated bench-press, seated pull-down, triceps press, shoulder press, and leg press. These exercises were performed at 85 to 90% 1RM, for 4 to 5 repetitions for 4 sets. Results of maximal strength gains in both groups were presented, but significant increases in swimming speed were only found in the HIIT 4 x 4 group. The authors concluded that HIIT and strength training performed concurrently improved muscular strength and sport specific adaptations in elite water polo players (Botonis et al., 2015).

Laird et al. (2016) evaluated performance improvements after resistance training and sprint interval concurrent training. This study utilized HIIT in the form of treadmill sprints using a 2:1 work rest ratio (20:10 s) with 8 repetitions for a total of 4 min. Twenty-eight recreationally active women were divided into two groups: concurrent sprint interval (CST), and resistance training (RT). The participants trained 3 days per week for a total of 11 weeks. The resistance training prescription was the same for both groups. Resistance training was divided into protocols A & B. Protocol A consisted of back squats, bent over rows, bench press, and sit ups. Protocol B consisted of squat jumps, deadlift, standing press, and back extensions. The CST group completed sprint intervals at least 4 hr after the resistance training. Results for this study were significant increases in 1RM back squat (37.5 ± 7.8 vs. 40.0 ± 9.6 kg) and average peak anaerobic power (7.4 ± 6.2 vs. 7.6 ± 6.4%) for both the CST and RT groups. Significant improvements in $\dot{V}O_{2max}$ (35.8 ± 4.2 to 38.8 ± 4.0 ml/kg/min) were observed only in the CST group. The authors concluded that 11 weeks of resistance and concurrent training resulted in similar physiological improvements, when compared to groups completing aerobic or resistance training alone (Laird et al., 2016).

Flow-Mediated Dilation and High-Intensity Interval Training

FMD is a non-invasive way to monitor endothelial function. The ability of an artery to vasodilate and vasoconstrict is an indicator of vascular health. Arteries that do not vasodilate with a given stimulus could indicate stiffening and disease, and this could be in the central vasculature (Rakobowchuk et al., 2005). HIIT has been used to help treat symptoms in populations with a diagnosed cardiovascular disease (Warburton et al., 2005). Healthy populations have also adapted the use of HIIT to influence endothelial function. Rakobowchuk et al. (2008) investigated the effects of sprint interval and traditional endurance training on peripheral artery stiffness and FMD in healthy humans. Twenty healthy men and women were recruited and split into two groups: sprint interval training (SIT; $n = 10$) and endurance training (ET; $n = 10$). Each group had 5 men and 5 women, and underwent 6 weeks of training. The SIT group completed 4 to 6 all out Wingate tests, lasting 30 s each and separated by 4.5 min recovery, 3 days a week. The ET group completed 40 to 60 min of cycling at 65% peak oxygen uptake 5 days a week. Popliteal endothelial function improved after training in both groups. Carotid artery distensibility was not altered by the training in either group. Popliteal artery distensibility was improved in both groups by the same amount. The authors concluded that SIT is a time-efficient way to elicit improvements in peripheral vascular structure and function comparable to ET, but alterations in central vasculature may require a longer training stimulus (Rakobowchuk et al., 2008).

Rakobowchuk et al. (2005) investigated FMD following whole body resistance training. Twenty-eight male participants took part in a 12-week training program that included whole body resistance training, 5 days a week, using a 3-day split cycle that repeated. The resistance training was divided into upper body push and pull, and lower body leg days. “Push” consisted of the following exercises: shoulder press, horizontal bench press, vertical bench press, triceps push down, and chest flys. “Pull” consisted of the following exercises: seated lat pull downs, wide-grip seated row, narrow-grip seated row, biceps curl, and seated rear flys. Leg day consisted of the following exercises: incline double leg press, leg curl, seated leg extension, and standing calf raises. Abdominal and lower back extension exercises were included every 3 days. At the end of

12 weeks, the participants had significant increases in strength, with increases in aggregate pulling (40 ± 0.02%), pushing (50 ± 0.02%), and leg (61 ± 0.02%) loads. Resting mean brachial artery diameter significantly increased from pre- to mid-testing and remained elevated at post-testing. Peak forearm blood flow significantly increased above pre-levels by mid-testing and remained elevated at post-testing. Peak and average shear rate in the brachial artery was not significantly different, nor was the relative FMD of the brachial artery. The authors concluded that peripheral artery remodeling does occur with resistance training in healthy men, and the increase in post occlusion could indicate improved vessel function (Rakobowchuk et al., 2005).

Sex Differences in Flow-Mediated Dilation

While FMD is a non-invasive way to assess endothelial function, there are noted differences on what could be considered normal function between the sexes. In some cases, the values differ so extremely that using the same specific "cut points" for men and women could seriously under-diagnose a potential issue in one sex (Patel et al., 2005). Patel et al. (2005) investigated gender-based differences as an indicator for coronary artery disease. This study included 209 outpatients (141 women and 68 men) who were referred for cardiovascular evaluation. Participants underwent myocardial perfusion stress testing to be classified as either having coronary artery disease (CAD) or being free of CAD. Participants with no evidence of ischemia or infarct were classified as being free of CAD, and participants were classified as having CAD if they had angiographic evidence of > 50% coronary stenosis, or any degree of fixed or reversible myocardial perfusion defects confirmed by angiography. Brachial FMD was performed on all participants with CAD (n = 64) and those who did not have CAD (n = 145). The FMD in women who had CAD (n = 33, 9.1 ± 0.8%) was higher than that of similarly aged males who had CAD (n = 31, 6.4 ± 0.5%). The authors of this study concluded that if the cut point as defined in men were used to evaluate women, brachial FMD would fail to diagnose 42% of women who did not have significant CAD. A higher FMD cut point is required to identify significant CAD in similarly aged women (Patel et al., 2005).

Hashimoto et al. (1995) also studied the sex differences with FMD. The authors of this study investigated the differences with FMD on the brachial artery by sex and menstrual cycle. Aged-matched volunteers (women n = 17, men n = 17) were examined using the non-invasive brachial artery FMD technique after sublingual nitroglycerin administration. The female participants were studied three times each, in the three different phases of one menstrual cycle (i.e., menses [M], follicular phase [FP], and luteal phase [LP]). Hashimoto et al. (1995) found that the FMD increase during M, when serum estradiol level was low (121.9 ± 12.5 pmol/L), was 11.22 ± 0.58%. This value was comparable to that in male participants (10.60 ± 0.75%). Percent FMD increased in FP (18.20 ± 0.81%) and LP (17.53 ± 0.74%) when compared to M. When serum estradiol level was high, FMD values were significantly larger in the FP (632.0 ± 74.5 pmol/L) and LP (533.8 ± 33.4 pmol/L) phases compared to the M phase. Endothelium-independent vasodilation by nitroglycerin increased in both the FP and LP. The authors concluded that endothelium-dependent vasodilation varies during the menstrual cycle, and that the endogenous estradiol may be involved in this menstrual cycle-related vasodilation (Hashimoto et al., 1995).

As the menstrual phase affects several physiological factors for women, there might be slight advantages or disadvantages in testing participants during certain phases. Janse de Jonge, Boot, Thom, Ruell, and Thompson (2001) investigated the influence of menstrual cycle phase on skeletal muscle contractile characteristics in women. In this study, women (n = 19) had strength, fatigability, and contractile properties of skeletal muscles measured during the different phases of the menstrual cycle. During each phase, participants underwent isometric quadriceps exercises, isokinetic extension/flexion exercises at different velocities, and dominant side hand grip exercises. No significant changes were found in these muscle function parameters across menstrual cycle phases, and the muscle function measurements had no significant correlations with any of the female reproductive hormones. The authors concluded that the fluctuations in female reproductive hormone concentrations throughout the menstrual cycle do not affect muscle contractile characteristics (Janse de Jonge et al., 2001).

Lebrun, McKenzie, Prior, & Tauton (1995) studied the effects of menstrual cycle phase on four variables of athletic performance: aerobic capacity, anaerobic capacity, isokinetic strength, and high intensity endurance. In this study, 16 women between the ages of 18 to 40 years, with a history of regular menstruation, had not taken contraceptives in the past six months, and who exercised regularly at a high intensity, were recruited. Women were tested during the early follicular (between Days 3 and 8) and midluteal phase (between Days 4 and 9 after ovulation) of the menstrual cycle confirmed by serum estradiol and progesterone assays (these times are during and after menstruation). The participants underwent physiological testing on two successive days, which was identical for each test period. Aerobic capacity was assessed using a ramp treadmill protocol to achieve $\dot{V}O_{2max}$. Anaerobic performance was assessed using a sprint test on a treadmill at 8 mph on a 20% incline until the participants were unable to continue. The second day of testing consisted of endurance performance and isokinetic strength testing. Endurance training consisted of running at 90% of $\dot{V}O_{2max}$ to fatigue. Isokinetic strength was assessed by peak torque generated by knee flexion. Lebrun et al. (1995) found no significant differences in aerobic capacity, anaerobic performance, high intensity endurance, and isokinetic strength between the follicular and luteal phases. Both absolute and relative $\dot{V}O_{2max}$ were slightly lower in the luteal when compared to the follicular group (3.19 ± 0.09 L/min vs. 3.13 ± 0.08 L/min; 53.7 ± 0.9 ml/kg/min vs. 52.8 ± 0.8 ml/kg/min). The authors concluded that cycle phase did not significantly impact the physiological variables measured in this study, but the cyclic increases in endogenous female steroid hormones of the menstrual cycle may have a slight, deleterious influence on aerobic capacity, with potential implications for individual athletes (Lebrun et al., 1995).

Flow-Mediated Dilation and Contraceptive Use

Differences in FMD% between the menstrual cycle phases have been noted above, but it should also be noted that a women's contraceptive use can also greatly alter endothelium function. Heidarzadeh et al. (2014) investigated the differences in FMD and carotid artery-intima thickness between a control group (i.e., non-hormonal contraceptives) and a group of women using combined oral contraceptives. A non-significant, inverse relationship between duration of combined oral contraceptive use and

FMD% was recorded, along with a non-significant positive relationship with carotid artery-intima thickness. The authors concluded that prolonged use of combined oral contraceptives can cause changes in endothelial structure and function (Heidarzadeh et al., 2014).

Franceschini et al. (2013) also investigated the combined oral contraceptive effect on FMD and carotid artery intima-media thickness. The authors in this study incorporated contraceptives containing levonorgestrel or chlormadinone exclusively. Sixty-four women, 21 in a nonhormonal control group (i.e., non-hormonal contraceptives), and 43 randomized to either chlormadinone (CMA) or levonorgestrel (LNG) were studied at baseline values and 6 months. The LNG group had nearly a threefold reduction of FMD compared to the CMA group, and a 7.5 greater reduction in FMD compared to the controls. The authors concluded that combined oral contraceptives containing LNG were associated with more changes in FMD and carotid artery intima-media thickness of healthy women than a combined contraceptive containing CMA and nonhormonal contraception (Franceschini et al., 2013).

There are clear positive physiological and performance adaptations from a combined aerobic and resistance training program in both healthy and clinical populations. Specifically, combined HIIT and resistance exercise can improve cardiorespiratory fitness, muscular strength, lean tissue mass, and FMD response. There are sex considerations when studying the endothelium function response to exercise. Women and men have significantly different endothelial responses to the same hyperemic stimulus (e.g., cuff occlusion and release). It appears the only time women's endothelial function is comparable to male counterparts is in the menses phase of the menstrual cycle. In this study, the acute FMD response to a training stimulus in women will be investigated, to potentially build a platform for future research.

CHAPTER III

METHODS

Participants

Young women, aged 18 to 35 years, were recruited for this study (*n* = 15), but only 10 participants completed the study. Participants who were recreationally active (i.e., exercise 2 to 3 days/week of moderate intensity ([40 to 60% HRR]) or vigorous ([60 to 90% HRR]) physical activity for at least the previous year) with a body mass index (BMI) between 18.5 to 29.9 kg/m^2 were eligible. Women were tested for FMD response during their menses.

Participants who had any known metabolic, pulmonary, or cardiovascular disease were excluded from participation for this study. Individuals that had a musculoskeletal injury that prevented physical activity or those who could not follow written or verbal instructions were also not eligible. Finally, any woman who was currently pregnant, was trying to become pregnant, or had an irregular menstrual cycle was not eligible to participate. Women utilizing birth control were included on a case by case basis, as some contraceptives' effects on endothelial function had not been investigated. Oral contraceptives that allowed for a menses between 5 to 7 days were allowed as our testing timeline required multiple testing days. Other contraceptives that did not allow for a regular menses were excluded.

Preliminary Procedures

Participants underwent a preliminary visit day where they were given a health questionnaire, university-approved consent form, and became familiarized with the study protocols. Participants were randomly assigned to either a high-intensity interval exercise group (HT) or a concurrent exercise group (CT).

After a preliminary visit, participants were asked when the next expected start of menses was to occur. Two weeks before the expected start of menses, participants underwent anthropometric measurements. Height and weight were measured using a stadiometer (Perspective Enterprises, Kalamazoo, MI) and a digital scale (Tanita Worldwide, Arlington Heights, IL). Body mass index (BMI) was calculated. Waist-to-hip ratio (WHR) was also measured, using a tape measure to determine the narrowest part of their waist above their navel, and around the largest part of their hips at their buttocks.

Cardiorespiratory fitness was also determined using the Bruce protocol on a motorized treadmill (Quinton Tm65, Miluakee, WI). Expired respiratory gasses were continuously collected by indirect calorimetry (True One 2400 Metabolic System, Parvomedics, Sandy, UT). Heart rate was monitored by a Polar heart rate strap and watch (Polar Inc, Lake Success, NY). Exercise continued until the participant reached maximum exertion. The test was deemed successful if a plateau in $\dot{V}O_2$ was reached (defined as < 150 ml/min $\dot{V}O_2$ despite an increase in workload), if the participants achieved a heart rate within 10 bpm of age-predicted HR_{max} (220-age), or if the respiratory exchange ratio was greater than 1.1. Participants were encouraged to continue to exercise throughout the test, and a spotter was used.

Protocol Overview

One week after the anthropomorphic measurements, participants in the CT group met back in the Pioneer Hall weight room (room 223) to establish an 8 to 12 repetition maximum for all the exercises used in the exercise protocol. Within 24 hr of the onset of menses (reported using a tracking application on the phone and self-report), each participant underwent FMD testing in the morning and started the exercise protocol in the evening, roughly 12 hr post-FMD (day 1 of the study). Twelve hours after the start of the first session of exercise (day 2), the participant underwent another bout of FMD testing. Participants completed a second exercise session 48 hr after the first exercise session (day 3), and a third exercise session 48 hr after the second session (day 5). A final FMD measurement occurred 12 hr after the last exercise session, while the participant was still on her menses (day 6).

Flow-Mediated Dilation Measurements

FMD was assessed using a portable ultrasound (GE Healthcare, Madison, WI) and an automatic cuff inflator (Hokanson Inc, Bellvue, WA). FMD of the brachial artery was assessed on the right arm of each participant. Each participant lay supine in a darkened room for 10 min, with her arm abducted at 90° with complete elbow extension. The brachial artery was scanned longitudinally above the antecubital space, and the diameter measured. With the transducer held in place, a cuff placed on the forearm was inflated to 250 mmHg for 4.5 min, then deflated as brachial artery diameter, peak blood flow, and shear stress measurements were recorded using the ultrasound 60 and 90 s after cuff deflation. The still frames saved from the baseline and peak measurements were uploaded into MATLAB (Mathworks, Natick, MA), and an edge detection method, known as Canny, was applied. The edge detection software enhanced the edges of the artery wall to allow for a more accurate measurement of arterial diameter. The distance between the artery walls was calculated in MATLAB. The FMD% was calculated by the following:

$$\text{FMD\%} = \frac{\text{Peak Diameter} - \text{Baseline Diameter}}{\text{Baseline Diameter}} \; x \; 100$$

Training Sessions

During training days, participants were asked to arrive at the gym in the evening, approximately 12 hrs after the first FMD session following the start of their menses (Pioneer Hall room 223). Participants were allowed a 5-min warm up and stretch routine before starting the respective exercise sessions.

High-intensity interval exercise group (HT)

For the HT group, participants completed a 5-min warm up on the treadmill at 50 to 60% HR_{max}. After the warm up, the participants completed eight, 1 min bouts at 85 to 95% of HR_{max} (determined from the speed and grade on the treadmill during the $\dot{V}O_{2max}$ test). One minute of active rest separated the bouts. Heart rate was continuously monitored during exercise using a Polar heart rate monitor that was fitted around the chest. Once the warm up was completed, the treadmill speed and grade were increased to the speed known to elicit the heart rate response desired with the participant on the treadmill. The participants completed the 1 min high-intensity exercise bout. The speed was then lowered for the 1 minute of active rest between working bouts. The participants completed eight high-intensity exercise bouts for each of the three exercise sessions over the 1-week period.

Concurrent exercise group (CT)

For the CT group, strength exercises included three split muscle group sessions (i.e., upper and lower body days). The participants completed 8 exercises in a circuit pattern for 3 sets with 2 min of rest between each circuit. There was no structured rest period between exercises. Participants were encouraged to move quickly from device to device between sets. The weights used in the week before the start of the study were used to achieve the 8 to 12 repetition range, the weights were kept the same between each set for each exercise unless participant was too fatigued. At this time, the weight was lowered for safety. The two upper body workout days were separated into exercises that targeted the chest, triceps, and deltoids, and those that targeted the latissimus dorsi, trapezius, and biceps. The lower body workout day included exercises that targeted the quadriceps, hamstrings, gastrocnemius, glutes, and hip adductor, and abductor muscles.
Exercises that targeted the chest, triceps, and deltoids (day 1) included:

1. Bench press- exercise with participants lying on a flat bench with feet planted on the floor, participants grasped barbell slightly wider than shoulder width apart over head. Participants pushed weight off rack until its directly over chest, participants bent arms at elbow to bring weight to chest and then pushed weight back upward until arms are fully extended.

2. Pectoral butterfly- a seated exercise where the participants had arms extended outward from chest either holding dumbbells and flexed arms extended towards chest until hands come together.

3. Vertical chest press- a seated exercise where the participants pushed against handles at chest level directly outward from their body.

4. Lying barbell triceps extension- an exercise where the participants lie on a bench with feet on the floor grasping a barbell closer than shoulder width apart directly in front of them. Participants arms fully extended at a 90-degree angle from their torso and the floor. The palms faced outward and the elbows were tucked in, bent at the elbow bringing the knuckles towards the face until the bar was a few inches from forehead, then they extended arms directly outward again.

5. Triceps pushdown- a seated exercise where the participants grasped two "arms" of the machine in front of body with palms facing towards each other (neutral grip) and elbows tucked close to body. The participants pushed the arms of the machine down until their arms are fully extended.

6. Shoulder press- a seated exercise where the participants pressed a weight from shoulder height directly above head until arms were fully extended.

7. Dumbbell front raise- a standing exercise where the participants grasped dumbbells with palms facing the thighs anteriorly, the participants then raised arms (alternatively) directly in front of body with arms fully extended until horizontal.

8. Dumbbell lateral raise- a standing exercise where the participants grasped dumbbell arms hanging at their sides palms facing side of thighs (neutral grip), simultaneously the

participant raised the weight to the sides with arms fully extended until horizontal forming a T-shape.

Exercises that targeted the latissimus dorsi, trapezius, and biceps (day 3) included:

1. Latissimus pulldown (machine)- grasp a bar overhead with palms facing away from body, wider than shoulder width apart, pull down to collar bones.
2. Seated row (machine)- a seated exercise where the participant grasped a long bar slightly greater than shoulder width apart at upper chest level with palms facing away from body (towards the bar), the participants leaned back slightly but kept back straight, retracts the scapula then pulled the bar towards chest with elbows flared out.
3. Low-pulley seated row (machine)- a seated exercise where the participant grasped a double handle bar with palms facing each other in a neutral grip, the participants leaned back slightly but kept back straight, retracted the scapula then pulled handles towards lower abdomen with elbows tucked close to the body.
4. Upright row- participant grasped barbell slightly closer than shoulder width apart with palms facing towards the body (pronated grip) resting on top of thighs, pulled weight upward towards chest ending at collar bones.
5. Shrugs- grasped dumbbells with arms hanging at side, palms facing thighs (neutral grip), participants elevated shoulders upwards.
6. Barbell biceps curl- grasped barbell with palms facing outwards from body (supinated grip), curled towards chest.
7. Dumbbell hammer curl- grasped dumbbells with arms hanging at sides palms facing thighs (neutral grip), curled towards shoulder.

8. Dumbbell reverse curl- grasped dumbbells with palms facing towards the body (pronated grip), curled knuckles towards shoulder.

Exercises that targeted lower body musculature (day 5) included:

1. Back squat- standing exercise with a weighted barbell, the barbell was laid across the shoulders of the participants with their hands grasping the barbell slightly wider than shoulder width apart. The participants (with back straight and legs wider than shoulder width apart) bent at the waist and knees until their thighs were parallel with the floor, the participants then pushed weight back up until they were standing upright again.

2. Leg press- seated exercise, feet were placed on flat surface in front of the participants, the participants extended legs pushing weight away from body (does not fully extend legs to "lock out" knees).

3. Lunges- a standing exercise where the participants kept one foot planted, the other foot stepped forward, the planted leg bends with the knee coming close to the floor but never touching.

4. Leg curl- a lying exercise, a pad was placed against the back of the ankles, the participants flexed their hamstrings bringing the pad closer to their gluteus.

5. Leg extension- seated exercise, a pad was placed on top of the ankles, the participants flexed their quadriceps extending their legs, bringing the pad out level with the seat.

6. Hip adduction- seated exercise, started with legs spread (pads on interior of thigh), pushed inward against pad until legs came together.

7. Hip abduction- seated exercise, started with legs spread (pads on exterior of thigh), pushed outward against pad to spread legs.

8. Calf raises- seated or standing exercise where weight is on the upper portion of body, the participant extended foot downward causing the gastrocnemius to flex.

The participants in the CT group also completed the HIIT protocol in a similar manner as the HT group. After strength training, the participants stood quietly for 5 to 10 min, until heart rate values returned to within 10 beats of baseline before starting the HIIT protocol.

Statistical Analysis

A Friedman's non-parametric two-way analysis-of-variance (ANOVA) with a significance level of $p = .05$ for each mode of training was performed to determine the significant responses and if a difference was present in the responses. The Wilcoxon test was used to test differences within groups between first and third FMD trials. Statistical computations were carried out using SPSS Statistical software package version 19 (IBM; Armonk, NY).

CHAPTER IV

RESULTS

Participants in this study included recreationally trained females (*n* = 10) divided into two groups, HT and CT. There were initially 15 participants, but due to injury (not related to the study procedures), health, and non-responses, only 10 participants fully completed all procedures. Descriptors (see Table 1) such as age, body mass, height, BMI, WHR, and $\dot{V}O_{2max}$ were measured at baseline. Only one participant in the 10 had an abnormal menstrual cycle (off by over a week due to sickness and stress) before testing, so testing and exercise were delayed for this participant until it returned to a normal schedule.

Table 1

Descriptive Measures

	HT (*n* = 5)	**CT (*n* = 5)**
Age (yrs)	20.0 ± 2.0	20.4 ± 2.5
Body Mass (kg)	61.5 ± 7.6	69.6 ± 8.2
Height (cm)	161.0 ± 4.2	163.8 ± 6.5
BMI (kg/m^2)	23.7 ± 2.2	25.9 ± 1.7
WHR	0.77 ± 0.05	0.76 ± 0.07
$\dot{V}O_{2max}$ (ml/kg/min)	36.9 ± 5.4	38.3 ± 4.8

Note. Values are mean ± SD. HT—high-intensity interval training exercise group; CT—concurrent training exercise group; BMI—body mass index; WHR—waist-to-hip ratio; $\dot{V}O_{2max}$—maximal oxygen consumption.

There were no observed statistically significant differences noted for FMD% across all time points and training programs, and no statistical differences were observed in brachial artery diameter measurements for either group across all time points (see Table 2). Some non-significant but noticeable variations were seen in the results, these were thought to be variations attributed to within participant variations that occur day-to-day or trial-to-trial.

Table 2

Flow-Mediated Dilation Measurements

		Group	**Baseline**	**After 1 Session**	**After 3 Sessions**
FMD%		HT	11.54 ± 6.92	12.11 ± 3.45	11.65 ± 3.58
		CT	9.15 ± 6.90	12.44 ± 5.22	9.40 ± 4.78
BA diameter (cm)	BO	HT	0.31 ± 0.02	0.33 ± 0.03	0.33 ± 0.03
		CT	0.34 ± 0.02	0.33 ± 0.02	0.33 ± 0.01
	AO	HT	0.36 ± 0.03	0.36 ± 0.04	0.38 ± 0.03
		CT	0.37 ± 0.01	0.37 ± 0.01	0.36 ± 0.00

Note. Values are mean ± SD. BA—brachial artery; HT—high-intensity interval training exercise group; CT—concurrent training exercise group. BO—before occlusion; AO—after occlusion.

A graphical representation of FMD% data for all participants can be found in Figure 1.

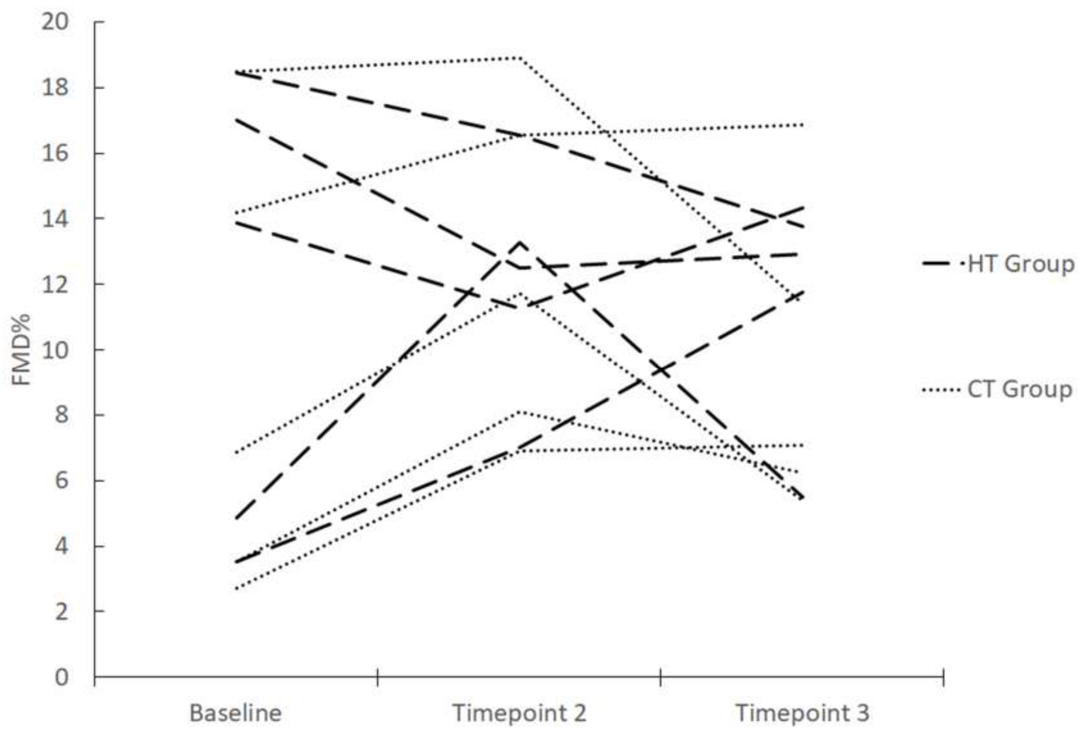

Figure 1. Flow-Mediated Dilation Changes

CHAPTER V

DISCUSSION

The purpose of this study was to compare the effect of two different modes of exercise on FMD in the brachial artery in recreationally active, college-aged women. FMD is a useful technique to non-invasively quantify endothelial function. When characterizing FMD% following acute exercise, the focus of most previous reports has included populations with a chronic disease or disorder that have inherent endothelial dysfunction, including those with cardiovascular disease (Bailey et al., 2018, Farsidfar et al., 2008), type 2 diabetes (Schreuder et al., 2014), obesity (Harris et al., 2008) and other at-risk populations, including post-menopausal women (Shah et al., 2019) and older adults (Yoo et al., 2017). While some studies have included the measurement of FMD following acute exercise in young adults (Dawson et al., 2013, Hwang et al., 2012), the acute effects of differing modes of exercise on endothelial health measured via FMD% have not been made. In this study, no statistically significant differences were noted for FMD% across all time points for either training program, and no statistically significant differences were observed in the brachial artery diameter for either group across all time points. The findings of this study lay a framework for future research studies conducted by strength and conditioning professionals and healthcare practitioners that include a characterization of endothelial function during specific modalities of exercise.

Baseline brachial artery diameter values in this study were similar to those reported by Rickenlund et al. (2005). The authors measured the brachial artery diameter in young female cyclists before and after oral contraceptive use. In that study, participants were taking oral contraceptives. Rickenlund et al. (2005) found that FMD% did not significantly increase due to oral contraceptive use alone (i.e., no additional intervention, such as exercise, was implemented). Though only one participant in this study used an oral contraceptive, oral contraceptive use may have factored into the non-significant findings observed with FMD% in this study.

While much of the literature includes males as participants, the studies that include females are limited. The studies that include a measurement of FMD following acute exercise in women are very limited. The baseline FMD% changes in this study were similar to those reported by Ballard et al. (2008), although the participants were males, but with a similar training history. Differences observed in FMD% between this study and the study by Rickenlund et al. (2005) may be related to the fact that the female participants recruited by Rickenlund et al. were endurance trained in specific sports (e.g., triathletes, cross-country skiers), included adult women (16 to 34 years), and were classified as “normal weight” according to BMI (Rickenlund et al., 2005). All of these factors can have an influence on FMD responses (Montero et al., 2014, Thijssen et al., 2011).

However, when characterizing FMD%, Green et al. (2013) found no differences in endothelium-dependent vasodilation in the brachial artery between groups of athletes using similar methods of ischemia to promote changes in artery diameter during FMD measurements. These participants had a history of performing sport specific training for at least 17 hr a week, for more than two years, and participated in a variety of sports including canoeing, squash, running, and cycling. The similar FMD% values in these athletes was thought to be due to the remodeling of peripheral arteries, leading to a larger diameter at baseline. The inherent remodeling of the endothelium with exercise training may lead to similar diameter changes in the active population (Green et al., 2013). Since only active individuals were recruited in this present study, this may explain why there were no significant differences between groups despite the intervention differences. It is interesting to note that when compared to sedentary individuals, FMD% values in the brachial artery are significantly lower in those who are active (Green et al., 2013).

The increased baseline brachial artery diameter in young, active adults has recently been confirmed by Montero et al. (2014). In their meta-analysis, the authors also found that, among those who are active, older individuals (i.e., greater than 40 years) exhibit greater responses to FMD when compared to younger individuals (i.e., less than 40 years). Not only can exercise training attenuate the decline in endothelial function that is associated with aging, but FMD% may also be age dependent (Montero et al., 2014). In the present study, the population was young, active females, all of whom were less than 40 years old. This may also explain why there was no statistical difference between the intervention groups. If the study was to include older participants, whether sedentary or active, who perform similar interventions, this may allow for statistical differences between exercise modalities to be found.

The non-significant results in FMD% may have also been due to the recruitment of a mixture of women some of who were classified as overweight according to BMI. Choo et al. (2014) found no differences between various exercise modalities (aerobic versus resistance versus a combination) with FMD% in overweight women. Although Choo et al. incorporated exercise training into their study and measured FMD% every 3 months, the FMD% values were similar to the FMD% values measured in this study. Any changes (or lack of changes) in FMD% may be a result of within-participant variations and lifestyle factors, including exercise habits and diet (Choo et al., 2014). These factors were not controlled in this study.

Limitations

There were some limitations associated with this study. This study was not sufficiently powered to examine variations in endothelial function, that may be, at least partially, explained by age or body composition. With such a small sample size (*n* = 10), FMD%, and brachial artery diameter (e.g., mean differences) may not be reflective of actual population parameters. Another limitation was that only females were recruited for this study. Although there was an attempt to control for menstrual cycle phase, FMD% may not be extrapolated to males. Another limitation was that there was no control group in this study that could have included recreationally active women who did not perform any exercise, or sedentary participants who performed similar interventions. Finally, only acute responses to exercise were investigated in this study. The structure of the endothelium, and thus measurements related to FMD, may change over time with more long-term exercise (Green et al., 2013).

Future Research

Future research studies could include an expansion of the participant pool to include men, older individuals, sedentary individuals from both sexes, or even those with a chronic condition that negatively affects the endothelium (e.g., heart disease, diabetes). Significant changes between groups and training status should be investigated. An expansion of the timeline for training and testing, or training over a longer period (i.e., 2 months or longer) could increase the possibility of physiological changes expressed, especially for the less trained individuals in the study. Finally, other variables that are indicative of arterial health, such as vascular endothelial growth factor or nitric oxide, should be investigated along with changes in FMD%.

Conclusion

In conclusion, although no statistical differences in the FMD values were observed, there is still much more to be learned from the effects of acute exercise on endothelial function. Incorporating changes, such as older adults and changes in intervention structure, could lead to significant differences in peripheral artery remodeling.

REFERENCES

American College of Sports Medicine. (2018). *ACSM's guidelines for exercise testing and prescription (10th ed.)* Philadelphia: Lippincott, Williams, & Wilkins.

Bailey, T. G., Perrisiou, M., Windsor, M. T., Schulze, K., Nam, M., Magee, R.,…Askew, C. D. (2018). Effects of acute exercise on endothelial function in patients with abdominal aortic aneurysm. *American Journal of Physiology, 313,* H19-H30. doi: 10. 1153/ajpheart. 00322.2017

Ballard, K. D., Miller, J. J., Robinson, J. H., & Olive, J. L. (2008). Aerobic capacity and postprandial flow mediated dilation. *International Journal of Exercise Science*, 1, 163-176.

Botonis, P. G., Toubekis, A. G., & Platanou, T. I. (2015). Concurrent strength and interval endurance training in elite water polo players. *Journal of Strength and Conditioning Research, 30*, 126-133. doi: 10. 1519/JSC.0000000000001091

Calder, A. W., Chilibeck, P. D., Webber, C. E., & Sale, D. G. (1994). Comparision of whole and split weight training routines in young women. *Canadian Journal of Applied Physiology, 19*, 195-199.

Celermajer, D. S., Sorenson, K. E., Gooch, V. M., Speigelhalter, D. J., Miller, O. I., Sullivan, I. D., Lloyd, J. K., & Deanfield, J. E. (1992). Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. *Lancet, 340,* 1111-1115.

Centers for Disease Control and Prevention (CDC). (2017). Obesity and Overweight. Retrieved from https://www.cdc.gov/nchs/fastats/obesity-overweight.htm

Choo, J., Lee, J., Cho, J. H., Burke, L. E., Sekikawa, A., & Jae, S. Y. (2014). Effects of weight management by exercising modes on markers of subclinical atherosclerosis and cardiometabolic profile among women with abdominal obesity: A randomized controlled trial. *BMC Cardiovascular Disorders, 14,* 82. doi: 10.1186/1471-2261-14-82.

Dawson, E. A., Green, D. J., Cable, N. T., & Thijssen, D. H. (2013). Effects of acute exercise on flow-mediated dilatation in healthy humans. *Journal of Applied Physiology, 115,* 1589-1598. doi:c10. 1152/japplphysiol.00450.2013

Donnelly, J. E., Blair, S. N., Jakicic, J. M., Manore, M. M., Rankin, J. W., Smith, B. K., & American College of Sports Medicine. (2009). ACSM Position Stand: Appropriate physical activity intervention strategies for weight loss and prevention of weight regain for adults. *Medicine & Science in Sports & Exercise*, *41*, 459-471. doi: 10. 1249/MSS.0b013e3181949333

Farsidfar, F., Kasikcioglu, E., Oflaz, H., Kasikcioglu, D., Meric, M., & Umman, S. (2008). Effects of different intensities of acute exercise on flow-mediated dilatation in patients with coronary heart disease. *International Journal of Cardiology*, *124*, 372-374.

Farzad, B., Gharakhanlou, R., Agha-Alinejad, H., Curby, D. C., Bayati, M., Bahraminejad, M., & Maestu, J. (2011). Physiological and performance changes from the addition of a sprint interval program to wrestling training. *Journal of Strength and Conditioning Research, 25*, 2392-2399. doi: 10. 1519/JSC.0b013e3181fb4a33

Franceschini, S. A., Vieira, C. S., Matins, W. P., Franca, J. B., & Ferriani, R. A. (2013). Effects of combined oral contraceptives containing levonorgestrel or chlormadinone on the endothelium. *Contraception, 87,* 766-772. doi: 10. 1016/j.contraception.2012.09.023

Gibala, M. J., Little, J. P., Van Essen, M., Wilkin, G. P., Burgomaster, K. A., Safdar, A.,… Tarnopolsky, M. A. (2006). Short-term sprint interval versus traditional endurance training: Similar initial adaptations in human skeletal muscle and exercise performance. *Journal of Physiology, 575*, 901-911.

Gibala, M. J., Little, J. P., MacDonald, M. J., & Hawley, J. A. (2012). Physiological adaptations to low-volume, high-intensity interval training in health and disease. *Journal of Physiology, 590*, 1077-1084. doi: 10. 113/jphysiol.2011.224725

Gori, T., Muxel, S., Damaske, A., Radmacher, M-C., Fasola, F., Schaefer, S.,…Munzel, T. (2011). Endothelial function assessment: Flow-mediated and constriction provide different and complimentary information on the presence of coronary artery disease. *European Heart Journal*, 33, 363-371. doi: 10. 1093/eurheartj/ehr361

Green, D. J., Rowley, N., Spence, A., Carter, H., Whyte, G., George, K.,…Thijssen, D.H. J. (2013). Why isn't flow-mediated dilation enhanced in athletes? *Medicine & Science in Sports & Exercise, 45,* 75-82. doi: 10. 1249/MSS.0b013e318269affe

Harris, R. A., Padilla, J., Hanlon, K. P., Rink, L. D., & Wallace, J. P. (2008). The flow-mediated dilation response to acute exercise in overweight active and inactive men. *Obesity, 16,* 578-584. doi: 10. 1038/oby.2007.87

Hashimoto, M., Akishita, M., Eto, M., Ishikawa, M., Kozaki, K., Toba, K.,…Ouchi, Y. (1995). Modulation of endothelium-dependent flow-mediated dilation of the brachial artery by sex and menstrual cycle. *American Heart Association Journal of Circulation*, 92, 3431-3435.

Heidarzadeh, Z., Asadi, B., Saadatnia, M., Ghorbani, A., & Fatehi, F. (2014). The effect of low-dosed combined oral contraceptive pills on brachial artery endothelial function and common carotid artery intima-media thickness. *Journal of Stroke and Cerebrovascular Disease*, 23, 675-680. doi: 10. 1016/j.strokecerebrovasdis.2013.06.007

Helgerud, J., Hoydal, K., Wang, E., Karlsen, T., Berg, P., Bjerkaas, M.,…Hoff, J. (2007). Aerobic high-intensity intervals improve $V0_{2max}$ more than moderate training. *Medicine & Science in Sports & Exercise*, 34, 665-671.

Hwang, I. C., Kim, K. H., Choi, W. S., Kim, H. J., Im, M. S., Kim, Y. J.,…Zo, J. H. (2012). Impact of acute exercise on brachial artery flow-mediated dilatation in young healthy people. *Cardiovascular Ultrasound, 10,* 39. doi: 10.1152/japplphysiol.00450.

Janse de Jonge, X. A., Boot, C. R., Thom, J. M., Ruell, P. A., & Thompson, M. W. (2001). The influence of menstrual cycle phase on skeletal muscle contractile characteristics in humans. *Journal of Physiology, 530*, 161-166.

Laird, R. H., Elmer, D. J., Barberio, M. D., Salom, L. P., Lee, K. A., & Pascoe, D. D. (2016). Evaluation of performance improvements after either resistance training or sprint interval-based concurrent training. *Journal of Strength and Conditioning Research, 30*, 3057-3065.

Laursen, P. B., & Jenkins, D. G. (2002). The scientific basis for high-intensity interval training: Optimising training programs and maximising performance in highly trained endurance athletes. *Sports Medicine, 32*, 53-73.

Lebrun, C. M., McKenzie, D. C., Prior, J. C., & Tauton, J. E. (1995). Effects of menstrual cycle phase on athletic performance. *Medicine & Science in Sports & Exercise*, 27, 437-444.

Meyer, B., Mortl, D., Strecker, K., Hulsmann, M., Kulemann, V., Neunteufl,T.,…Berger, R. (2005). Flow-mediated vasodilation predicts outcome in patients with chronic heart failure: comparison with B-type natriuretic peptide. *Journal of the American College of Cardiology*, 46, 1011-1018.

Montero, D., Padilla, J., Diaz-Cãnestro, C., Muris, D. M. J., Pyke, E., Obert, P., & Walther, G. (2014). Flow-mediated dilation in athletes: Influence of aging. *Medicine & Science in Sports & Exercise*, 46, 2148-2158. doi: 10. 1249/MSS.0000000000000341

Patel, A. R., Kuvin, J. T., Sliney, K. A., Rand, W. M., Chiang, J. C., Udelson, J. E.,...Karas, R. H. (2005). Gender-based differences in brachial artery flow-mediated vasodilation as an indicator of significant coronary artery disease. *American Journal of Cardiology, 96,* 1223-1226.

Pyke, K. E., Dwyer, E. M., & Tschakovsky, M. E. (2004). Impact of controlling shear rate on flow mediated dilation in the brachial artery of humans. *Journal of Applied Physiology, 97*, 499-508.

Pyke, K. E., & Tschakovsky, M. E. (2005). The relationship between shear stress and flow mediated dilation: Implications for the assessment of endothelial function. *Journal of Physiology, 568*, 357-369.

Rakobowchuk, M., McGowan, C. L., De Groot, P. C., Hartman, J. W., Phillips, S. M., & MacDonald, M. J. (2005). Endothelial function of young healthy males following whole body resistance training. *Journal of Applied Physiology, 98*, 2185-2190.

Rakobowchuk, M., Tanguay, S., Burgomaster, K. A., Howarth, K. R., Gibala, M. J., MacDonald, M. J. (2008). Sprint interval and traditional endurance training induce similar improvements in peripheral arterial stiffness and flow-mediated dilation in healthy humans. *American Journal of Physiology, 295*, R236-R242. doi: 10. 1152/ajpregu.00069.2008

Rickenlund, A., Eriksson, M. J., Schenck-Gustafsson, K., & Hirschberg, A. L. (2005). Oral contraceptives improve endothelial function in amenorrheic athletes. *Journal of Clinical Endocrinology and Metabolism*, 90, 3162- 3167.

Schreuder, T. H., van Lotringen, J. H., Hopman, M. T., & Thijssen, D. H. (2014). Impact of endothelin blockade on acute exercise-induced changes in blood flow and endothelial function in type 2 diabetes mellitus. *Experimental Physiology, 99,* 1253-1264.
doi: 10. 1113/expphysiol.2013.077297

Shah, M., Bailey, S., Gloeckner, A., Kreutzer, A., Adams-Huet, B., Cheek, D., & Mitchell, J. (2019). Effect of acute exercise on postprandial endothelial function in postmenopausual women: A randomized cross-over study. *Journal of Investigative Medicine.* doi: 10.1136/jim-2019-000992

Shechter, M., Issachar, A., Marai, I., Koren-Morag, N., Freinark, D., Shahar, Y.,…Feinberg, M. S. (2009). Long-term association of brachial artery flow-mediated dilation and cardiovascular events in middle-aged subjects with no apparent heart disease. *International Journal of Cardiology, 134,* 52-58.
doi: 10. 1016/j.ijcard.2008.01.021

Shechter, M., Shechter, A., Koren-Morag, N., Feinberg, M. S., & Hiersch, L. (2014). Usefulness of brachial artery flow-mediated dilation to predict long-term cardiovascular,events in subjects without heart disease. *American Journal of Cardiology, 113,* 162-167.
doi: 10. 1016/j.amjcard.2013.08.051

Sperlich, B., De Marees, M., Koehler, K., Linville, J., Holmberg, H. C., & Mester, J. (2011). Effects of 5 weeks of high-intensity interval training vs. volume training in 14-year old soccer players. *Journal of Strength and Conditioning Research, 25*, 1271-1278. doi: 10. 1519/JSC.0b013e3181d67c38

Thijssen, D. H., Black, M. A., Pyke, K. E., Padilla, J., Atkinson, G., Harris, R. A.,…Green, D. J. (2011). Assessment of flow-mediated dilation in humans: A methodological and physiological guideline. *American Journal of Physiology, 300,* H2-H12. doi: 10. 1152/ajpheart.00471.2010

Warburton, D. E., McKenzie, D. C., Haykowsky, M. J., Taylor, A., Shoemaker, P., Ignaszewski, A. P., & Chan, S. Y. (2005). Effectiveness of high-intensity interval training for the rehabilitation of patients with coronary artery disease. *American Journal of Cardiology, 95*, 1080-1084.

Yoo, J. K., Pinto, M. M., Kim, H. K., Hwang, C. L., Lim, J., Handberg, E. M., & Christou, D. D. (2017). Sex impacts the flow-mediated dilation response to acute aerobic exercise in older adults. *Experimental Gerontology, 91,* 57-63. doi: 10. 1016/l.exger.2017.02.069

APPENDIX A

FMD Raw Data

Group	BA Base (cm)	BA Max Base (cm)	FMD Base	BA Base 2 (cm)	BA Max 2 (cm)	FMD 2	BA Base 3 (cm)	BA Max 3 (cm)	FMD 3
HT	0.3046	0.3985	17.012	0.3667	0.4125	12.4896	0.3626	0.4094	12.9155
HT	0.3066	0.3625	13.876	0.3301	0.3672	11.2628	0.3204	0.3910	14.3376
HT	0.3485	0.3655	4.8568	0.3413	0.3866	13.2589	0.3722	0.3931	5.4958
HT	0.3089	0.37	18.445	0.3116	0.363	16.536	0.298	0.339	13.768
HT	0.3008	0.3114	3.5288	0.2756	0.2949	7.0185	0.3203	0.3579	11.7436
CT	0.3483	0.3578	2.731	0.3495	0.3737	6.9075	0.3457	0.3702	7.087
CT	0.3089	0.3660	18.47	0.3036	0.3611	18.9203	0.3221	0.3589	11.4245
CT	0.3457	0.3947	14.187	0.3229	0.3763	16.5561	0.3154	0.3686	16.8673
CT	0.3535	0.3659	3.5202	0.3515	0.3800	8.1003	0.3439	0.3653	6.2245
CT	0.3376	0.3608	6.861	0.3175	0.3547	11.731	0.3360	0.3541	5.3768

APPENDIX B

CT Group Training Volume

Participant #	Volume (lbs) for Day 1	Volume (lbs) for Day 3	Volume (lbs) for Day 5	Total Volume (lbs)
1	5388	8490	14090	27968
4	6302	6468	13200	25970
7	10008	12132	37740	59880
10	4728	7608	19890	32226
13	8028	8282	20990	37300

APPENDIX C

Informed Consent

Exercise Physiology Laboratory
Texas Woman's University
304 Administration Dr.
Denton, TX 76204-5647

PI: Ashley Allen, B.S
Phone: 936-672-6078
Aallen31@twu.edu

TEXAS WOMAN'S UNIVERSITY CONSENT TO PARTICIPATE IN RESEARCH
for a Research Study entitled

"Acute Effects of High Intensity Interval Training vs. Concurrent training and flow mediated dilation in college-aged women"

PURPOSE

You are being asked to participate in a research study using two different modes of exercise and their effect on flow mediated dilation (FMD): high intensity interval training (HIIT), and a concurrent training group that involves strength training and HIIT. Flow mediated dilation is the hyperemic response of a blood vessel following occlusion. This means FMD is the measurement of the arteries diameter expansions (hyperemic response) after blood flow is allowed to return. High intensity interval training is a form of interval training alternating periods of high intensity (85 to 95% of maximal exertion) exercise with less intense rest periods. There is research published on the effectiveness of HIIT on $V0_{2max}$ especially in sedentary individuals and HIIT on FMD in post cardiac event populations. There are little to no research studies in which a comparison is made between the two exercise modalities effect on FMD in adult college aged women. **The purpose of this study is to compare the effect of two different modes of exercise on flow mediated dilation in the brachial artery. A secondary measure will include maximal oxygen uptake, body fat, and muscular strength.**

PARTICIPANT REQUIREMENTS and PRELIMINARY SCREENING

Participant Criteria

Participants for this study will be adult females at TWU, aged 18 to 35 years old. Enrollment is open to adults of all ethnicities.

In order to be eligible to participate, you must have the following characteristics:

1. Are a female TWU student, faculty, or staff aged 18 to 35
2. Are recreationally active 2 to 3 times a week for at least one year
3. Are not underweight, or obese according to BMI (BMI between 18.5-29.9)
4. Are without orthopedic problems that could be exacerbated by exercise

Initials______

5. Are without a diagnosed congenital heart condition, known cardiovascular condition, or metabolic condition that would preclude you from exercise
6. Are free from any musculoskeletal injuries that would preclude you from exercise
7. Are not pregnant or plan to become so during the study
8. Do not have an irregular menstrual cycle
9. Can follow written/ verbal instructions

If you are currently taking medications, we ask that you do not change the schedule or dosage of your medications throughout the duration of the study.

EXPERIMENTAL METHODS and APPROACH

The preliminary visit, testing, and training sessions will take place in Pioneer Hall on TWU's campus in Denton, TX.

Preliminary Visit

An initial visit will occur at Pioneer Hall at TWU for all participants. At this time, you will be given a health/ exercise readiness questionnaire, a PAR-Q, become familiarized with the study protocols, and the informed consent to allow for participation in this study. During this visit, you will be allowed to ask questions about the study and your potential role in it. You will be randomly assigned to a high-intensity interval training only group (HT), or a concurrently trained exercise group (CT).

Testing Sessions

All experimental data will be collected in four or five testing sessions. Prior to the testing sessions, all experimental procedures will be reviewed with you, and you will be verbally re-assented. After a preliminary visit, you will be asked when the next expected start of their menses is. Two weeks before the expected start of menses, you will undergo anthropometric measurements (e.g., height, weight, BMI) and body fat percentage, and complete an incremental maximal oxygen uptake (V02max) test on a treadmill. One week after the anthropometric measurements, (if you are in the CT group) you will be asked to establish an 8 to 12 repetition maximum for all the exercises used in the training protocol. Approximately one week after this, and within 24 hr of the start of menses (reported using a tracking application on the phone and self-report), you will start the first training session. A total of 3 evening training sessions will take place, each 48 hr apart from each other. Twelve hours before the start of the first training session, you will undergo FMD testing. Twelve hours after the first training session, you will undergo a second FMD test. A final FMD measurement will take place 12 hr after the last training session while you are still in the follicular phase (menses). All FMD measurements will take place in the morning.

Height will be assessed using a stadiometer, and weight will be assessed using a digital scale. These two measurements be used to calculate your BMI. Body fat percentage will be assessed using dual-energy x-ray absorptiometry (DEXA). You will lay on the table supine with your arms at your side and asked to remain motionless for 6 minutes while the machine scans you from head to toe.

Your muscular strength will be assessed using a 8 to 12 repetition range for all the exercises listed in training protocol. The weight used in the repetition range for the testing session will be used in the training sessions to follow.

Initials_____

V02max will be assessed using the Bruce protocol and a metabolic cart. The Bruce protocol is a six stage (3 minutes per stage) treadmill test that increases speed and grade at the beginning of each new stage. You will be asked to complete a dynamic stretching protocol to minimize risk of muscular strain/ sprain. You will attach a heart rate monitor to your chest before testing. You will be fitted with head gear and a mouth piece that allows expiration to be analyzed by the metabolic cart. You will be spotted with one tester behind them as the other operates the treadmill. You will be allowed a cool down on the treadmill, at a lower speed and grade.

Flow mediated dilation will be assessed using a portable ultrasound and an automatic cuff inflator. Flow mediated dilation of the brachial artery will be assessed on your right arm. You will lie supine in a darkened room for 10 minutes, with your arm abducted at 90◦ with complete elbow extension. The brachial artery is then scanned longitudinally above the antecubital space, and the diameter measured. With the transducer held in place, a cuff placed on the forearm is inflated to 250 mmHg for 4.5 minutes then deflated with diameter, peak flow, shear stress measurements being recorded 60 and 90 seconds after deflation.

Training Sessions

Participants will be randomized into 2 groups and will be asked to meet at TWU's fitness area on the second floor in Pioneer Hall room 223.

1) HIIT group 2) HIIT and strength group (Concurrent). The training will last for one week and will be performed three times on nonconsecutive days, for a total of 3 training sessions.

HIIT group (HT)

For the HT group, you will be asked to complete a 5-min warm up on the treadmill at 50 to 60% $\dot{V}O2max$, followed by eight, 1 min bouts at 85 to 95% of $\dot{V}O2max$ based upon heart rate (determined from the speed on the treadmill during the $\dot{V}O2max$ test). One minute of rest will separate the bouts. Heart rate will be monitored during exercise using a Polar heart rate monitor that will be fitted around the chest. Once the warm up is completed the treadmill speed will be increased to the speed known to elicit the heart rate response desired. You will complete the one-min HIIT bout, then grasp onto the side hand rails, and jump off placing feet on either side of the moving belt for the rest period. You will complete eight HIIT bouts for each of the three-training sessions over the 1-week period.

Concurrent group (CT)

For the CT group, strength training will include split muscle group training (i.e., upper, and lower body days). The participants will be asked to complete 8 exercises in a circuit pattern for 3 sets with 2 min of rest between each circuit bout. The weights used in the week before the start of the study will be used to achieve the 8 to 12 repetition range.

Upper body workout days will be separated into; chest/ triceps/ deltoids, and latissimus dorsi/ trapezius/ biceps. The lower body workout day will work the quadriceps, hamstrings, gastrocnemius, gluteus, and hip adductor/ abductor muscles.

Exercises for chest/ triceps/ deltoids will be:

1. bench press
2. pectoral fly
3. vertical chest press
4. lying barbell triceps extension
5. triceps pushdown
6. shoulder press
7. dumbbell front raise
8. dumbbell lateral raise

Initials_____

Exercises for latissimus dorsi/ trapezius/ biceps will be:

1. latissimus pulldown (machine)
2. seated row (machine)
3. low-pulley seated row (machine)
4. upright row
5. shrugs
6. barbell biceps curl
7. dumbbell hammer curl
8. dumbbell reverse curl

Exercises for lower body days will be:

1. back squat
2. leg press
3. lunges
4. leg curl
5. leg extension
6. hip adduction
7. hip abduction
8. calf raises

The participants in the concurrent group will also complete the HIIT protocol in a similar manner as the HT group. After strength training, the participants will sit quietly for 5 to 10 min until heart rate values return to within 10 beats of baseline before starting the HIIT protocol.

In order for you to complete all requirements of this study, the maximum time commitment is:

Preliminary Visit:	**1.0 hours**
Testing Sessions HIIT (4 total):	**4.0 hours**
Testing Sessions Concurrent (5 total):	**5.0 hours**
Training Sessions HIIT (3 total):	**1.75 hours**
Training Sessions Concurrent (3 total):	**4.0 hours**
TOTAL TIME COMMITMENT (HIIT)	**6.75 hours**
TOTAL TIME COMMITMENT CONCURRENT	**10.0 hours**

Participant Benefits

For your participation, you will receive:

1. Your individual results from the testing

2. A written summary of the findings upon completion of the study

3. Instruction on proper weight lifting form and technique

There are currently no monetary benefits for participation in this study

Initials_____

Potential Risks and Protection of Participants

RISK	STEPS TO MINIMIZE RISK
Injury sustained while exercising on the treadmills	The primary risk of running on treadmills is falling off the treadmill. You will have appropriate supervision to ensure the risk of falling is minimized. When you are on the treadmill, one person will be standing on one side and another behind you, handrails are also located on either side and you will be directed to hold on if they feel they will lose balance.
RISK	STEPS TO MINIMIZE RISK
Sudden cardiac death	According to the American College of Sports medicine, there is little risk during submaximal exercise. This study uses maximal and submaximal exercise increasing the risk associated with testing. All investigators are AED/CPR certified. All of the physiologic risks inherent with exercise testing will be mitigated through preliminary screening, adherence to standards of practice for exercise testing that are published by the American College of Sports Medicine, and the monitoring of each test by trained personnel. Individuals with congenital heart disease or known cardiovascular disease will be excluded.
RISK	STEPS TO MINIMIZE RISK
Injury sustained while strength training with free weights and machine resistance systems	The primary risk involved with strength training is pulls/ strains or ligaments, tendons, and muscles. Although there is little risk associated with submaximal exercise, there is maximal testing in this study. All the physiologic risks associated with maximal and submaximal exercise will be reduced though screenings, adherence to exercise testing that is published by the American College of Sports Medicine, and the proper guidance of trained personnel.

Initials_____

RISK	STEPS TO MINIMIZE RISK
Injury sustained during V02max tests	The primary risk involved with the completion of a V02max test is, fatigue, dizziness, and syncope. While there is an increased risk due to the test being a max test the subjects risk of injury will be minimized by the presence of a trained test performer and a spotter behind the participant on the treadmill. You are meant to push to your personal maximum but will be allowed to stop the test when you deem it necessary.
RISK	STEPS TO MINIMIZE RISK
Anxiety/ Stress	The investigators will be available to talk to the you over any concern/ fear they have regarding aspects of the study and will answer questions honestly.
RISK	STEPS TO MINIMIZE RISK
Radiation	The primary risk involved with using the DEXA scanner is radiation. Though the radiation you are exposed to, is less than that of a chest x-ray. The technician running the scan will take the steps necessary to minimize the risk of prolonged exposure to radiation by positioning you correctly and having the test run properly.
RISK	STEPS TO MINIMIZE RISK
Discomfort due to flow mediated dilation techniques	The primary risk involved with flow mediated dilation is the discomfort/ loss of feeling associated with occluding blood flow in the forearm while running the test. The investigators will minimize this discomfort by having two investigators present during the test, and holding the inflation no longer than the protocol states.
RISK	STEPS TO MINIMIZE RISK
Emotional discomfort	Participation is voluntary and you may withdraw from the study at any time. Every attempt will be made to reduce your discomfort.

RISK	STEPS TO MINIMIZE RISK
Loss of confidentiality	It is possible that there might be a loss of your confidentiality with data stored offline. To minimize this risk, all data forms collected will be coded using alphanumeric IDs. A single identification form linking names with their respective IDs will be kept in a separate folder from the other data. Persons not associated with the study will have no access to the folders. Data collection sheets will be locked in a file cabinet in Pioneer Hall 123B. There is also a potential risk of loss of confidentiality in all email, downloading, and internet transactions.
RISK	STEPS TO MINIMIZE RISK
Loss of anonymity	It is possible that multiple participants may be tested at one time, or that testing may take place such that you exposed to the general public or students; because of this, participants will be informed, before the study begins, that the loss of anonymity may be present. You have the ability to request a separate time to complete the pool or lab assessment; however, anonymity cannot be protected even when the pool or lab is closed due to evening classes in Pioneer Hall and the glass windows that allow for the pubic to view the pool or lab. You may withdraw from the study at any time without penalty.
RISK	STEPS TO MINIMIZE RISK
Lightheadedness, nausea, and/or dizziness	It is possible that the testing and/or training sessions may cause lightheadedness, nausea, and/or dizziness. To minimize this risk the investigators will allow for rest periods and will act as safety spotters during maximal exertions. Your participation is voluntary and you are allowed to withdraw from the study at any time without penalty.

Initials_____

RISK	STEPS TO MINIMIZE RISK
Loss of time	It is possible that you may feel the time commitment is too lengthy and requires too much time. Participation in the study is voluntary and you may withdraw from the study at any time without penalty.
RISK	**STEPS TO MINIMIZE RISK**
Coercion	Services provided to you will not be affected by participation/non-participation in the study. Participation is voluntary and you may withdraw from the study at any time.

At the beginning of each session, all of the procedures will be briefly reviewed with you. We will obtain your verbal consent to participate in the day's procedures.

The researchers will try to prevent any problem that could happen because of this research. You should let the researchers know at once if there is a problem and they will help you. However, TWU does not provide medical services or financial assistance for injuries that might happen because you are taking part in this research.

YOUR RIGHTS TO PRIVACY

Confidentiality will be protected to the extent that is allowed by law. All individual information obtained in this study will remain confidential and your right to privacy will be maintained. Data collected will be used for research purposes only and will be limited to access by the investigators of this study. Only data reported as group means or responses will be presented in scientific meetings and published in scientific journals. Data will be destroyed within 5 years of study completion.

QUESTIONS ABOUT THIS RESEARCH

As investigators, it is our obligation to explain all of the procedures to you. We want to make sure that you understand what is required of you and what you can expect from us in order to complete this study. Please do not hesitate to inquire about the research, your rights and responsibilities as the participant, or our roles as the investigators now or at any time throughout the study.

YOUR CONSENT TO PARTICIPATE

Participation in this research is entirely voluntary. Your decision whether or not to participate will not jeopardize your future relations with Texas Woman's University and the Department of Kinesiology. You may withdraw your consent and discontinue participation at any time and for any reason without prejudice. Discontinuing your participation will involve no penalty.

Failure to comply with all of the procedures and to follow the instructions necessary for reliable and valid scientific measurements may result in termination of your participation in this study without your consent. You may be asked to withdraw if you fail to comply with all of the requirements for participation listed above. If you are withdrawn from participation by one of the investigators, our decision will not jeopardize your future relations with Texas Woman's University and the Department of Kinesiology.

CONTACT INFORMATION

Ashley Allen, B.S
Principal Investigator, Kinesiology
Phone: 936-672-6078
aallen31@twu.edu

Rhett Rigby, PhD
Professor, Kinesiology
Office: 940-898-2473
brigby@twu.edu

YOU WILL BE GIVEN A COPY OF THIS SIGNED AND DATED CONSENT FORM TO KEEP. IF YOU HAVE ANY QUESTIONS ABOUT THE RESEARCH STUDY YOU SHOULD ASK THE RESEARCHERS. IF YOU HAVE ANY QUESTIONS ABOUT YOUR RIGHTS AS A PARTICIPANT IN THIS RESEARCH OR THE WAY THIS STUDY HAS BEEN CONDUCTED, YOU MAY CONTACT THE TEXAS WOMAN'S UNVERSITY OFFICE OF RESEARCH AND SPONSORED PROGRAMS AT 940-898-3378 OR VIA EMAIL AT IRB@twu.edu

____________________	______	____________________	______
Participant's Signature	**Date**	**Investigator Obtaining Consent**	**Date**

____________________ ____________________

Printed Name **Printed Name**

Approved by the
Texas Woman's University
Institutional Review Board
Approved: August 31, 2017
Modifications Approved:
July 13, 2018