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MENSTRUAL CYCLE CHANGES IN QUADRICEP MUSCULAR ARCHITECTURE AND OTHER FUNCTIONAL PARAMETERS IN COLLEGE-AGED FEMALES COMPARED TO MALES

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MENSTRUAL CYCLE CHANGES IN QUADRICEP MUSCULAR ARCHITECTURE AND OTHER FUNCTIONAL PARAMETERS IN COLLEGE-AGED FEMALES COMPARED TO MALES

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ABSTRACT

Women have often been excluded from research studies due to the fluctuations of hormones throughout the menstrual cycle and the difficulty in scheduling through different phases and the possible interaction it may have with other testing variables. The purpose of this study was to investigate how the menstrual cycle may affect lower limb muscular architecture and functional parameters, such as: force production, range of motion (ROM) and body composition in females during the follicular (FOL), ovulatory (OV) and luteal (LUT) phases of the menstrual cycle. This study utilized a short-term longitudinal repeated measured design which consisted of 4 visits. There were three groups: Non-contraceptive users (NC), Oral-contraceptive users (OC) and a male control group (CON). Visit 1 began with an explanation of inclusion and exclusion criteria and familiarization for testing equipment. Visits 2-4 were scheduled based off of the participant's menstrual cycle phase by menstrual cycle questionnaires or estimated in the CON group by: FOL (days 1-4) , OV (days 12-15) and LUT (days 15-18). Visits 2-4 included hip and knee ROM goniometer assessment, a DXA scan, pennation angle (PA) measurements of the vastus medialis (VM), vastus lateralis (VL), rectus femoris (RF), and the anterior portion of the vastus intermedius (VIA) of the dominant leg using B-mode ultrasound and knee extension and flexion maximal voluntary isometric contraction (MVICs) testing on the dominant leg. Sigma Plot (Systat Software, San Jose, CA v. 12.5) was used to perform statistical analysis. Statistical significance was set at $p < 0.05$. 3-Way Repeated-measures ANOVA was used to identify differences between quadriceps muscle architecture, body composition, ROM and force production during the FOL, OV and LUT phases of the menstrual cycle between groups (OC, NC, CON) and time (FOL, OV, and LUT phases). There were no significant differences found in force production, pennation angle, range of motion or

body composition across the three groups (OC, NC, CON) or time points (follicular, ovulatory, luteal), The lack of significant changes in variables between the NC and OC groups indicates oral contraceptives may not play a role in altering changes in body composition, hip and knee ROM, isometric knee strength and quadriceps PA. However, it is important to control for the testing phase for future research studies to minimize error with hormone fluctuation through the menstrual cycle.

CHAPTER I

INTRODUCTION

Research studies show that female athletes have significantly more anterior cruciate ligament (ACL) tears compared to male athletes.[1] Women have a 4-8-fold greater risk of ACL tears than men do, especially during the pre-ovulation, particularly the first two days of menses.[2] This data showing a sex difference warrants an explanation regarding the potential physiological mechanisms responsible for this difference and if these differences are related to hormone fluctuations during the menstrual cycle. In addition, data has shown strength varies through the menstrual cycle. However the studies show conflicting results with some studies showing no changes[3] to others observing significant increases in strength during the ovulatory phase compared to luteal and follicular phases.[4] Body composition changes in relation to the mensural cycle have also been assessed by researchers but are limited by poor body composition methods (BMI) and did not test in all menstrual cycle phases (follicular, ovulatory and luteal).[5] Similarly, mobility and flexibility has not been assessed through all three phases and limited by equipment used to assess flexibility (Sit-and-Reach machine).[5] No other study, to our knowledge, has assessed quadriceps muscular architectural characteristics, such as Pennation angle (PA), through all three phases. PA is the angle in which the muscle fascicle extends from the deep aponeurosis (the extension of the tendon joining the muscle).[6] This angle allows us to correlate the direction of the muscle to how that muscle produces force. Water retention occurs during a woman's menstrual cycle, so the pressure accumulated may alter the pennation angle, thus altering the way women may produce

force and ultimately contribute to why women have an increased risk for injury when compared to men.

Birth control use in female athletes has been a topic of interest for many years now. Many studies are inconclusive when asked does contraceptive use provide a "protective effect" from injury.[2,7] Many studies fail to control for different types of contraceptives used, examples of popular types include: oral contraceptive (birth control pill), Intra-uterine device (IUD), Hormone implants, Contraceptive injection (Depo-Provera), Transdermal contraceptives (birth control patch), Vaginal contraception ring (birth control ring) and others. Hormone concentration varies between contraceptive type and brand.[8,9] One of the largest constraints in research studies that assess differences in users and non-users is when the researcher does not require adequate time for the hormone levels to stabilize. Some studies only required 2 months of being on birth control[5], while others required over 6 months to ensure hormone levels to regulate.[3] Many contraceptives will diminish or regulate menstrual cycle phases and influence impact hormones levels, so that why is crucial to allow time to ensure valid results.

There are many gaps in the literature when assessing menstrual cycle changes in body composition, force production, mobility and other parameters. To address these gaps, we are going to test during all three phases: follicular, ovulatory and luteal. To control for the potential testing effect a male only control group will be included in this study. Results from our study will inform future research protocols on how and when to assess female participants according to their menstrual cycle. Our results, whether or not significance is found, will help future researchers and physicians when developing injury

prevention protocols, rehabilitation programs, and future training approaches for both female athletes and females that are recreationally active.

Significance

Many studies have evaluated functional parameters (body composition, force production and mobility), but are restricted by not controlling for menstrual cycle. There have been no studies to our knowledge to assess pennation angle changes through the menstrual cycle. New knowledge in how the lower limbs work together in the presence of bilateral asymmetry will allow for the development of new rehabilitation programs to reduce asymmetry and potentially prevent injury.

Purpose of this study

The purpose of this study is to assess changes in lower limb muscular architecture (pennation angle (PA)) and functional parameters, such as: strength, mobility, lean mass, fat mass, in females during the luteal, ovulatory and follicular phases of the menstrual cycle. A secondary purpose of this study is to examine differences in the same physiological variables between non-contraceptive users and hormonal contraceptive users.

Research Questions

1. What are the differences in hip and knee ROM across menstrual cycle phases?
2. What are the differences in body composition values across menstrual cycle phases?
3. What are the differences in force production (FP) across menstrual cycle phases?

4. What are the differences in pennation angle (PA) across menstrual cycle phases?

Hypotheses

#1 H_0: There will not be a difference in hip and knee ROM across menstrual cycle phases.

#1 H_1: There will be a difference in hip and knee ROM across menstrual cycle phases.

#2 H_0: There will not be a difference in body composition values across menstrual cycle phases.

#2 H_1: There will be a difference in body composition values across menstrual cycle phases.

#3 H_0: There will not be a difference in force production across menstrual cycle phases.

#3 H_1: There will be a difference in force production across menstrual cycle phases.

#4 H_0: There will not be a difference in pennation angle across menstrual cycle phases.

#4 H_1: There will be a difference in in pennation angle PA across menstrual cycle phases.

Limitations

1. Results of this study will not apply to the entire population.
2. Results of this study will not include females who have irregular menses, metrorrhagia, or any birth control that doesn't allow for a monthly menstrual cycle.
3. Subjects will be recruited from the Norman and Oklahoma City areas.

Delimitations

1. Healthy participants between the ages of 18-35.

2. Subjects without metabolic, respiratory or cardiovascular diseases.

3. Healthy participants free of any musculoskeletal injury.

4. Healthy participants who have participated in fitness training ≥ 2-3 times a week for at least 2 months prior to testing.

Assumptions

1. All participants will provide accurate medical information and health history.

2. All participants will be assumed to be honest when filling out questionnaires.

3. All participants will be assumed to have followed pre-testing guidelines before coming in for testing.

4. All participants will be assumed to have exerted maximal effort in all MVC attempts.

5. Participants will be assessed during the follicular, ovulatory and luteal phases of their menstrual cycle.

Operational definitions

1. **Aponeurosis:** extension of the tension into the muscle, to which the muscle fascicle attach.[6]
2. **Muscle thickness:** the area between the aponeuroses of the muscle.[6]
3. **Pennation angle:** angle of pull of muscle fascicles relative to the tendon; the internal angle composed of the fascicle and deep aponeurosis.[6]
4. **Fascicle length:** the distance between the intersection composed of the superficial aponeurosis and fascicle and the intersection composed of the deep aponeurosis and the fascicle.[6]
5. **Follicular Phase:** Days 1-9 of the menstrual cycle, which includes menses and relatively lower levels of estradiol and progesterone.[9]
6. **Ovulation Phase:** Days 10-14 of the menstrual cycle, which includes ovulation and highest levels of estradiol for the cycle.[9]
7. **Luteal Phase:** Days 15-30 of the menstrual cycle, which includes the remaining days of the menstrual cycle and highest levels of progesterone for the cycle.[9]
8. **Force production (FP):** the product of mass and acceleration.[10]
9. **Isometric contraction:** a muscle action in which the muscle length does not change because the contractile force is equal to the resistive force.[10]
10. **Asymmetry:** ratios between the left and right side of the body, along with the lengths, circumferences, and shapes of the human musculature.[11]
11. **Strength Asymmetry:** is the deviation of strength between muscle groups and is often referred to as muscular imbalance.[11]

12. **Dual-Energy X-Ray Absorptiometry (DXA):** Measures the differential attenuation (absorption) by bone and soft tissue of the transmitted x-rays at two energy levels (high and low); Measures 3-C of body composition, bone mineral, fat, and fat-free soft tissue.[12]

13. **Ultrasound:** Ultrasound beam is propagated through skin and partially reflected with B-mode (brightness-modulation) instruments which use high-frequency sound waves (1-10 MHz).[13]

CHAPTER II

REVIEW OF LITERATURE

Menstrual Cycle Physiology

The mensural cycle is a very complex process that involves various organs (i.e., hypothalamus, anterior pituitary and ovaries) and hormones (i.e. estrogen and progesterone) that prepares the oocyte for release and the uterus for potential pregnancy.[14] The menstrual cycle can range between 21 days to 35 days, but averages 28 days for most women.[3,5,14,15] There are three main phases within a woman's menstrual cycle: follicular, ovulatory and luteal. Each phase is regulated by some type of feedback mechanism, which either positively or negatively affects hormones production.[14–16] The following section will discuss the physiology of the female menstrual cycle.

Gonadotrophin releasing hormone (GnRH) is secreted in a pulsatile manner by the hypothalamus.[14–16] GnRH is then transported to the anterior pituitary, where its 7-transmembrane G-protein receptor is activated. The activation of its 7-transmembrane G-protein signals to secrete follicle stimulating hormone (FSH) and luteinizing hormone (LH).[16] FSH and LH then provides feedback to the ovaries to alter hormone production.[16] There are two types of cells in the ovarian follicle involved in hormone production: theca cells and granulosa cells. LH stimulates the production of theca cells.[16] Theca cells simulate the production of progesterone and androstenedione by activating the enzyme, cholesterol desmolase.[16] After the secretion of androstenedione, androstenedione molecules are diffused to nearby granulosa cells. At the granulosa cells, FSH converts androstenedione to testosterone then 17-beta-estradiol by activating the enzyme, aromatase.[16] Additionally, the granulose cells also produce inhibin and activin, which aid

in inhibiting and stimulating the amount of FSH and LH the anterior pituitary produces.[14,16]

Negative feedback to the anterior pituitary is seen during both the follicular and luteal phases of the menstrual cycle, which lowers the amount of FSH and LH being produced. This negative feedback subsequently decreases the levels of 17-beta-estradiol and progesterone produced. Once a critical amount of 17-beta-estradiol is reached, positive feedback to the anterior pituitary can occur[16]. Positive feedback is seen only during the ovulatory phase of the menstrual cycle, which increases the amount of FSH and LH being produced. This regulation is very complicated and has high variability between women.[14,17]

Body Composition and Range of Motion

A study by Luiz da Silda Texeira et al. examined body composition (body mass, height and BMI) as well as sit-and-reach flexibility measurements in the follicular and luteal phases of the menstrual cycle.[5] They had a control group who had no hormone contraceptive (n=20) and an experimental group who has used hormone contraceptives for a least 2 months (n=24). The follicular phase was defined as the first day of menstruation, the ovulatory phase was defined between days 10 and 14 and the luteal phase was defined from the 15th day and lasts until the end of the cycle. No significant body composition differences were found between or within the control or experimental group. This may be due to the error that is associated with 2-component models of body composition, specifically BMI for any indication of fat percentage.[18] Fat mass was estimated through the Siri equation, which is not always specific to populations, such as women and ethnicity. Flexibility was also not different between or within groups.[5] These results in

flexibility may be due to the lack of controlling for pre-testing conditions, such as exercising prior to testing. In addition, the experimental group was only required to be using oral contraceptives for two cycles or two months, where as other studies have required three-four months to allow stabilization of hormonal contraceptives.[19,20] Podfigurna-Stopa et al. assessed 27 young women (21.8±3.9 yrs) diagnosed with functional hypothalamic amenorrhea and age/gender matched controls.[20] Functional hypothalamic amenorrhea (FHA) refers to weight loss-related amenorrhea. Body and skeletal composition was assessed from a DXA scan. Although not significantly different, the FHA group had reduced total body fat mass (TBFM), total body lean mass (TBLM), bone mineral density (BMD) and bone mineral content (BMC). This study suggests that body composition is affected, even if not significantly, by the absence of a menstrual cycle.[20]

Force Production

The female reproductive hormones which fluctuate throughout the menstrual cycle, are known to influence numerous cardiovascular, respiratory, thermoregulatory and metabolic parameters, which may affect exercise physiology and performance.[21] A study by Ross et al. assessed eight female soccer player in an endurance test, counter movement jump and sprint test in the early follicular phase (FP) and mid luteal phase (LP) of the menstrual cycle.[21] The endurance test was lower during the mid LP (2833±896 m) as compared to the early FP (3288±800 m), although it was not significant. Menstrual cycle changes have been seen to not impact maximal intensity whole body sprinting when assessed in eight female athletes who had not been on oral contraceptive for four months prior.[19] A study by Janse de Jonge assessed the influence of menstrual cycle phase on

skeletal contractile characteristics in 19 women who had not been on oral contraceptive for 6 months prior.[3] Oestrogen, progesterone, FSH and LH were measured to assess menstrual cycle phase (follicular, ovulatory, and luteal). Isometric quadricep strength, fatigability and electrically stimulated contractile properties and fatigability were also assessed, as well as handgrip strength. No significant differences were found between any of the phases of the menstrual cycle in isometric quadricep or handgrip strength. The author suggests hormone fluctuations hormone concentrations throughout the menstrual cycle do not affect muscle contractile characteristics, although it was limited by only assessing two muscle groups.[3] The study was also limited by a small sample size.

The menstrual cycle has shown fluctuation in performance and functional parameters, but due to the limitations of controlling for hormone contraceptive use and the lack of assessing other lower musculature, there is more research needed to understand what variables are affected. Women have a 4-8-fold greater risk of anterior cruciate ligament (ACL) tear than men do, especially during the pre-ovulation stage of their menstrual cycl.[7] Lefevre et al. conducted a prospective study to define the amount of ACL tears according to menstrual cycle in female recreational skiers who had sustained an ACL tear during skiing.[7] Menstrual cycle phases and oral contraceptive use were determined by a questionnaire. Women who were post-menopausal and women who had irregular cycles (>30 days) were excluded from the study. The following phases were noted at the time of the ACL tear in one hundred and seventy-two participants: follicular phase (n=58, 33.72%), ovulatory phase (n=63,36.63%), and luteal phase (n=51, 29.65%). There was a significant difference in distribution found ($\chi 2$=48.32; P=0.00001). Of the participants, 30.8% were taking oral contraceptives. Regardless oral contraceptives use, ACL tears

were 2.4-fold more frequent in pre-ovulatory than post-ovulatory phase (85/119 (71.4%) vs. 36/53 (67.9%); p=0.64). ACL tears in women are more frequent in the pre-ovulatory phases (follicular and ovulatory) than post-ovulatory phase (luteal). However, this study did not find any significant effects of birth control on ACL tear risk.[7] Another study by Slauterbeck et al. observed that in a cohort of 38 female athletes a significant amount of ACL tears occurred in the first two days of menses.[2] In conclusion, research suggests that ACL tears are occurring more often in the follicular and ovulatory phases. ACL tears also have associated changes in muscular architecture, such as shorter fascicle lengths.[22] Due to the variability in the literature, investigation is still needed for what measurements are altered during a female's menstrual cycle. We will further discuss muscle architecture furthermore in the next section.

Muscle Architecture

Some muscle fibers are parallel to the muscle aponeurosis, which is the extension of the tension into the muscle, to which the muscle fascicle attach.[6] Other muscle fibers are angled which allows for greater amount of the fiber to exert on the same length of aponeurosis, which is referred to as pennation angle (PA).[23,24] PA is most accurately measured through muscle dissection using a goniometer, but can also be measured in a non-invasive way using an imaging technique, such as ultrasound.[23–28]

The quadriceps and hamstrings are two primary muscle groups responsible for hip and knee actions, especially for ambulation, athletic performance and activities of daily living (ADLs). A better understanding of muscle architecture (PA) and function of the quadriceps and hamstring muscles could allow us to improve performance and reduce the risk for lower body injury. Studies have shown that muscular imbalance between the

quadriceps and hamstring muscles can lead to an increased risk of injuries. However, there is very little research regarding the amounts of muscular imbalance in each of these muscle groups and none that we found that address menstrual cycle potential interactions.[11,29] The more we learn about the relationship between muscle architecture and muscle performance and the role of female sex hormone fluctuations play on these variables can allow us to better educate practitioners and coaches to better prescribe exercise and also improve research studies for females in the future.

The quadricep muscles include the vastus lateralis (VL), vastus intermedius (VI), vastus medialis (VM) and rectus femoris (RF).[26] Blazevich et al. completed a study in sixteen women (19.9±3.1 years, 170.0±0.04 m, 64.6±7.9 kg) and 15 men (20.6 ± 2.6 yrs, 1.80±0.09 m, 76.0±13.0 kg), all participants were non-resistance trained.[26] The aim of the study was to determine correlations between knee extension strength and quadriceps architecture, assumption of a similarity in the mean structure of the combined VL, VI, VM and RF was created.[26] Pennation angle (PA), muscle thickness (MT) and fascicle length (FL) of the all four quadricep muscles were examined using ultrasound. Subjects were testing in a supine position with their knees bent and supported at a 45˚ angle. Three images of each muscle (VL, VM, RF, and VI) were collected at distal, middle and proximal portions of the muscle with VI being examined in two portions, anterior and lateral. The PA of each muscle was measured from approximately 3-4 cm from the deep aponeurosis to the center point of the deep and superficial aponeuroses to avoid error from curvature of fascicles as they neared the deep and superficial aponeuroses. MT was determined to be the average of the distance between the aponeuroses at the three measurement sites in each muscle and FL was estimated using PA and MT.

Significant correlation was found for within-muscle architecture for the VL for muscle thickness at each site and for PA between proximal and middle ($r = 0.48$, $P < 0.01$) and proximal and distal ($r = 0.48$, $P < 0.01$) sites. VM displayed a significant correlation for PA between distal and proximal sites ($r = 0.41$, $P < 0.05$) as well as MT at proximal and middle sites ($r = 0.57$, $P < 0.01$). RF displayed a significant correlation for PA between middle and distal sites ($r = 0.38$, $P < 0.05$) and MT for all sites ($0.56 < r < 0.74$, $P = 0.000–0.002$). The anterior portion of the VI displayed a significant PA correlation for the middle and distal sites ($r = 0.47$, $P < 0.05$) and for MT between the proximal and middle ($r = 0.52$, $P < 0.01$) and proximal and distal sites ($r = 0.38$, $P < 0.05$). The lateral portion of the VI displayed a significant correlation for PA between the proximal and distal sites ($r = 0.71$, $P < 0.001$) and no significant correlations for MT. A difference index (provides quantitative assessment of intermuscular between the mean architecture of the three superficial quadriceps muscles) was calculated to compare overall muscle structure, or architectural similarity, and revealed a low difference index for the VL, VM, and RF muscles but not for either portion of the VI. Trends between muscles were calculated using z-scores to provide a parameter for the entire quadriceps group. It was determined higher angles in the VM were indicative of higher angles in the VL and RF. A mirrored trend was also seen for individuals with a larger RF PA having a larger PA in their VL and VM. These trends were not seen in the VI when compared to other muscles.[26]

MT of one muscle was not an accurate determinant of MT in other muscles except between VL and the anterior portion of the VI. Regression equations were used to determine each variables contribution of whole muscle architecture. It was found the MT of the VM and the PA of the VM were the two best predictors of whole muscle

architecture. When looking at the interaction of muscle parameters the VL, VM, and portions of the VI showed significant correlations between MT and PA, but not for RF. This study suggests that the quadriceps muscles (VL, VI, VM, RF) have similar architecture, which indicates assessment of one muscle can help infer to the structure of the other muscles.[26] PA has been shown to be a strong indicator of MT[26]. Since the VM is a strong predictor of the whole muscle architecture, it was assumed that measuring the PA of the VM and the VI will provide a sense of the whole quadriceps muscle group. This assumption is necessary in order to investigate correlations between muscle architecture of the quadriceps and performance of the muscle group as a whole when it is not feasible to measure the MT and PA of each muscle individually.[26]

The hamstring muscles include the semitendinosus (ST), semimembranosus (SM) and bicep femoris, which is usually classified as bicep femoris long-head (BFlh) and bicep femoris short-head (BFsh). Chleboun et al. tested the reliability of using ultrasound to measure BFlh PA using an Acuson 128XP real-time ultrasonography scanner (Acuson Sequoia, Acuson Corporation, CA, USA) with a 5MHz 8.0-cm transducer and through in vivo measurement of dissection 18 female recreational athletes (23.0±1.8 years).[27] This study examined ultrasound measurements at the knee and hip angles of 0, 40, and 90 degrees. Three to seven pictures were taken along the long head of the rectus femoris. In vivo measurements were made by removing entire fibers from cadavers and measuring the angle with a goniometer. The researchers found that measurements from the ultrasound were slightly less accurate than the in vivo measurements but not significantly different ($p > 0.05$, ICC = 0.87).[27] The researchers concluded that bending at the joints resulted in significant changes in PA with the highest PA coming at a 90-degree hip angle and a 0-

degree knee angle. The authors suggest ensuring maximal overlap and optimal force production capabilities, a seated position of 90° hip and 90° knee flexion should be used.[27]

A study by Baroni et al. examined PA, FL and CSA of the rectus femoris (RF) and vastus lateralis (VL) of 20 male volunteers before and after a non-training control period of 4 weeks, and post- 4, 8, and 12 weeks of isokinetic eccentric training.[30] Significant increases in RF and VL had significant changes in muscle architecture within the first 4 weeks of training. MT increased by 7–10%, fascicle length increased 17–19%, while pennation angle was unchanged.[30] The authors concluded that increased muscle thickness was not related to PA changes, but FL changes. A study by Blazevich et al. study examined changes in the muscle size, muscle architecture (muscle size, pennation angle (PA), and fascicle length (FL) of the vastus lateralis (VL) and rectus femoris (RF) muscles) strength, and sprint/jump performances of concurrently training athletes during 5 weeks of resistance training (RT).[31] There were three different programs implemented: squat-lift training, forward back squat training and sprint-jump training. They found an increase in VL in squat-lift training and forward hack squat training was statistically different to the decrease in sprint-jump training subjects ($P < 0.05$ at distal, $P < 0.1$ at proximal). VL FL increased for sprint-jump training only at the distal site ($P< 0.05$). Furthermore, MT of both the VL and RF increased significantly at the proximal sites ($p<0.05$). These studies suggest that adaptions can occur within a 4-5-week resistance training program. When assessing through a month, for example throughout the menstrual cycle, it is crucial to control for physical activity to avoid changes to the muscular architecture that may hinder results. Since there is an increased amount of water retention, which may cause excessive swelling, muscular architecture changes throughout the

menstrual cycle are possible. No studies have focused on menstrual cycle changes in muscle architecture and few have controlled for the menstrual cycle while testing muscle architecture variables, thus warranting further investigation.

In summary, the menstrual cycle has shown measurement fluctuations during specific phases. Muscle architecture, such as pennation (PA), plays a significant role in force production and dynamic exercise. Many studies have evaluated functional parameters (body composition, force production, mobility), but are restricted by not including menstrual cycle fluctuations through the follicular, ovulatory and luteal phases. Due to the inconsistency in literature results, in regarding what changes with the hormonal fluctuations of the menstrual cycle, research is needed to bridge the gaps.

CHAPTER III

METHODOLOGY

This chapter will aim to provide a thorough description of the methods used for this study, which includes participant requirements, participant inclusion and exclusion criteria, descriptions of the data collection protocols, instrumentation, and statistical analyses.

Sample

G-Power Analysis was used to determine sample size using a moderate effect (3 groups, 0.8 power, 0.3 f, $p \leq 0.005$), which revealed that nine subjects were needed for each group. Thirty-five participants consented to partake in this study; six of those participants were excluded, which is discussed in Chapter IV. Twenty-eight participants (10 non-contraceptive users, 9 oral contraceptive users and 9 males) between the ages of 18 and 35 completed this study. All subjects signed an informed consent form approved by the University of Oklahoma Institutional Review Board (Health Science Center). Individuals were recruited from the Norman and Oklahoma City area by flyers, word of mouth, and e-mail to participate.

Inclusion Criteria

Non-Contraceptive Users (NC)

1. Be in the age range of 18-35 years old.
2. Be female.
3. Have a regular menstrual cycle for the last 6 months.
4. Not-taking oral contraceptives for at least 6 months prior to testing.
5. Active in an exercise program ≥ 2-3 days a week for at least 2 months.

Oral-Contraceptive Users (OC)

1. Be in the age range of 18-35 years old.

2. Be female.

3. Have a regular menstrual cycle for the last 6 months.

4. Regularly taking oral contraceptives for at least 6 months prior to testing.

5. Active in an exercise program ≥ 2-3 days a week for at least 2 months.

Male Control Group (CON)

1. Be in the age range of 18-35 years old.

2. Be male.

3. Active in an exercise program ≥ 2-3 days a week for at least 2 months.

Exclusion Criteria

Non-Contraceptive Users (NC)

1. Pregnant women will be excluded from the study.

2. Outside the age range of 18-35 years.

3. Not being female.

4. Individual has a prior injury, which limits knee range of motion.

5. Individuals who have irregular menses, metrorrhagia, or any birth control that doesn't allow for menses.

6. Individuals unable to perform a knee maximal voluntary contraction.

7. Individual has undergone surgery that may alter muscle architecture of the quadriceps and/or hamstrings.

8. Individuals with cardiovascular diseases.

9. Individuals with neurological diseases or damage.

10. Individuals have metal implants in the lower limbs that would impact body composition assessments.

Oral-Contraceptive Users (OC)

1.Pregnant women will be excluded from the study.

2. Outside the age range of 18-35 years.

3. Not being female.

4. Individual has a prior injury, which limits knee range of motion.

5. Individuals who have irregular menses, metrorrhagia, or any birth control that doesn't allow for menses.

6. Individuals unable to perform a knee maximal voluntary contraction.

7. Individual has undergone surgery that may alter muscle architecture of the quadriceps and/or hamstrings.

8. Individuals with cardiovascular diseases.

9. Individuals with neurological diseases or damage.

10. Individuals have metal implants in the lower limbs that would impact body composition assessments.

Male Control Group (CON)

1. Outside the age range of 18-35 years.

2. Not being male.

3. Individual has a prior injury, which limits knee range of motion.

4. Individuals unable to perform a knee maximal voluntary contraction.

5. Individual has undergone surgery that may alter muscle architecture of the quadriceps and/or hamstrings.

6. Individuals with cardiovascular diseases.

7. Individuals with neurological diseases or damage.

8. Individuals have metal implants in the lower limbs that would impact body composition assessments.

Research Design

This study utilized a short-term longitudinal design with repeated measures. There were three groups: Non-contraceptive users (NC), Oral-contraceptive users (OC) and a male control group (CON). The NC group was required to not be on any contraceptives (hormonal or non-hormonal) for at least six months prior to testing. The OC group was required to be taking hormonal oral contraceptives for at least six months prior to testing. Medication use was assessed through the Medical History Questionnaire to ensure no hormonal medication was being used by the NC and CON group. Participants were requested to avoid any lower body resistance training or endurance training 24 hours prior to testing and to continue exercise as usual throughout the duration of the study. Testing protocols during each visit were the same for all participants. Visit 1 began with an explanation of inclusion and exclusion criteria and a brief explanation of the protocols included in the study. Each participant had the study explained to them and sign an

informed consent. Participants also completed a physical activity readiness questionnaire (PAR-Q), menstrual history questionnaire (if applicable), Health Insurance Portability and Accountability Act (HIPAA) form and an International Physical Activity Questionnaire (IPAQ). Height, weight, resting blood pressure (BP), resting heart rate (HR) were assessed at the beginning of each visit. Visit 1 concluded with a familiarization of the maximal voluntary isometric contraction muscle protocol (MVIC) for knee extension and flexion, and range of motion of the hip and knee. All testing and familiarization were performed on the participant's dominant leg.

Visits 2-4 were scheduled based off of the participant's menstrual cycle phase. The luteal (days 1-4), ovulatory (days 12-15) and follicular (days 18-21) phases were defined visits based off of menstrual cycle questionnaires (if necessary). The control group (CON) followed a similar timeline that started as an estimated Day 1 (follicular phase). This was estimated, since the males recruited for the control group do not have a menstrual cycle to compare to, but we estimated the timing used for our female groups to control for any training effect that may occur. All measurements were taken by the same researcher and sites were standardized to control for placement and angles used through testing for each participant.

Visits 2-4 began by measuring weight, resting blood pressure, resting heart rate, hip and knee range mobility, hydration status and a pregnancy test (if necessary). Mobility measurements were taken on an observation table with a goniometer in the following order: hip flexion, knee flexion, knee extension, hip extension. After mobility measurements, the distance of the femur was assessed with a measuring tape between the superior anterior iliac spine (main bony landmark on the hip) and the superior border of

the patella (top of the knee cap). Marks were made at 22%, 56%, and 73% of the thigh to allow for consistent measurements between visits. Following initial assessments, a body composition assessment was done via Dual-Energy X-Ray Absorptiometry (DXA) scan. The DXA scan assessed the absorption of bone and soft tissue of the transmitted x-rays at two energy levels. Radiation exposure was minimal, but pregnancy tests were taken for all female participants. Pennation angle (PA) measurements of the quadriceps muscles (vastus medialis (VM), vastus lateralis (VL), rectus femoris (RF), and the anterior portion of the vastus intermedius (VI)) of the dominant leg were performed using B-mode ultrasound. PA is the angle in which the muscle fascicle extends from the deep aponeurosis (the extension of the tendon joining the muscle). Ultrasound measurements were followed by an isometric force production protocol. Assessment of knee extension and flexion was done in the dominant leg with a force dynamometer. Legs were assessed in a random order (knee extension/flexion). Three warm-ups and three MVICs were taken for each knee extension and flexion.

Initial Assessment:

During the initial assessment, weight, blood pressure and heart rate were taken to assess for any potential adverse reactions associated with hypertension (high blood pressure) or any other heart disorders. During this initial assessment femur length (largest bone going from the hip to knee) was measured to prepare for ultrasound measurements for muscle architecture assessment. Marks were made at 22%, 56%, and 72% of the thigh to allow for consistent measurements between visits.

Range of Motion (ROM) assessment:

During ROM assessment, hip and knee range of motion (ROM) were assessed. The dominant leg was assessed in the following order: hip flexion, knee flexion, knee extension and hip extension. All measurements were made on an observation table (massage table) and subjects were instructed to relax completely. Extension and flexion were assessed for the dominant hips and knee with a goniometer (Lafayette Instrument Company, Lafayette, IN, USA). Each measurement was assessed passively, meaning the maximum mobility that the participant can do with assistance from the researcher, by pushing the joints together.

For passive measurements of hip flexion, subjects started supine with their pelvis in a neutral position with the knees extended on a table. The fulcrum of the goniometer was placed over the greater trochanter of the femur, the proximal arm was aligned with the lateral midline of the pelvis and the distal arm was aligned with the lateral midline of the femur (lateral epicondyle for reference). Hip flexion took place by firmly applying pressure to lift the thigh off the table, allowing the knee to flex. Stabilization of the pelvis was maintained to avoid posterior tilt or rotation of the pelvis.[32,33]

For passive measurements of knee flexion, subjects started supine with their pelvis in a neutral position with the knees extended on a table. The fulcrum of the goniometer was placed over the lateral epicondyle of the femur, the proximal arm was aligned with the lateral midline of the femur (greater trochanter for reference) and the distal arm was aligned with the lateral midline of the fibula (lateral malleolus for reference). Knee flexion took place by firmly applying pressure above the knee and below to knee allowing the

knee to flex at a hip flexion of 90 degrees. Stabilization of the femur was maintained to prevent hip rotation, abduction or adduction.[32,33]

For passive measurements of knee extension, subjects started supine with their pelvis in a neutral position with the knees extended on a table. A folded towel was placed under the ankle to ensure that knee is in full flexion. The fulcrum of the goniometer was placed over the lateral epicondyle of the femur, the proximal arm was aligned with the lateral midline of the femur (greater trochanter for reference) and the distal arm was aligned with the lateral midline of the fibula (lateral malleolus for reference). Knee extension took place by applying pressure on the lower thigh and exerting a slight downward pressure. Stabilization of the femur was maintained to prevent hip rotation, abduction or adduction. [32,33]

For passive measurements of hip extension, subjects started prone with their pelvis in a neutral position with the knees extended on a table. The fulcrum of the goniometer was placed over the greater trochanter of the femur, the proximal arm was aligned with the lateral midline of the pelvis and the distal arm was aligned with the lateral midline of the femur (lateral epicondyle for reference). Hip extension took place by firmly applying pressure above the knee to raise the thigh off the table. Stabilization of the pelvis was maintained to avoid posterior tilt or rotation of the pelvis. [32,33]

Body Composition Assessment

Body composition was measured using a whole-body Lunar Dual-energy X-ray absorptiometry (DXA) scanner (with software version 13.60.033, GE-Lunar Prodigy Advanced, Madison, WI). The DXA scanner was calibrated each day prior to data collection. Subjects removed their shoes, jewelry, and any clothing or personal items that

may contain metal prior to scan. Prior to scanning, urine specific gravity (U.S.G.) was assessed and must be between the range of 1.005-1.030. If USG measurements were outside of the range, subjects were given water and reassessed 30 minutes later or rescheduled for another day. All females had pregnancy test prior to DXA scans. No participants became pregnant at any time during the study, so none were disqualified from the study due to fetal adverse effects related to radiation in DXA scans. After clearing the USG levels and pregnancy test, subjects were positioned in a supine position with the middle of the table aligned with the middle (sagittal plane) of their body. Participants were asked to place their arms at their side within the measurement zone, hands pronated perpendicular to the table, leaving space between their arms and your sides. Straps were used to secure feet to limit movement during the scan. Following the scan, regions of interest were placed around each leg to examine composition of each leg individually. DXA scans were assessed three times for each participant to distinguish body composition differences seen with the menstrual cycle in all three groups: OC, NC, and CON and all three time points.

Muscle architecture assessment

Muscle pennation angle (PA) was collected using a LOGIQ S8 ultrasound apparatus (GE Healthcare, Little Chalfont, United Kingdom). All measurements were made with the probe angled perpendicular to the leg and parallel to the muscle such that an imaginary line extending out from the probe would go straight through the muscle roughly in the sagittal plane of the body. Prior to measurements a water-soluble gel was applied to the area being measured to maximize acoustic perfusion into the muscle, thus minimizing the amount of pressure applied to obtain a clear image of muscle fascicles.

Three images were taken of the following muscles; Vastus medals (VM), vastus lateralis (VL), rectus femoris (RF), and vastus intermedius (VIA) on the dominant leg. All images were transferred to a USB drive to further analyzed by an on-screen protractor (MB-Ruler 5.3, MB-Software Solutions, Iffezheim, Germany).

The measurement process began by locating the anterior superior iliac spine and the superior border of the patella as described in the initial measurements section above. The distance between these two landmarks was considered the subject's thigh length and measurement locations were oriented based on a straight-line measurement location between the two. To locate the proximal end of the vastus intermedius anterior (VIA) and the rectus femoris (RF) we used a marker to indicate the area lying 73% of thigh length distal from the anterior superior iliac spine along the measurement location. To locate the proximal end of the vastus lateralis (VL) we used a marker to indicate the area lying 56% of thigh length distal from the anterior superior iliac spine along the measurement location. To locate the proximal end of the vastus intermedius (VM) we used a marker to indicate the area lying 22% of thigh length distal from the anterior superior iliac spine along the measurement location. Any proximal, distal, medial, or medially or laterally deviation from these points required to find an accurate PA measurement were noted.

For quadriceps assessment, subjects were seated with both hips and knees at a 90˚ angle. The back, knees, and feet were adjusted and supported to ensure those angles could be maintained with the entire lower body completely relaxed. We measured PA of the muscles in this order: dominant leg (VM, VL, VIA, RF).

Force Production Assessment

Familiarization for maximal voluntary isometric contractions (MVICs) for both knee extension and knee flexion consisted of having subjects sit on the KinCom dynamometer (KinCom model: KC125AP, Isokinetic International, East Ridge, TN 37412) and adjusting the seat until the knee and hip angles were both 90˚. The KinCom was adjusted so the rotational axis of the dynamometer head aligns with the knee. Seat and dynamometer head position were recorded for future visits. Straps were fastened to secure the upper and lower body to the seat, to ensure that quadriceps and hamstring muscles were isolated. The dominant ankle was strapped to the load cell of the KinCom.

Subjects were asked to perform isometric knee extensions at perceived efforts of 25%, 50%, and 75% until they felt comfortable with the strength protocol. Participants were asked to perform three MVICs at full (maximal) effort. The same process was completed for isometric knee flexion.

Statistical Methods

Sigma Plot (Systat Software, San Jose, CA v. 12.5) was used to perform statistical analysis. Statistical significance was set at $p < 0.05$.

3-Way Repeated-measures ANOVA was used to identify differences between quadriceps muscle architecture, body composition, mobility and force production during the follicular, ovulatory and luteal phases of the menstrual cycle between groups (OC, NC, CON) and time (follicular, ovulatory, and luteal phases). If significant main effects were observed, pairwise comparisons using a Bonferroni Post-Hoc were completed.

If no interaction or main effect was observed, a 2-Way Repeated Measures ANOVA was run on female group (OC, NC) and time (follicular, ovulatory and luteal) to

see if there were differences between the female groups. If significant main effects were observed, pairwise comparisons using a Bonferroni Post-Hoc were completed. Cohen's d values were calculated for all female data in the OC and NC groups. The highest score were used and data is displayed as mean±SD. Cohen's d values (d) effect sizes were deemed as: large (0.8), medium (0.5) or small (0.2-0.3).[34] Raw and absolute % difference values were found for two time points: follicular to ovulatory phases (F-O) and ovulatory to luteal phases (O-L) as well as Cohen's d values for all female data in the OC and NC groups.

If no interaction or main effect was observed through 2 x 3 Repeated Measures ANOVA, 1-Way ANOVA was used to assess female characteristics of both the NC and OC groups collapsed (n=19). Significant findings were followed by pairwise comparisons using a Bonferroni Post-Hoc. Partial eta^2 (η^2) were calculated for all collapsed female data.[34] Partial eta^2 (η^2) effect sizes were deemed as: large (0.14), medium (0.06) or small (0.01).[34]

If no interaction or main effect was observed, a 2-Way Repeated Measures ANOVA was run on group (males, females) and time (follicular, ovulatory and luteal) to see if there were differences between gender. If significant main effects were observed, pairwise comparisons using a Bonferroni Post-Hoc were completed.

CHAPTER IV

RESULTS

The first goal of this project was to assess changes in lower limb muscular architecture (pennation angle (PA)) and functional parameters, such as: strength, mobility, whole body lean mass, whole body fat mass, in females during the luteal, ovulatory and follicular phases of the menstrual cycle. The second goal was to examine differences in physiological variables between non-contraceptive users and hormonal contraceptive users. This chapter will discuss the results for the study which included: subject characteristics, group differences, and menstrual cycle changes in ROM, body composition, force production and muscle architecture.

SUBJECT CHARACTERISTICS

Thirty-four participants were recruited for this study. There were six participants who consented that were unable to finish their visits due to: knee extension measurements over the maximal value (n=1), incorporating a diet or using weight-loss medication during the study (n=2) and moving outside traveling distance and/or no reply to scheduling visits (n=3). Nine CON males, 10 OC females and 9 NC females completed the entirety of the study results in 28 participants total. All participants were active in a resistance exercise program at least 2-3 days a week for at least 6 weeks prior to their first visit. All participants were deemed Health Enhancing Physical Activity (HEPA) active by IPAQ analysis, which is defined by (a) vigorous-intensity activity on at least 3 days achieving a minimum of at least 1500 MET-minutes/week or (b) 7 or more days of any combination of walking, moderate-intensity or vigorous intensity activities achieving a minimum of at least 3000 MET-minuets/week. Participants were all asked to continue the same exercise

and nutritional regimen as normal throughout the study. Both female groups (OC and NC) had a regular menstrual cycle every month for the last 6 months. OC individuals had been taking hormonal, oral contraceptives for at least 6 months prior to their first visit. All NC individuals had not taken any contraceptive (hormonal or non-hormonal) the past 6 months. All participants were asked to refrain from lower body resistance exercise for at least 24 hours prior to testing.

A One-Way ANOVA was used to assess differences in group characteristics, which revealed that male CON participants were significantly taller ($p < 0.001$ for OC and NC) and weighed more ($p < 0.001$ for OC and NC) when compared to female participants in both OC and NC groups. Age was not significantly different for any of the groups ($p > 0.05$). The OC and NC were not significantly different for age, height and weight ($p > 0.05$). Menstrual cycle length was an average of 27.75 ± 1.5 for combined groups (OC, NC). Group characteristics are displayed in Table 1.

Table 1: Subject Characteristics (Mean ± SD)

	Groups		
Variables	**CON (n = 9)**	**OC (n = 10)**	**NC (n= 9)**
Age (years)	25.2 ± 2.53	23.2 ± 2.5	23.65 ± 4.3
Height (in)	70.7 ± 2.78*	64.81 ± 2.62	64.06 ± 2.71
Weight (lb)	191.9 ± 21.11*	137.2 ± 17.14	144.82 ± 40.5
Cycle Length (days)	N/A	27.5 ± 1.0	28.0 ± 2.0

Note: Differences if present were denoted using *($p<0.05$). Standard deviations represent variability. CON: male controls, OC: oral-contraceptive users, NC: non-contraceptive users.

GROUP DIFFERENCES

- **Groups: 3 x 3 (CON, OC, NC)**
    - 3-Way Repeated-measures ANOVA was used to identify differences between quadriceps muscle architecture, body composition, mobility and force production between groups (OC, NC, CON) and time (follicular, ovulatory, and luteal phases).
- **Contraceptive Use 2 x 3 (OC, NC):**
    - 2-Way Repeated Measures AN OVA was run on female group (OC, NC) and time (follicular, ovulatory and luteal) to see if there were differences between the female groups across the menstrual cycle.
- **Females: 1 x 3 (Collapsed Females)**
    - 1-Way ANOVA was used to assess female characteristics of both the NC and OC groups collapsed (n=19) across the menstrual cycle.
- **Gender: 2 x 3 (Males, Collapsed Females)**
    - 2-Way Repeated Measures ANOVA was run on gender (female, male) and time (follicular, ovulatory and luteal) to see if there were differences between gender across the menstrual cycle.

RANGE OF MOTION

3 x 3 (CON, OC, NC)

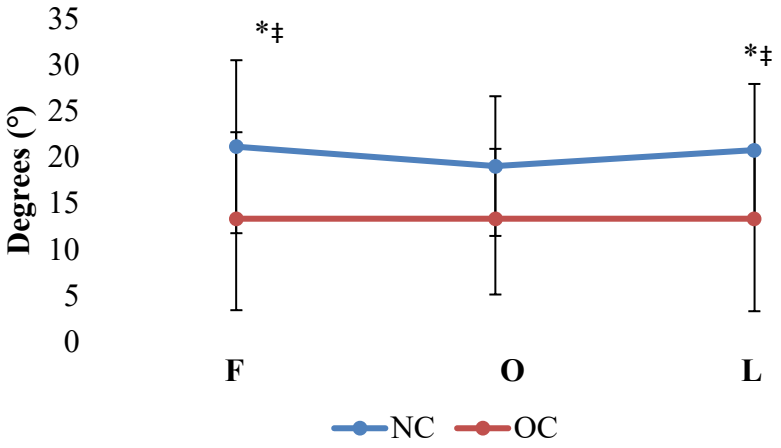
The 3 x 3 Repeated Measures ANOVA reveled no significant group main effect for hip extension ($p=0.07$), hip flexion ($p=0.09$), knee extension ($p=0.79$) or knee flexion ($p =0.07$). There also was no significant time main effect found for hip extension ($p=0.8$),

hip flexion (p=0.81), knee extension (p=0.57) or knee flexion (p=0.15). Likewise, no group x time interaction was found for hip extension (p=0.22), hip flexion (p=0.53), knee extension (p=0.22) or knee flexion (p=0.58). Results are shown in Table 2. η^2 showed small effect sizes for all group ROM measurements.

2 x 3 (OC, NC)

The non-significant 3 x 3 Repeated Measures ANOVA was then followed by the 2 (OC, NC) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA to further investigate changes within the females to compare contractive use. There was a significant group main effect for hip extension with the NC group displaying significant higher hip extension than the OC group (p=0.049), shown in figure 1. There was no group main effect for hip flexion (p=0.835), knee extension (p=0.87) or knee flexion (p=0.59). There was no time main effect for hip extension (p=0.567), hip flexion (p=0.54), knee extension (p=0.496) or knee flexion (p=0.09). There was a group x time interaction where hip extension in the NC group was significantly higher in both the follicular and luteal phases when compared to the OC group ($p<0.05$ for both). There was no group x time interaction for hip flexion (p=0.739), knee extension (p=0.134) or knee flexion (p=0.49).

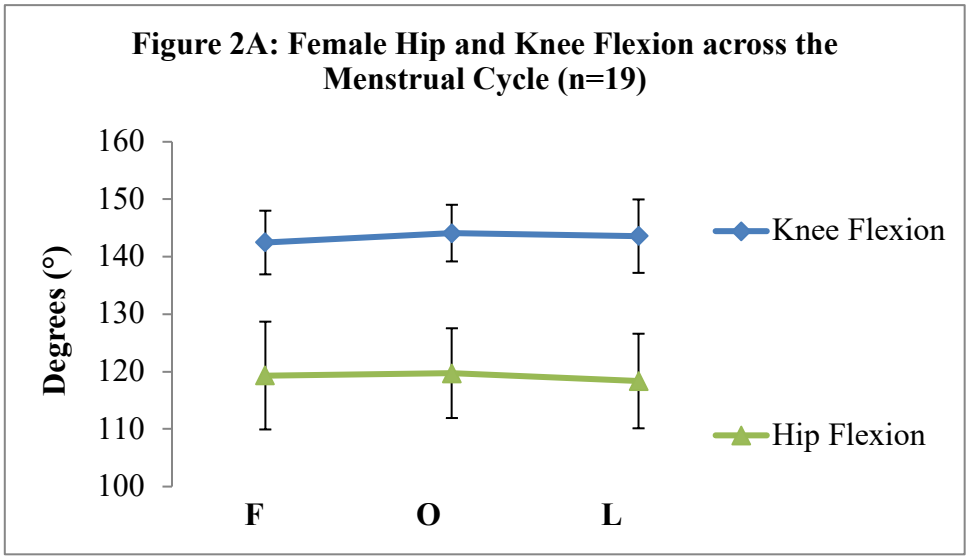
Figure 1: Passive Hip Extension during the Menstrual Cycle

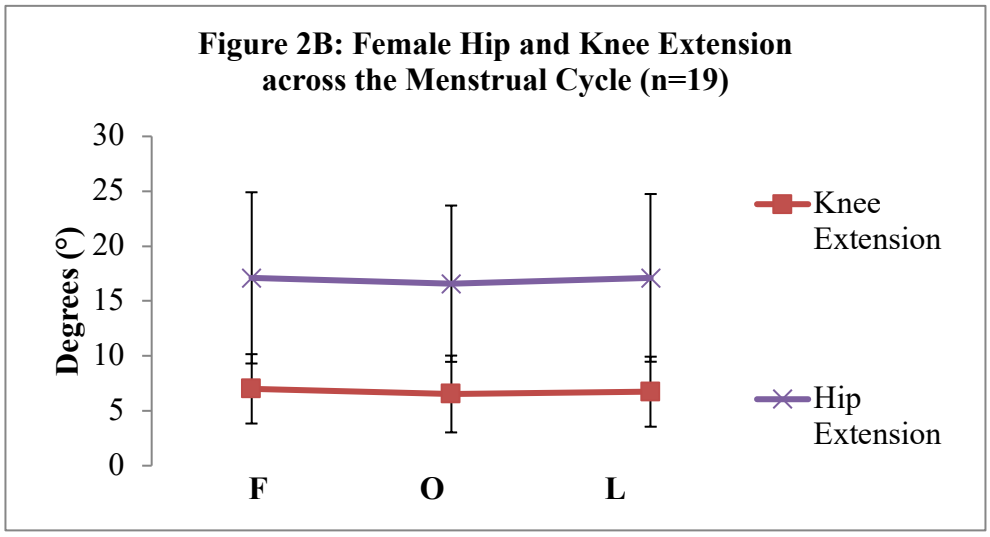


Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Group main effects shown through the 2x3 (OC vs NC) RM ANOVA were denoted using *($p<0.05$). ‡ denotes group x time interaction during the 2x3 (OC vs NC) RM ANOVA. Standard deviations represent variability.

1 x 3 (Collapsed Females)

A One-Way Repeated Measures ANOVA was used to assess female characteristics (n=19), which showed that there were no significant visit main effect (follicular, ovulatory and luteal) for hip extension ($p=0.67$), hip flexion ($p=0.55$), knee extension ($p=0.52$) or knee flexion ($p=0.09$) shown in Figures 2A and 2B.





Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Differences if present were denoted using *($p<0.05$). Standard deviations represent variability.

2 x 3 (Males, Collapsed Females)

Due to the non-significance found in the female groups, a 2 (male, female) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA was run to further investigate sex differences in ROM through the menstrual cycle. There was no group main effect for hip flexion ($p=0.34$), knee extension ($p=0.12$), hip extension ($p=0.22$) or knee

extension (p=0.51). There was no time main effect for hip extension (p=0.94), hip flexion (p=0.71), knee extension (p=0.73) or knee flexion (p=0.23). There was no group x time interaction for hip extension (p=0.73), hip flexion (p=0.27), knee extension (p=0.7) or knee flexion (p=0.42).

Table 2: ROM across the Menstrual Cycle (mean±SD)

ROM		CON (n=9)	OC (n=10)	NC (n=10)	η^2
Hip Extension (^{0})	**F:**	13.22 ± 8.14	14.6 ± 5.9	21.2 ± 7.2*‡	0.0053
	O:	13.33 ± 8.15	15.4 ± 6.5	19.1 ± 6.6*	
	L:	12.89 ± 7.52	14.9 ± 6.4	20.8 ± 6.9*‡	
Hip Flexion (^{0})	**F:**	108.1 ± 13.9	119.4 ± 9.4	120.2 ± 9.9	0.0047
	O:	109.3 ± 15.5	120.3 ± 7.6	120.2 ± 8.2	
	L:	110.7 ± 14	118.6 ± 7.2	118.2 ± 10	
Knee Flexion (^{0})	**F:**	191.9 ± 21.1	215.3 ± 55.2	240.9 ± 72.5	0.0045
	O:	191.8 ± 21.1	220.3 ± 62.5	246.2 ± 71.3	
	L:	192.4 ± 21.9	240.5 ±80	231.4 ± 56.7	
Knee Extension (^{0})	**F:**	5.9 ± 2.5	7.00 ± 3.54	7.11 ± 3.14	0.0099
	O:	5.9 ± 1.9	6.56 ± 2.92	6.67 ± 4.33	
	L:	6.1 ± 2.9	7.67 ± 3.32	6.11 ± 3.10	

Note: CON: male controls, OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory, L: luteal. η^2 was used to determine time effect size for all three groups. Group main effects shown through the 2x3 (OC vs NC) RM ANOVA were denoted using *($p<0.05$). ‡ denotes group x visit interaction during the 2x3 (OC vs NC) RM ANOVA. Standard deviations represent variability.

BODY COMPOSITION

3 x 3 (CON, OC, NC)

The 3 x 3 Repeated Measures ANOVA revealed a significant group main effect for scale weight, Bone Mineral Density (BMD), Fat-free (g), Lean mass (g) and total mass, all of which were higher in the CON group vs the OC and NC groups ($p<0.001$ for all variables). There also was a significant group effect where gynoid (% fat) was found to be

higher in the OC (p=0.02) and NC (p=0.03) groups when compared to the CON group. A/G ratio was also found to be significantly lower in the NC (p=0.003) group when compared to the CON group. Scale weight was significantly higher in the CON (192.048±9.27 lbs) than both the OC (137.68±8.79 lbs, p<0.001) and NC groups (146.026±9.27 lbs, p=0.004). There was similarly no time main effect found for Tissue (% fat) (p=0.08), Android (% fat) (p=0.62), and Gynoid (% fat) (p=0.57). A significant time effect was found in the CON group, lean mass was found to be higher during their ovulatory visit (62813±2115 g) when compared to their follicular visit (62168±2115 g) (p=0.044). Furthermore, a significant time effect was found in the CON group, total mass was found to be higher during their luteal visit (88.09±4.2 kg) when compared to their follicular visit (87.4±4.2 kg) (p=0.028). There was no time effect for scale weight (p=0.12), BMD (p=0.63), Fat-free (g) (p=0.16), tissue (%fat) (p=0.07), android % fat (p=0.14), gynoid (% fat) (p=0.57) and A/G ratio (p=0.38). No group x time interaction was found for scale weight (p=0.89), BMD (p=0.89), Fat-free (g) (p=0.57), lean mass (p=0.55) total mass (p=0.79), tissue (%fat) (p=0.51), android % fat (p=0.9), gynoid (% fat) (p=0.897) and A/G ratio (p=0.98). Results are shown in Table 3. $\eta 2$ showed small effect sizes for all group body composition measurements.

2 x 3 (OC, NC)

The non-significant 3 x 3 Repeated Measures ANOVA in the females was then followed by the 2 (OC, NC) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA to further investigate changes within the females to compare contractive use. Scale weight (p=0.61), BMD (p=0.5), fat free (g) (p=0.72), lean mass(g) (p=0.72), tissue

(% fat)(p=0.88), total mass (p=0.69), android (% fat) (p=0.44), gynoid (% fat) (p=0.91) and A/G ratio (p=0.18) had no significant group (OC, NC) main effect. Scale weight (p=0.14), BMD (p=0.22), fat free (g) (p=0.22), lean mass(g) (p=0.81), tissue (% fat)(p=0.18), total mass (p=0.27), android (% fat) (p=0.28), gynoid (% fat) (p=0.57) and A/G ratio (p=0.52) had no significant time (follicular, ovulatory, luteal) main effect. Scale weight (p=0.63), BMD (p=0.46), fat free (g) (p=0.25), lean mass(g) (p=0.9), tissue (% fat)(p=0.28), total mass (p=0.94), android (% fat) (p=0.7), gynoid (% fat) (p=0.6) and A/G ratio (p=0.86) had no significant group x time interaction.

1 x 3 (Collapsed Females)

A One-Way Repeated Measures ANOVA was used to assess female characteristics (n=19), which showed that there were no significant visit main effect (follicular, ovulatory and luteal) for scale weight (p=0.14), A/G Ratio (p=0.49), Gynoid % fat (p=0.59), Android % fat (p=0.27), Fat free mass (p=0.26), Lean (g) (p=0.81), Fat (g) (p=0.12), total mass (p=0.25), tissue % fat (p=0.2) and BMD (p=0.23).

2 x 3 (Males, Collapsed Females)

Due to the non-significance in female groups, a 2 (male, female) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA was run to further investigate sex differences in body composition across the menstrual cycle. The 2 x 3 Repeated Measures ANOVA revealed a significant group main effect for scale weight, Bone Mineral Density (BMD), Fat-free (g), Lean mass (g) and total mass, all of which were higher in males ($p<0.001$ for all variables). Tissue (% fat) was significant higher in females (32.23±1.63%) vs males (25.36±2.37%, p=0.025). This is the only difference found between the 3 x 3 RM ANOVA, which could be attributed to having a higher sample size

with the combined OC and NC groups. There also was a significant group effect where gynoid (% fat) was found to be higher in females than males (p=0.003). A/G ratio was also found to be significantly lower in the females when compared males (p=0.004). Android (% fat) had no significant group main effect (p=0.66). There was a significant time main effect in men where follicular total mass significantly increased from follicular to luteal phase (87±4.13g vs 88.09±4.13g, p=0.04). Scale weight (p=0.17), BMD (p=0.52), fat free (g) (p=0.08), lean mass(g) (p=0.2), tissue (% fat)(p=0.09), android (% fat) (p=0.17), gynoid (% fat) (p=0.68) and A/G ratio (p=0.37) had no significant time (follicular, ovulatory, luteal) main effect. Scale weight (p=0.85), BMD (p=0.65), fat free (g) (p=0.5), lean mass(g) (p=0.42), tissue (% fat)(p=0.66), total mass (p=0.45), android (% fat) (p=0.88), gynoid (% fat) (p=0.9) and A/G ratio (p=0.86) had no significant group x time interaction.

Table 3: Body Composition across the Menstrual Cycle (mean±SD)

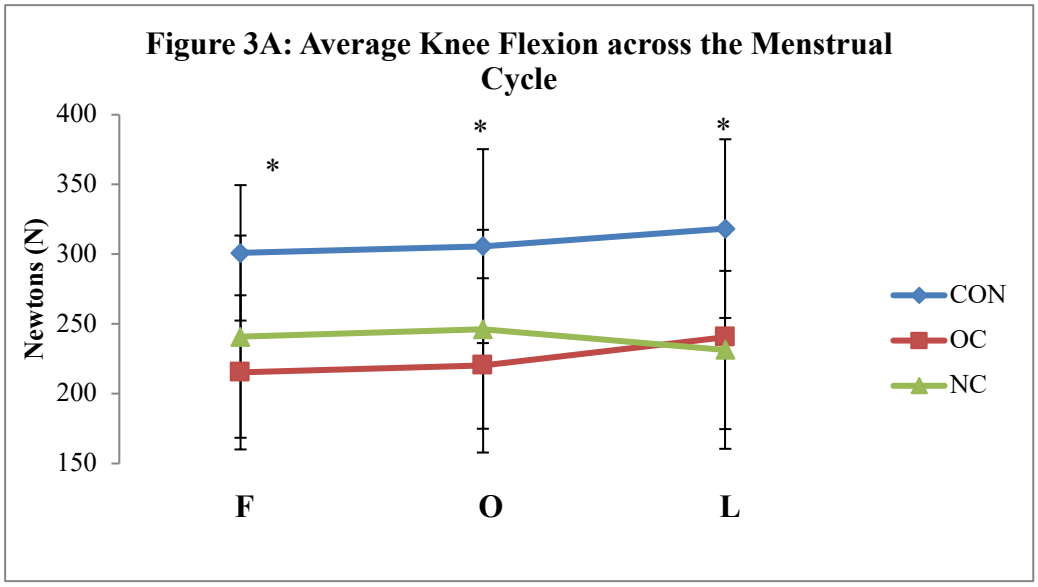
		CON n=9)	OC (n=10)	NC (n=9)	η^2
A/G Ratio	**F:**	1.11 ± 0.09	0.94 ± 0.2	0.83 ± 0.2*	0.0035
	O:	1.13 ± 0.09	0.95 ± 0.22	0.84 ± 0.19*	
	L:	1.13 ± 0.09	0.96 ± 0.21	0.84 ± 0.18*	
Android (% fat)	**F:**	34.57 ± 6.48	37.43 ± 8.25	34.58 ± 12.85	0.0004
	O:	34.36 ± 6.73	37.22 ± 9.23	33.89 ± 12.69	
	L:	34.91 ± 6.84	37.71 ± 9.23	34.41 ± 12.50	
BMD	**F:**	1.37 ± 0.08*	1.17 ± 0.07	1.19 ± 0.067	0.0058
	O:	1.37 ± 0.08*	1.17 ± 0.074	1.2 ± 0.082	
	L:	1.366 ± 0.09*	1.18 ± 0.074	1.2 0 ± 0.071	
Fat free (g)	**F:**	66347.7 ± 7439.73*	42756.89 ± 5772.15	43686.56 ± 6254.84	0.0076
	O:	66776.8 ± 8002.91*	42827.33 ± 5737.99	44113.89 ± 6503.52	
	L:	66332.3 ± 7493.45*	42806.56 ± 5570.91	43930.78 ± 6615.83	
Gynoid (% fat)	**F:**	31.08 ± 5.75	39.46 ± 6.74*	39.99 ± 7.58*	0.003
	O:	30.98 ± 5.28	39.28 ± 7.37*	39.74 ± 7.80*	
	L:	31.09 ± 5.78	39.24 ± 6.74*	40.18 ± 7.24*	
Lean (g)	**F:**	62280 ± 7535.42*†	40295.78 ± 5692.9	41411.33 ± 5971.34	0.0074
	O:	62702.1 ± 7631.88*	40394 ± 5590.47	41513.89 ± 6208.72	
	L:	62471.3 ± 7385.85*†	40369 ± 5407.89	41399 ± 6363.83	
Scale Weight	**F:**	191.93 ± 21.06	136 ± 17.6	144.8 ± 40.5	0.0045
	O:	191.78 ± 21.06	136.6 ± 17.6	144.7 ± 40.3	
	L:	192.43 ± 21.87	136.7 ± 17.8	145.5 ± 40.4	
Tissue (% fat)	**F:**	25.52 ± 5.14	32.02 ± 6.46	32.03 ± 9.86	0.0018
	O:	25.12 ± 5.10	32.04 ± 6.56	31.61 ± 9.87	
	L:	25.44 ± 4.96	32.14 ± 6.47	32.14 ± 9.56	
Total Mass	**F:**	87.4 ± 9.12*†	61.87 ± 8.27	64.97 ± 18.34	0.0047
	O:	87.83 ± 9.58*	61.92 ± 7.88	65.11 ± 18.33	
	L:	88.09 ± 9.76*†	62.18 ± 8.19	65.25 ± 18.27	

Note: CON: male controls, OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory, L: luteal. η^2 was used to determine time effect size for all three groups. Group main effect in the 3x3 RM ANOVA were denoted using *($p<0.05$). † denotes group x visit interaction with significant increases from F to L in the 3x3 RM ANOVA. Standard deviations represent variability.

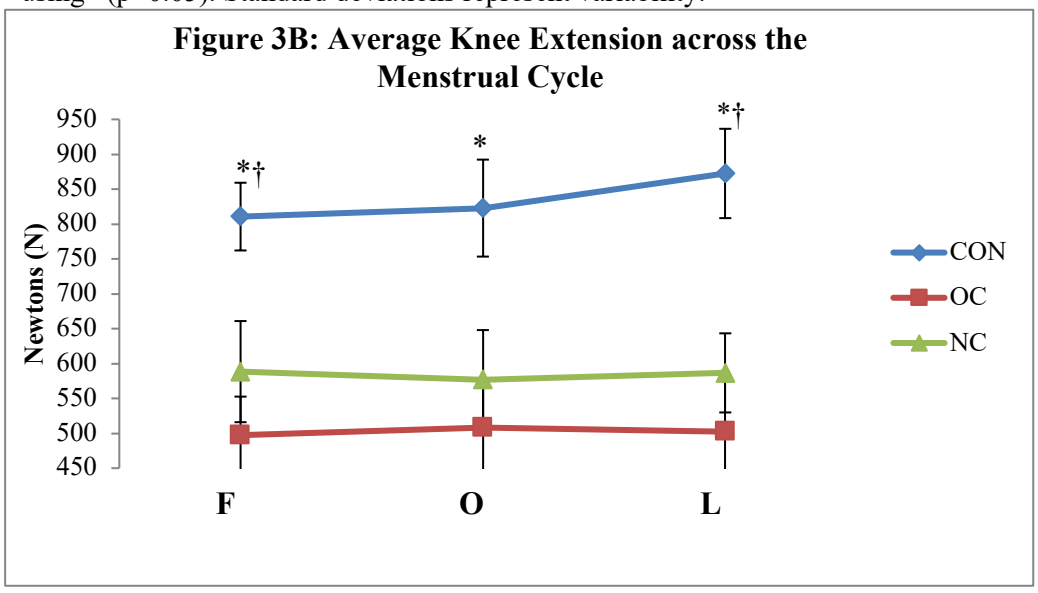
FORCE PRODUCTION

3 x 3(CON, OC, NC)

During the 3x3 Repeated Measures ANOVA, there was a significant group main effect where the CON group had higher knee extension ($p<0.001$ vs OC, $p=0.002$ vs NC) and knee flexion ($p=0.038$ vs OC), shown in Figures 3A and 3B. There was a significant time main effect ($p=0.007$), where knee extension values were higher in the luteal phase than the follicular phase in the CON group. Muscle quality was assessed as lean mass of the dominant limb and knee extension force production for all subjects. Running the ANOVA again with muscle quality values showed no significant group mina effect (p=), time main effect (p=) or group x time interaction (p=), which signifies the increase in lean mass was contributing to the males' increase in knee extension. This may be due to participants not adhering to testing guidelines to maintain the same amount of exercise through the study. There was no time main effect found in the OC or NC groups ($p>0.05$). There was no significant group x time effect for knee extension ($p=0.132$) or knee flexion ($p=0.116$). Results are displayed in Table 4. $\eta 2$ showed small effect sizes for all group force production measurements.



Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Significant group main effects CON > OC were denoted using *($p<0.05$). Standard deviations represent variability.



Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Significant group main effects CON > OC and NC were denoted using *($p<0.05$) from the 3x3 RM ANOVA. † denotes group x visit interaction with significant increases from F to L in the 3x3 RM ANOVA. Standard deviations represent variability.

2 x 3 (OC, NC)

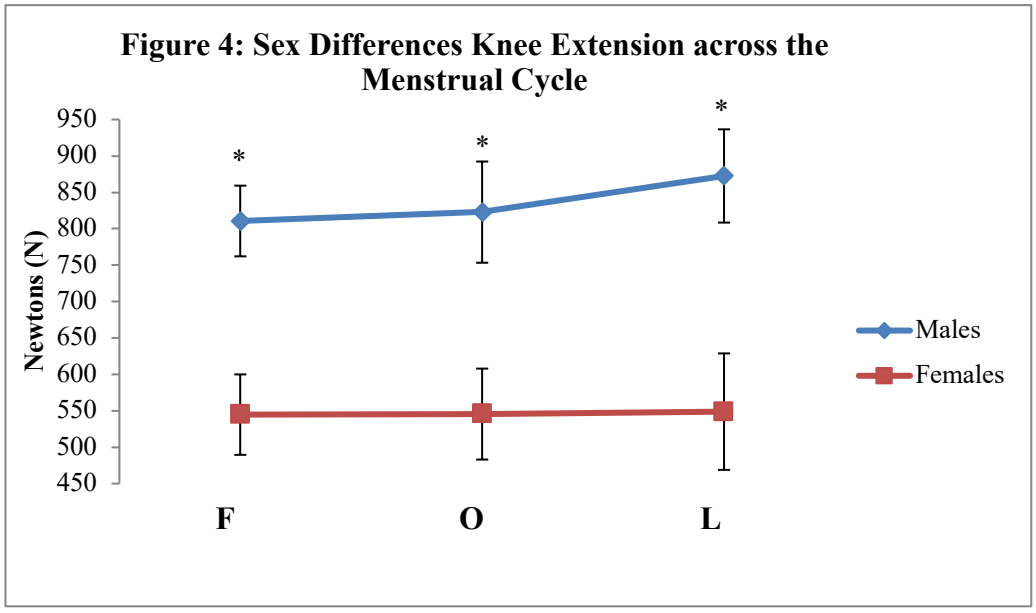
The non-significant 3 x 3 Repeated Measures ANOVA was then followed by the 2 (OC, NC) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA to further investigate changes within the females to compare contractive use. No group main effect was found for knee flexion ($p=0.83$) or knee extension ($p=1.99$). No time main effect was found for knee flexion ($p=0.63$) or knee extension ($p=0.96$). No group x time interaction was found for knee flexion ($p=0.054$) or knee extension ($p=0.707$).

1 x 3 (Collapsed Females)

A One-Way Repeated Measures ANOVA was used to assess female characteristics ($n=19$), which showed that there was no significant visit main effect (follicular, ovulatory and luteal) for knee extension ($p=0.95$) or knee flexion ($p=0.6$).

2 x 3 (Males, Collapsed Females)

Due to the non-significance in female groups, a 2 (male, female) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA was run to further investigate sex differences in force production across the menstrual cycle. There was a significant group main effect showing males had greater knee extension values than the females (males 835.31±45.34 N vs females 546.35±31.2 N, $p<0.001$). There was a significant visit main effect in knee extension in the males, shown in Figure 4. Males' knee extension was significantly higher during the luteal phase than the follicular phase ($p=0.007$). There was a significant group main effect showing males had greater knee flexion values than the females (males 308.4±20.53 N vs females 236.05±14.13 N, $p=0.007$). There was no significant time main effect ($p=0.173$) or group x time interaction ($p=0.732$) in knee flexion.



Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Significant group main effects CON > NC and OC were denoted using *($p<0.05$) from the 2x3 (Gender) RM ANOVA. † denotes group x visit interaction with significant increases from F to L in the 2x3 (Gender) RM ANOVA. Standard deviations represent variability.

Table 4: Isometric Knee Strength across the Menstrual Cycle

		CON (n=9)	OC (n=10)	NC (n=9)	η^2
Knee Flexion	**F:**	301 ± 48.5*	215.3 ± 55.2	240.9 ± 72.5	0.0023
	O:	305 ± 69.5*	220.3 ± 62.5	246.2 ± 71.3	
	L:	318.4 ± 64*	240.5 ± 80	231.4 ± 56.7	
Knee Extension	**F:**	810.6 ± 172.5*	497.3 ± 73.3	588.5 ± 162.1	0.005
	O:	822.8 ± 180.1*	508.2 ± 94.2	576.7 ± 140	
	L:	872.5 ± 160.6*	502.6 ± 76.6	586.6 ± 157.4	

Note: CON: male controls, OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory, L: luteal. η^2 was used to determine time effect size for all three groups. Significant group main effects CON > NC and OC were denoted using *($p<0.05$) from the 3x3 RM ANOVA. † denotes group x visit interaction with significant increases from F to L in the 3x3 RM ANOVA. Standard deviations represent variability.

PENNATION ANGLE

3 x 3 (CON, OC, NC)

The 3 x 3 Repeated Measures ANOVA showed were no significant group main effect (p=0.53), time main effect (p=0.27) or group x time interaction (p=0.97) in the vastus medialis (VM). The vastus lateralis (VL) also showed no significant group main effect (p=0.83), time main effect (p=0.11) or group x time interaction (p=0.97). Results revealed no significant group main effect (p=0.73), time main effect (p=0.63) or group x time interaction (p=0.97) in the rectus femoris (RF). Similarly, the vastus intermedius anterior (VIA) had no significant group main effect (p=0.95), time main effect (p=0.055) or group x time interaction (p=0.8). Concluding, there was no significant group main effect (p=1.0), time main effect (p=0.84) or group x time interaction (p=0.99) in total PA across the three menstrual cycle phases and three groups (CON, OC, NC). Results are shown in Table 5. $\eta 2$ showed small effect sizes for all group pennation angle measurements.

2 x 3 (OC, NC)

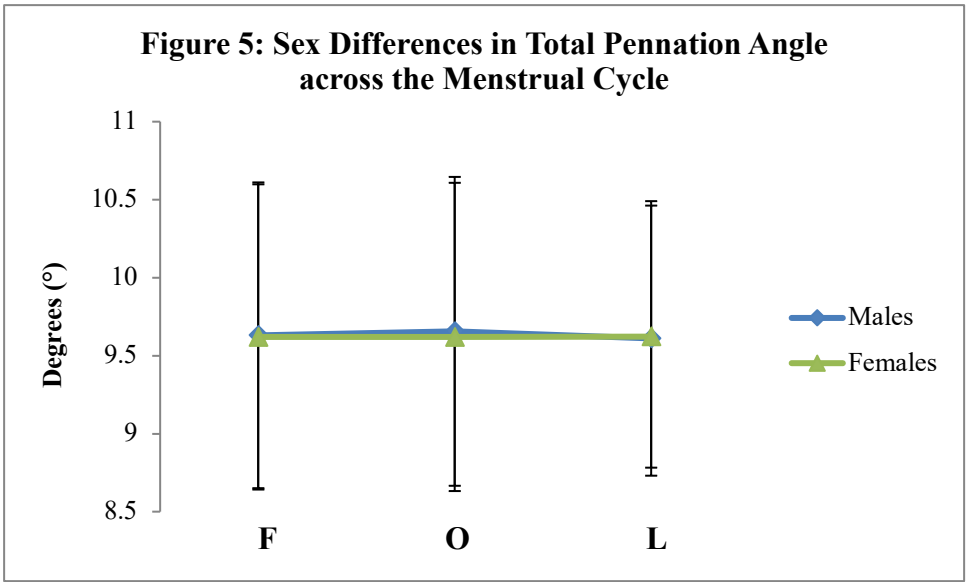
The non-significant 3 x 3 Repeated Measures ANOVA was then followed by the 2 (OC, NC) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA to further investigate changes within the females to compare contractive use. No significant group main effect was found for VM (p=0.29), VL (p=0.52), RF (p=0.49), VIA (p=0.77) or total PA (p=0.997). No significant time main effect was found for VM (p=0.38), VL (p=0.09), RF (p=0.65), VIA (p=0.33) or total PA (p=0.95). No significant group x time interaction was found for VM (p=0.74), VL (p=0.89), RF (p=0.56), VIA (p=0.87) or total PA (p=0.91).

1 x 3 (Collapsed Females)

A One-Way Repeated Measures ANOVA was used to assess female characteristics (n=19), which showed that there were no significant changes in time (follicular, ovulatory and luteal) for VIA (p=0.3), VM (p=0.38), VL (p=0.08) RF (p=0.61), or PA total (p=0.25).

2 x 3 (Males, Collapsed Females)

Due to the non-significance in female groups, a 2 (male, female) x 3 (follicular, ovulatory, luteal phases) Repeated Measures ANOVA was run to further investigate sex differences in pennation angle across the menstrual cycle. No significant group main effect was found for VM (p=0.95), VL (p=0.78), RF (p=0.89), VIA (p=0.89) or total PA (p=0.98). No significant time main effect was found for VM (p=0.26), VL (p=0.2), RF (p=0.5), VIA (p=0.15) or total PA (p=0.8). No significant group x time interaction was found for VM (p=0.93), VL (p=0.83), RF (p=0.98), VIA (p=0.51) or total PA (p=0.95). Total PA between females and males is shown in Figure 5.



Note: OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory and L: luteal. Differences if present were denoted using *($p<0.05$). Standard deviations represent variability.

Table 5: Quadriceps Pennation Angles across the Menstrual Cycle NC vs OC (mean±SD)

		CON (n= 9)	OC (n=10)	NC (n=9)	η^2
PA total	**F:**	9.63 ± 0.98	9.8 ± 0.9	9.6 ± 1.1	0.00002
	O:	9.66 ± 0.99	9.8 ± 0.9	9.6 ± 1	
	L:	9.58 ± 0.84	9.7 ± 0.8	9.6 ± 1	
RF	**F:**	13.06 ± 1.46	13.3 ± 1.5	12.6 ± 1.9	0.0229
	O:	12.96 ± 1.51	13.2 ± 1.6	12.6 ± 2.1	
	L:	12.9 ± 1.29	13.1 ± 1.4	12.6 ± 1.9	
VIA	**F:**	4.39 ± 0.91	4.7 ± 1	4.3 ± 1	0.048
	O:	4.68 ± 1.03	4.8 ± 0.9	4.4 ± 1.1	
	L:	4.45 ± 0.81	4.6 ± 0.8	4.4 ± 1	
VL	**F:**	11.07 ± 1.21	11.3 ± 0.5	11.2 ± 0.7	0.013
	O:	10.95 ± 0.9	11.2 ± 0.8	10.9 ± 0.5	
	L:	11.04 ± 0.95	11.3 ± 0.8	11 ± 0.5	
VM	**F:**	9.83 ± 1.56	9.8 ± 1.8	10.4 ± 1.81	0.004
	O:	10.04 ± 1.83	9.8 ± 1.9	10.6 ± 1.8	
	L:	9.942 ± 1.54	9.9 ± 1.9	10.4 ± 1.9	

Note: CON: male controls, OC: oral-contraceptive users, NC: non-contraceptive users, F: follicular, O: ovulatory, L: luteal. η^2 was used to determine time effect size for all three groups. Differences in mean±SD if present were denoted using *($p<0.05$). Standard deviations represent variability.

DISCUSSION

This section will present a detailed account of the results found in this study for both groups separated and combined. Results will be examined with respect to previous literature.

MAIN FINDINGS

1. **Weight, BMD, Fat-free mass (g), lean mass (g), total mass, android (% fat), force production as determined by knee extension MVIC, is significantly higher in males (CON) when compared to females (OC, NC).**
2. **Isometric knee strength and knee and hip ROM did not significantly change through the menstrual cycle for contraceptive users or non-contraceptive users.**
3. **The PA of the individual's muscles of the QF; the VM, VL, RF, and VI; did not significantly change throughout the menstrual cycle in any group.**

BODY COMPOSITION / ROM

As expected, CON males had significantly higher weight ($p<0.005$) and lean mass (g) and total mass ($p<0.001$) when compared to females in the OC and NC groups. The CON group had significantly higher lean mass during their ovulatory visit (62813±2115g) when compared to their follicular visit (62168±2115 g) ($p=0.044$). This finding in the CON group is limited by not having repeated IPAQ and nutrition logs every week of testing. Although participants were encouraged to keep the same diet and exercise schedule, we cannot justify why there was an increase between their follicular and

ovulatory phases in lean mass. Gynoid fat was significantly higher in both female groups (OC, NC) when compared to males (p=0.012 for both OC and NC). This is in accordance with other research studies, which illustrates that females have more gynoid fat, while men have more android fat.[35,36,37] Fat deposition regions (android, gynoid and android/gynoid (A/G) ratio) are other body composition measurements that were assessed via DXA. Android body type is more common in males and is indicative of increased risk for cardiovascular disease and diabetes mellitus[35]. Meanwhile, women on average have more gynoid body types. A/G ratio, similar to hip-waist ratios, are commonly used to assess where the majority of fat is located which can help us non-invasively understand overall health and risk for disease.[35] Gynoid % fat was significantly higher in the female groups (OC and NC) than males (CON) (p=0.012 in both OC and NC), which is in line with past research.[35] CON males had significantly higher A/G ratios compared to female (OC and NC) (p=0.004 in both OC and NC), which is linked to the lower gynoid fat in the males.

The data of this study suggests that hip and knee ROM and body composition measurements via DXA may not be altered significantly throughout the menstrual cycle. This is similar to the findings by Luiz da Silda Texeira et al. who assessed body composition (body mass, height and BMI) as well as sit-and-reach flexibility measurements in the follicular and luteal phases of the menstrual cycle in contraceptive and non-contraceptive women.[5] They found no significant body composition or flexibility differences were found between or within the control or experimental group.[5] When comparing contraceptive and non-contraceptive users, we found that on average the non-contraceptive users had more hip extension ROM through all three phases when compared to contraceptive-users (p=0.049). We also found that there was a 11% absolute difference

between menstrual cycle phases ($p<0.05$).This may be due to the increased laicity that occurs during menses and ovulation.[38] It has been shown to be a link to hamstring strains and ACL tears.[1,2,8] Samuelson et al. completed a systematic review over hormonal contraceptives having a "protective benefit" for potential injury.[8] Although that literature is very conflicting, ROM may be a future topic of interest for female athletes and track menstrual cycle changes. The 1-Way Repeated Measures ANOVA showed no significant changes in time (follicular, ovulatory and luteal) for any ROM variables in all females combined.

Our results also coincide with Podfigurna-Stopa et al. who assessed body composition in women with functional hypothalamic amenorrhea.[38] Similar to our results, they found no significant differences in total body fat mass (TBFM), total body lean mass (TBLM), bone mineral density (BMD). All of our data (% fat, total mass, lean mass, fat free mass) showed $\leq$1% absolute difference in all DXA variables Data shows that water retention occurs during the pre-ovulatory phases (follicular and ovulatory phases), but these results suggest that using a DXA scan may limit our understanding of what physiological variables are impairing fluid and electrolyte balance, leading to sodium and water retention during the menstrual cycle. The 1-Way Repeated Measures showed no significant changes in time (follicular, ovulatory and luteal) for any body composition variables in all females combined. The lack of group differences and collapsed females differences across the menstrual cycle all led to the rejection of the hypothesis that ROM or DXA body composition will change through the menstrual cycle. [39] It is important to note that when comparing the female groups for differences in contraceptive-use, statistics

were underpowered (<0.8), concluding that a larger sample size may have altered result even with small effect sizes.

FORCE PRODUCTION

As expected, the males in this study were significantly stronger than both female groups in knee extension (<0.001) and knee flexion (p=0.038). Results showed neither of the two female groups (OC nor NC) had significant isometric force production changes throughout their menstrual cycles. A study by Janse de Jonge assessed isometric quadriceps strength in oral contraceptive through hormone assays.[3] No significant differences were found between any of the phases of the menstrual cycle in isometric quadriceps strength, which corresponds with our results. Our results do not align with Sarwar and colleagues who found that isometric strength increased during the ovulatory phase in non-contraceptive women.[4] This study looked at early and mid-follicular, mid-ovulatory and mid and late-luteal phases, while we only assessed early follicular (menses), mid-ovulatory and mid-luteal. This may be due variations in scheduling menstrual cycle visits off of menstrual cycle questionnaires, which is a limiting in both studies when compared to hormonal assays.

As previously mentioned, group differences as determined by 2-way RM ANOVA analysis were not significant for any factors in the OC and NC groups. The 1-Way Repeated Measures ANOVA showed no significant changes in time (follicular, ovulatory and luteal) for any force production variables in all females combined. The lack of group differences and collapsed females' differences across the menstrual cycle all led to the rejection of the hypothesis that force production will change through the menstrual cycle.

The menstrual cycle phases appeared to have no effect on force production, but it is still recommended to control for menstrual cycle potential fluctuations when testing female participants in the future.

PENNATION ANGLE

The results of this study showed PA in the QF muscles was not significantly altered through the follicular, ovulatory and luteal phases of the menstrual cycle, regardless of group. To our knowledge, no other studies have assessed menstrual cycle changes on PA. This coincides with Baroni et al. who after 4, 8 and 12 weeks of resistance training found that PA was unchanged.[30] The 1-Way Repeated Measures ANOVA showed no significant changes in time (follicular, ovulatory and luteal) for any pennation angle variables in all females combined. The lack of group differences and collapsed females differences across the menstrual cycle all led to the rejection of the hypothesis that quadriceps pennation angle will change through the menstrual cycle. It is possible the small sample size and lack of experience of ultrasound measurements by the researchers limited the study's ability to find interactions between phases. If possible, future research should examine these factors with a larger group and with a researcher or trained individual who is experienced in the use of ultrasonography.

MENSTRUAL CYCLE CONSIDERATIONS

Previous research has extensively examined menstrual cycle fluctuations in exercise performance testing between various phases and samples.[4,17,40] However, due to small sample sizes and requiring < 6 months for the regulation of hormone levels for oral contraceptive use or non-use, research was still needed to investigate changes in body composition, force production and mobility. To our knowledge, no research has examined

the change in pennation angle across the menstrual cycle. Gaining an understanding of how the menstrual cycle relates to tested measurements could be the next step towards using these variables to determine training needs in athletes or to identify deficiencies resulting from prior injury or disease in clinical populations.

CHAPTER V
CONCLUSION

The purpose of this study was to assess changes in lower limb muscular architecture (pennation angle (PA)) and functional parameters, such as: strength, mobility, lean mass, fat mass, in females during the luteal, ovulatory and follicular phases of the menstrual cycle. A secondary purpose of this study is to examine differences in physiological variables between non-contraceptive users and hormonal contraceptive users.

Research Questions

1. What are the differences in hip and knee ROM across menstrual cycle phases?
2. What are the differences in body composition values across menstrual cycle phases?
3. What are the differences in force production (FP) across menstrual cycle phases?
4. What are the differences in pennation angle (PA) across menstrual cycle phases?

Hypotheses

#1 H_0: There will not be a difference in hip and knee ROM across menstrual cycle phases.

#1 H_1: There will be a difference in hip and knee ROM across menstrual cycle phases.

→ Fail to reject the null hypothesis. No hip and knee ROM differences were found across the menstrual cycle phases.

#2 H_0: There will not be a difference in body composition values across menstrual cycle phases.

#2 H_1: There will be a difference in body composition values across menstrual cycle phases.

→ Fail to reject the null hypothesis. No body composition differences were found across the menstrual cycle phases.

#3 H_0: There will not be a difference in force production across menstrual cycle phases.

#3 H_1: There will be a difference in force production across menstrual cycle phases.

→ Fail to reject the null hypothesis. Force production differences were found across the menstrual cycle phases.

#4 H_0: There will not be a difference in pennation angle across menstrual cycle phases.

#4 H_1: There will be a difference in in pennation angle PA across menstrual cycle phases.

→ Fail to reject the null hypothesis. No pennation angle differences were found across the menstrual cycle phases.

CLINICAL SIGNIFICANCE

The absence of menstrual cycle research and the amplified injury risk in females suggest that we should expect to see alterations in testing measurements through the menstrual cycle. The lack of significant changes in variables between the NC and OC groups indicates oral contraceptives may not play a role in altering changes in body composition, hip and knee ROM, isometric knee strength and quadriceps PA. However, it is possible a larger study would suggest otherwise. Although there were no significant changes in our measurements, it is important to control for the testing phase for future research studies to minimize error with hormone fluctuation through the menstrual cycle.

FUTURE RESEARCH

This research should be taken and applied to future research studies. Future researchers should examine these measures on a larger scale in a population who are various types of birth control. Through recruiting, there were many hormonal and non-hormonal IUD users that could not participant in the current study, which would only

enhance exercise endocrinology research. Female athletes could also be used to examine changes in our testing variables through the menstrual cycle and throughout a sport's season. The second direction would be a longitudinal study of female athletes with hamstring strains and ACL tears and how their muscle architecture, body composition and force production differ from the results seen in the current study's healthy population. Assessing other lower limb architecture, strength and ROM may be helpful in discovering what is truly being altered through the menstrual cycle.

LIMITATIONS

The results of this study are only representative of college-aged participants 18-30 years old from the Norman area. Additionally, muscle architecture was estimated based on locations previously determined to be most representative of whole muscle architecture. Although the interrater reliability for pennation angle was < 2%, since there were repeated measures occurred a week apart, image locations may not have been as precise as intended. This study was limited by individual variability of hormone levels that were not assessed and estimated by a Mensural History Questionnaire. Participants were encouraged to give maximal effort on all MVIC testing, but once again the time between visits as well as a training effect may have influenced results. Five participants did not complete the study, although our sample was still within our G-Power analysis predicted range, a higher sample size in each group would have been ideal.

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APPENDIX A

IRB Approval Letter
Informed Consent
Research Privacy Form
Physical Activity Readiness Questionnaire
IPAQ
Menstrual History Questionnaire
Medical History Questionnaire

The UNIVERSITY of OKLAHOMA

Institutional Review Board for the Protection of Human Subjects

Initial Submission – Board Approval

Date: March 29, 2019
From: Karen Beckman, MD, Chair
To: Rebecca Larson, PhD

IRB#: 10260
Meeting Date: 03/04/2019
Approval Date: 03/28/2019
Expiration Date: 02/28/2020

Study Title: Menstrual Cycle Changes in Quadricep Muscular Architecture and Other Functional Parameters in College-Aged Females Compared to Males

Reference Number: 678727

Study Status: Active - Open | CR Req
Risk/Benefit Assessment: Research not involving greater than minimal risk.

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 and the study documents approved for this submission, open this study in IRIS 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*.

Informed consent and research privacy authorization must be obtained using the currently approved, stamped forms. You must retain all original, signed forms, as applicable.

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 an annual check-in form to the IRB upon notification approximately 45 days prior to the check-in due date indicated above.

The following are also required if applicable to this research study:

- You may *not begin your study* until the contract through Office of Research Administration (ORA) is finalized and signed as per OUHSC institutional policy.
- If this study involves collaboration with multi-center sites requiring a reliance agreement for OUHSC to serve as IRB of record for non-OU sites, submit a modification as soon as possible to add each non-OU site and non-OU collaborator to the application and begin the reliance agreement process.

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

Consent Form to Participate in a Research Study
University of Oklahoma Health Sciences Center (OUHSC)
University of Oklahoma Norman Campus (OU)
Study Title: Menstrual Cycle Changes in Quadricep Muscular Architecture and Other Functional Parameters in College-Aged Females Compared to Males
Sponsor: OU Department of Health and Exercise Science
Principal Investigator: Rebecca D. Larson, Ph.D.
Phone Number: 352-359-8432 (cell) or 405-325-6325 (office)

KEY INFORMATION ABOUT THE RESEARCH STUDY

You are being asked to participate in a research study. Research studies are voluntary and include only people who choose to take part. This consent form begins with a 'Key Information' section to provide important information to help you decide whether or not to participate in this study. More detailed information is provided after the key information. Please take your time, discuss this with family and friends, and ask the investigator and study team any questions you may have.

WHY HAVE I BEEN ASKED TO PARTICIPATE IN THIS STUDY?
You are being asked to participate in this research study because you are a healthy individual who is free from any known diseases or injuries that may affect the lower limbs.

WHY IS THIS STUDY BEING DONE AND HOW LONG WILL IT LAST?
Past studies have shown that female athletes have significantly more anterior cruciate ligament (ACL) tears compared to male athletes. The purpose of this study is to investigate changes within leg muscles and functions, such as: strength, range of motion, lean mass, and fat mass, in females during the different phases of the menstrual cycle. A secondary purpose of this study is to examine differences in your physiology between non-contraceptive users and hormonal contraceptive users. Males are included for comparison.

You will be in the study for a total of 4-5 weeks during which you will visit the Body Composition and Physical Performance Lab on 4 occasions. Each visit will take approximately 1-2 hours to complete.

WHAT WILL I BE ASKED TO DO IN THIS STUDY?
If you decide to participate in this study, you will be asked to take part in 4 study visits. The first visit will include filling out paperwork and introducing you to the equipment for the study. Visits 2-4 will each include vital signs (heart rate, blood pressure, height and weight), the testing of joint angles in the hip and knee, DXA scans to assess amounts of fat mass and fat-free mass (body composition), ultrasound testing of the front thigh, and force measurements of pushing the knee forward and pulling the knee backwards to get strength measurements. A total of 3 DXA scans and 3 ultrasound testing visits will be required for study.

WHY MIGHT I WANT TO PARTICIPATE IN THIS STUDY?
If you agree to take part in this study, there will not be direct medical benefit to you. We hope that the information learned from this study will benefit future exercise physiology research.

WHY MIGHT I NOT WANT TO PARTICIPATE IN THIS STUDY?
While participating in the study you will be asked to contract your leg muscles which can result in mild discomfort and/or muscle tenderness following contraction. You may find the strength testing uncomfortable. You may also experience heavier than normal breathing while contracting maximally. You will be closely monitored for any possible ill effects. To further increase your safety, you will be screened for risk factors.

This study involves exposure to radiation from an x-ray procedure that is being performed for research purposes only, and not required for medical care.
The radiation exposure in this study may be hazardous to an unborn child. Therefore, females will be asked to perform a simple urine test to determine possible pregnancy.
The researchers do not know all of the adverse effects that could happen. For a complete description of known risks, refer to the Detailed Information section of the consent form.

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.

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 75 women and men will take part in this study.

WHAT IS INVOLVED IN THE STUDY?
If you decide to participate in this study, you will be asked to come to the Body Composition and Human Performance Lab for 4 visits. There will be three groups: Non-contraceptive users (NC), Hormonal Oral-contraceptive users (OC) and a male control group (CON). NC and OC groups will be assessed by menstrual cycle questionnaires. The NC will be required to not be on any contraceptives (hormonal or non-hormonal) for at least six months prior to testing. The OC group will be required to be taking hormonal oral contraceptives for at least six months prior to testing. Medication use will be assessed through the Medical History Questionnaire to make sure no hormonal medication is being used by the NC and CON group. We will request you to avoid any lower body resistance training or endurance training 24 hours prior to testing and to avoid caffeine use 4 hours prior to testing. You will be encouraged to continue exercise as usual throughout the study. Testing protocols during each visit will be the same for everyone.

Scheduling:
Visit 1 can occur at any time. Visits 2-4 will be scheduled based on the participant's menstrual cycle phase. We will use the luteal (days 1-4), ovulatory (days 12-15) and follicular (days 18-21) phases to schedule visits based on menstrual cycle questionnaires (if necessary). The non-contraceptive users (NC) and oral-contraceptive users (OC) groups will begin Visit 2 on the first few days (days 1-4) of their

menses (where you begin to menstruate). Visit 3 will test in the ovulatory phase and Visit 4 will test in the luteal phase.

Visit 1:
Visit 1 will begin with an explanation of inclusion and exclusion criteria and a brief explanation of the protocols included in the study. Each participant will have the study explained to them and sign an informed consent and Health Insurance Portability and Accountability Act (HIPAA) form. You will also complete a physical activity readiness questionnaire (PAR-Q), menstrual history questionnaire (if applicable), and an International Physical Activity Questionnaire (IPAQ). Height, weight, resting blood pressure (BP), resting heart rate (HR) will be assessed at the beginning of each visit. Visit 1 will include a familiarization with the maximal voluntary isometric contraction muscle protocol (MVIC) for knee extension and flexion, hip and knee mobility assessment and assessment.

Visits 2-4:
Visits 2-4 will start by measuring weight, resting blood pressure, resting heart rate, hip and knee range mobility, hydration status and a pregnancy test (if necessary). Mobility measurements will be taken with a goniometer, which measures the angle your hip and knee joints. The goniometer will be lined up with your hip, knee and ankle bones to see the maximal mobility (flexibility in the four measurements. After mobility measurements, the length of the femur will be assessed. Next, you will complete a body composition assessment using a Dual-Energy X-Ray Absorptiometry (DXA) scan. Then we will assess the muscles of your dominant leg using B-mode ultrasound. Ultrasound measurements will be followed by our force production protocol. We will assess knee extension and flexion through maximal voluntary isometric contractions (MVICs) in your dominant leg with a force dynamometer. Three warm-ups and three MVICs will be taken for each knee extension and flexion.

There will be five main procedures used in this study: initial assessment, mobility assessment, body composition assessment, muscle architecture assessment and force production.

Initial assessment:
During the initial assessment, weight, blood pressure and heart rate will be assessed. During this initial assessment we will also measure the femur length (largest bone going from the hip to knee) to prepare for ultrasound measurements for muscle architecture assessment. Marks will be made at 22%, 56%, and 73% of the thigh to allow for consistent measurements between visits.
Mobility assessment:
During mobility assessment, we will assess hip and knee range of motion (ROM).
Body composition assessment:
Body composition will be measured using a whole-body Lunar dual-energy x-ray absorptiometry (DXA) scanner, a type of x-ray. Prior to scanning, urine will be assessed and be between the range of 1.005-1.030 to ensure proper hydration levels. If you are outside of the range, you will be given water and reassessed 30 minutes later or rescheduled for another day. Women will be asked to take a pregnancy test before every testing. If you become pregnant at any time during the study, you will be disqualified from the study due to fetal adverse effects related to radiation in DXA scans. After clearing the USG levels and pregnancy test, you will then be asked to lay down on your back. You will have three DXA scans during this study to distinguish body composition differences seen during the menstrual cycle in all two groups: OC and NC, and estimated in the CON group.
Muscle architecture assessment:
Three images will be taken of the following muscles of the front of your upper leg; Vastus medialis (VM), vastus lateralis (VL), rectus femoris (RF), and vastus intermedius (VIA) on your dominant leg. All images will be transferred to a USB drive to be further analyzed through an image processing software. Your

name will be removed from your data and it will be only identifiable with your participant ID so that it will be confidential.
For quadriceps assessment, you will be seated with both hips and knees at a 90° angle. Your back, knees, and feet will be adjusted and supported to ensure those angles could be maintained with the entire lower body completely relaxed.

Force production assessment:
We will use introduce you the equipment being used for maximal voluntary isometric contractions (MVICs) for both knee extension and knee flexion. That will consist of having you sit on the KinCom, which is a machine that tests how much force you can kick forward or backwards and adjusting the seat until your knee and hip angles are both 90°. Straps will then be fastened to secure your upper body to the seat to ensure that your quadriceps and hamstring muscles are isolated.
You will then be asked to perform isometric knee extensions at perceived efforts of 25%, 50%, and 75% until you feel comfortable with the strength protocol. You will then be asked to perform three MVICs at full (maximal) effort. The same process will be completed for isometric knee flexion.

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

- If it is determined that it is in your medical best interest.
- New information becomes available.
- You become pregnant.
- You fail to follow study requirements.
- The study is stopped by the sponsor.

WHAT ARE THE RISKS OF THE STUDY?
While participating in the study you will be asked to maximally contract your quadriceps, hamstring, and calf muscles which can result in mild discomfort and/or muscle tenderness following contraction. You may find the dynamometer seat or attachments uncomfortable. You may also experience heavier than normal breathing while contracting maximally. While rare and uncommon, you may experience faintness, nausea, and/or lightheadedness. You will be closely monitored for any possible ill effects. To further increase your safety, you will be screened for risk factors.

This study involves exposure to radiation from three DXA scans that are being performed for research purposes only, and not required for medical care. The amount of radiation exposure is equivalent to less than the daily amount of natural background radiation exposure people in the United States receive. The risk of radiation is cumulative over your lifetime.
The radiation exposure in this study may be hazardous to an unborn child. As such you will be asked to perform a simple urine test to determine possible pregnancy. The test will be free. A negative pregnancy test is needed prior to participating in the DXA scan. For unexpected pregnancies, subjects are encouraged to speak with their family physician. The three DXA scans and other research procedures are not for diagnostic purposes and if you have any questions about your test results, you should see a physician. You will be given the results of your DXA scan (body composition) on the last visit 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. Other drugs may be

given to make side effects less serious and uncomfortable. Many side effects go away shortly after the force production protocol is stopped.

For more information about risks and side effects, ask the researcher or contact Dr. Rebecca Larson at 352-359-8432 (cell) or 405-325-6325 (office).

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 an average person; 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 AND MEN:
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 DXA scan while you are pregnant or breastfeeding may involve risks to an embryo, fetus, or infant, including birth defects which are currently unforeseeable.

If you currently are taking oral contraceptives (for OC group only), in order to reduce your risk of pregnancy, you should regularly and consistently take your medication, while you are in this 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 will be withdrawn from the study. Payment for all aspects of obstetrical, child, or related care will be your responsibility.

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. 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.

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.

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. 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 The University of Oklahoma 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. 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.

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. Rebecca Larson at 352-359-8432 (cell) or 405-325-6325 (office).

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**

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: **Menstrual Cycle Changes in Quadricep Muscular Architecture and Other Functional Parameters in College-Aged Females Compared to Males**

Leader of Research Team: **Rebecca D Larson, PhD**

Address: **Department of Health and Exercise Science, 1401 Asp Avenue, Room 117 HHC, Norman OK 73019**

Phone Number: **405-325-6325**

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 can include physical findings from questionnaires, dual X-ray absorptiometry (DXA) scan, and isometric dynamometer tests.

Purposes for Using or Sharing PHI. If you give permission, the researchers may use your PHI to investigate changes within leg muscles and functional parameters, such as: strength, range of motion, lean mass, and fat mass, in females during the different phases of the menstrual cycle. Males are included for comparison.

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 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 outside the research team

[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.**

PHI Research Authorization

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

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.

FORM 3.1 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		
☐	☐	1.	**Has your doctor ever said that you have a heart condition and that you should only do physical activity recommended by a doctor?**
☐	☐	2.	**Do you feel pain in your chest when you do physical activity?**
☐	☐	3.	**In the past month, have you had chest pain when you were not doing physical activity?**
☐	☐	4.	**Do you lose your balance because of dizziness or do you ever lose consciousness?**
☐	☐	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?**
☐	☐	6.	**Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?**
☐	☐	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.

Supported by: Health Canada / Santé Canada

continued on other side...

IRB NUMBER: 8669
IRB APPROVAL DATE: 06/22/2017

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 Questionnaires (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***

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***

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***

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.

Department of Health and Exercise Science
University of Oklahoma

MENSTRUAL HISTORY QUESTIONNAIRE

Participant ID:_____ Date:_____

We are asking you to give us as complete a menstrual history as possible. All information is strictly confidential.

Are you pregnant (circle your response)
YES- Do not complete the rest of this form
NO- Continue to section A.

SECTION A: CURRENT MENSTRUAL STATUS

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)

Jan Feb Mar Apr May Jun Jul Aug Sep Oct Nov Dec

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.

3. When was the date of the onset of your last period?

4. When do you expect your next period?

5. What is the average length (number of days) of your menstrual flow? _____ days

How many of these days do you consider "heavy"?_____days

6. Do you take oral contraceptives or any other medication that includes estrogen and/or progesterone?

If yes, how long have you been taking this medication?_____

What is the brand name and dosage of this medication?_____

Has this medication affected your menstrual cycle (regularity, length and amount of flow)? If yes, indicate changes.

Date:_____

Medical History Questionnaire

Name: _____ Date of Birth: _____
Address: _____ Phone Number: _____
_____ alt #: _____
Email: _____
Age: _____
Dominant side: Left Right (circle)
Blood Pressure: _____/_____
Height: _____ Weight: _____ Shoe Size: _____
Sex: Male Female (circle) Gender: Male Female (circle)
Ethnicity: Caucasian African American Hispanic Asian Other: _____
Emergency contact name and number: _____

Please answer the following questions:

1. Have you ever been diagnosed with diabetes?
   Y N If "yes," when where you diagnosed? _____
2. Have you ever been told by a physician that you have Osteoporosis/Osteopenia?
   Y N
3. Have you ever had a heart attack or stroke?
   Y N If "yes," what and when? _____
4. Have you ever been diagnosed with any disease affecting the brain, spine, or nerves? (ex: Multiple sclerosis, brain tumors, epilepsy, Parkinson's disease, Neuropathy, ALS, etc.)
   Y N If "yes," what and when? _____
   _____
5. Have you ever been diagnosed with arthritis?
   Y N If "yes," when?
6. Have you had any injuries of the lower limbs specifically involving bone, tendon, or ligament damage?
   Y N If "yes," what and when? _____
   _____
7. Have you had any injuries of the lower limbs specifically involving the muscles?
   Y N If "yes" what and when? _____
   _____
8. Do you experience frequent pain in your lower limbs?
   Y N If "yes," where and how often? _____

9. Do you have a decreased range of motion or mobility in your hips, knees, or ankles?
   Y N If "yes," how much and where? _____
10. Do you use an assistive device for walking?
   Y N If "yes," what? _____
11. Do you experience any difficulties producing and maintaining rapid and repetitive movements?
   Y N If "yes," then describe. _____
12. Are you currently on any kind of medications?
   Y N If "yes," what medication, amount taken, time on medication, and reason.
   _____
   _____
13. Is there anything else you feel that the researchers should be aware of?

_____

_____

_____

I certify that these answers are accurate and complete

_____

Your Signature: Date:

Witness: _____ Date:_____

APPENDIX B
Data Collection Sheet

□ NC □ OC □ CON Subject ID: MC-___-0____

□ Familiarization □ Follicular □ Ovulatory □ Luteal Date: ___/___/ 2019

Day of cycle: ____ Researcher: _____

I. Initial Assessments:

Dominant limb □ Left □ Right Thigh length: _____cm

Height: _____ in. Weight _____ lbs. 22%: _____ cm

56%: _____ cm

BP: ____/_____mmHg RHR: ____ bpm 73%: _____ cm

II. Mobility Assessment:

Hip flexion ←_____° Knee extension→_____°

Knee flexion ←_____° Hip extension→_____°

III. Body Composition Assessment:

USG: _____ (1.005- 1.03)

VI. Muscle Architecture Assessment:

Muscle:	1:	2:	3:	Trial Av.:	Muscle Av.:
VM 1					VM total:
VM 2					
VM 3					
VL 1					VL total:
VL 2					
VL 3					
RF 1					RF total:
RF 2					
RF 3					
VIA 1					VIA total:
VIA 2					
VIA 3					

Data entered by: _____

Date: _____