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THE EFFECTS OF THE MENSTRUAL CYCLE ON AGILITY

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TABLE OF CONTENTS

ABSTRACT.....	v
CHAPTER I. INTRODUCTION.....	1
CHAPTER II. REVIEW OF LITERATURE.....	4
2.1 Effects of the Menstrual Cycle on Substrate Kinetics.....	5
2.2 Effects of the Menstrual Cycle on the Anabolic Response Post-Exercise.....	6
2.3 Mediators of Agility.....	7
2.4 Effects of the Menstrual Cycle on the Neuromuscular System.....	10
2.5 Effects of the Menstrual Cycle on Joint Laxity.....	12
2.6 Effects of the Menstrual Cycle on Joint Stiffness.....	16
2.7 Effects of the Menstrual Cycle on the Prevalence of Injuries.....	16
2.8 Oral Contraceptives and Athletic Performance.....	17
CHAPTER III. METHODS.....	19
3.1 Study Design.....	19
3.2 Reactive Agility Test.....	21
3.3 T-Test.....	22
3.4 Statistical Analysis.....	23
CHAPTER IV. RESULTS.....	24
CHAPTER V. DISCUSSION.....	27
5.1 Limitations.....	30
5.2 Future Considerations and Conclusions.....	30
REFERENCES.....	31
APPENDIX.....	38
Appendix A. IRB Approval.....	39
APPENDIX B. Informed Consent.....	40
APPENDIX C. Screening Questionnaire.....	43
APPENDIX D. AHA/ACSM Questionnaire.....	44
APPENDIX E. Reactive Agility Test.....	46
APPENDIX F. T-Test.....	47
APPENDIX G. Abbreviations.....	48

LIST OF FIGURES

FIGURE 1. EFFECT OF MENSTRUAL CYCLE PHASE ON T-TEST AGILITY PERFORMANCE.....26

LIST OF TABLES

TABLE 1. EFFECT OF MENSTRUAL CYCLE PHASE ON AGILITY PERFORMANCE.....	25
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THE EFFECTS OF THE MENSTRUAL CYCLE ON AGILITY

KARIM G. KHENISER

ABSTRACT

It has long been speculated that the menstrual cycle affects athletic performance. Whether athletic performance is impacted by menstrual cycle phase is dependent upon an array of factors: the type of activity (i.e., aerobic or anaerobic); oral contraceptive (OC) use; and, possibly, if mental processing is required during the execution of the task. Prior research has indicated that alterations in estradiol (E2) affects cognitive performance. However, there is a paucity of information about whether E2 modulates the speed of cognitive processing in a sports setting, which may cause alterations in sport performance throughout the menstrual cycle. Additionally, variations in E2 levels during the menstrual cycle are associated with changes in joint laxity, which may translate into changes in quick and powerful movements, such as multidirectional running patterns that are required in many sports. **PURPOSE:** The present study aimed to deduce if fluctuations in E2 influence agility performance in the reactive agility test (RAT) and T-Test. The former requires mental processing during its execution, whereas the T-Test does not. **METHODS:** Subjects ($n=10$; BMI 22.4 ± 1.4 -kg/m²; weight 61.3 ± 5.6 -kg; age 24.2 ± 5 -years; height 165.58 ± 8.29 -centimeters) were tested on the RAT and T-Test, during mid-cycle ([MC], high E2) and the early follicular phase ([EF], low E2) of the menstrual cycle. Subjects monitored urine levels of luteinizing hormone (LH), to determine when the surge in E2 occurs and correspondingly the MC test timing. **RESULTS:** With respect to the T-Test, the results indicated that the subjects were significantly faster during EF,

relative to MC (12.3 ± 0.89 -s vs. 12.56 ± 0.90 -s, $p=0.007$). However, there were no significant differences in RAT agility times between EF and MC. CONCLUSION: Cognitive function (i.e., mental processing) during the execution of sports-related movements is not affected, but joint laxity and the subsequent alterations in biomechanics may hinder the ability to execute athletic maneuvers that require sudden changes in direction. Strength and conditioning coaches and athletes should conduct agility trials during EF to have greater consistency between trials; especially when assessing the efficacy of a strength and conditioning paradigm.

CHAPTER I

INTRODUCTION

Substantial interest has been devoted to studying how the menstrual cycle may affect athletic performance and mediate injury risk. As it pertains to the latter, there is more than a three-fold higher incidence of anterior cruciate ligament injuries (ACL) among women. The mechanisms that are causal to ACL-related injuries are multifactorial: sexual dimorphism in muscular strength, morphological differences, and the exposure to pulsatile secretions of ovarian hormones. Of note, the latter has garnered an overarching degree of attention, when studying the etiology of ACL injuries. Although the menstrual cycle is characterized by cyclic changes in hormones, to delineate the effects of endogenous hormones on injury risk, three distinct and segmented hormonal milieus are isolated during the menstrual cycle and are of critical importance. Specifically, estrogen (E) levels are significantly attenuated, increased, and increased during the early follicular phase (EF), mid-cycle (MC), and the mid-luteal phase (ML), respectively. Moreover, progesterone (PG) concentrations are low during EF and MC and upregulated in the ML phase, while testosterone (T) levels are elevated during MC.

One prevailing thought is that these perturbations in endogenous hormones lead to increases in knee joint laxity, which may alter biomechanical patterns (e.g., increased knee valgus during landing) and contribute to debilitating injuries (Shultz et al., 2012a). Similarly, reciprocal decreases in musculotendinous stiffness (change (Δ) in force/ Δ length), which is specifically related to the menstrual cycle, also represents one mechanism that heightens risk to injury (Shultz, Schmitz, & Beynnon, 2011).

Although the aforementioned has yielded pertinent information, with regards to ascertaining the precipitating factors that lead to ACL injuries, there appears to be no conclusive and significant evidence to indicate that the menstrual cycle affects athletic performance. To date, muscular strength has been shown to be unaltered by fluctuations in endogenous hormones (Dibrezzo, Fort, & Brown, 1991; Elliott, Cable, Reilly, & Diver, 2003; Friden, Hirschberg, & Saartok, 2003; Janse de Jong, Boot, Thom, Ruell, & Thompson, 2001; Lebrun, McKenzie, Prior, & Taunton, 1995), and substrate kinetics is equally unaffected by the menstrual cycle during aerobic physical activity, as evidenced by novel well-controlled trials (Horton, Miller, & Bourret, 2006; Horton, Miller, Glueck, & Tench, 2002; Jacobs, Casazza, Suh, Horning, & Brooks, 2005; Smekal et al., 2007). However, higher concentrations of E in fertile females and infusion of E in post-menopausal women improves verbal memory (i.e., cognitive function) and laxity modulates biomechanical patterns (Rosenberg & Park, 2002; Wolf et al., 1999); consequently, it is conceivable that the former may affect agility times during open-skilled (requires mental processing) in relation to closed-skilled (does not require mental processing) agility movements, whilst laxity may affect biomechanical patterns. Taken

together, agility can be modified by impaired mental processing and increased joint laxity.

Thus, given that agility is required in almost all sport and significantly influences athletic performance, it is prudent to elucidate how it varies across the menstrual cycle. In doing so, scientists and strength coaches can more readily delineate when to best conduct agility tests and effectively prescribe individualized strength and conditioning paradigms. More importantly, cycle phase can serve as a predictor of athletic performance, and agility times can serve as an ancillary marker of increases in knee laxity (i.e., increased laxity may be paralleled by decreases in agility times, given that biomechanics is altered by lax tissues). The purpose of this investigation was to determine if agility is affected by the menstrual cycle, by conducting agility drills during MC and EF. The hypothesis was that agility performance will be hindered during MC, relative to EF, due to alterations in knee laxity, cognitive function, and biomechanical patterns.

CHAPTER II

REVIEW OF LITERATURE

The menstrual cycle of the female reproductive system is marked by two distinct phases: the considerable inter-individual variability of the follicular phase (FP) and the luteal phase (LP), which is usually 12-14 days in length (Practice Committee of the American Society for Reproductive Medicine [PCASRM], 2012). Lower mean resting plasma concentrations of E and PG during the FP differentiates the aforesaid phases. Further, the E peak occurs during the latter portion of the FP and there are a diverse group of estrogens: 17 β -estradiol (E₂), estrone, and estriol, which are formed by androgenic precursors (Coelingh Bennink, 2004); E₂ is the principal E and PG peaks roughly 6-8 days post-ovulation (PCASRM., 2012). From a physiological perspective, the phases serve as intermediates to one another and their temporal nature is marked by the development of the dominant follicle in the FP and an active corpus luteum in the LP (American College of Sports Medicine [ACSM], 2006). Specifically, the FP denotes the time from menses to ovulation, whereas the LP is defined as the interval from ovulation until the onset of the next menses. Each phase is arbitrarily defined as being 14 days in duration; however, this can

fluctuate based on habitual physical activity, use of oral contraceptives (OC), and energy consumption. For example, women who adhere to an exercise regimen frequently display an elongated FP and a condensed LP (Widmaier, Raff, & Strang, 2011). There is also a high prevalence of anovulation in women who are recreationally active. However, maintenance of luteinizing hormone (LH) pulsatility and ovarian function is related to energy availability rather than the stress of exercise (Loucks & Thuma, 2003; Loucks, Verdun, & Heath, 1998). Subsequently, consuming an adequate energy intake (i.e., 30kcal/kg of lean body mass), even in the face of heightened levels of energy expenditure, preserves ovarian function (Loucks & Thuma, 2003).

2.1 Effects of the Menstrual Cycle on Substrate Kinetics

With the aforementioned changes in circulating hormones, the menstrual cycle affects a myriad of physiological parameters, which could in-turn affect athletic performance (Janse de Jonge, 2003; Lebrun, 1993; Shultz, et al., 2012b). For instance, absolute and relative maximal oxygen consumption (Vo_{2max}) are significantly lower during the LP, relative to the FP (Lebrun et al., 1995). Similarly, endogenous hormonal perturbations during the menstrual cycle have been shown to alter substrate utilization during aerobic physical activity, such that free- fatty acid (FFA) oxidation is heightened during ovulation and/or the LP (Ashley, Bishop, Smith, Reneau, & Perkins, 2000; Bonen, Haynes, & Graham, 1991; Casazza, et al., 2004; Devries, Hamadeh, Phillips, & Tarnopolsky, 2006; Suh, Casazza, Horning, Miller, & Brooks, 2003). However, the results were marred by confounding variables. For instance, Devries et al. (2006) included women on triphasic OC and subjects not on OC; relative to OC subjects, women not on OC had higher circulating concentrations of E2 and PG during the

LP and elevated levels of E2 and T during FP, but no differences were noted between the groups, with regards to carbohydrate storage and utilization. Given that the aforementioned study combined subjects who were and weren't on OC into one group, the results from this study should be interpreted with caution. On the other hand, longitudinal and/or well conducted trials that controlled for confounding variables (e.g., food intake, which alters substrate utilization) have demonstrated no deviations in substrate utilization during the menstrual cycle (Horton et al., 2006; Horton et al., 2002; Jacobs et al., 2005; Smekal et al., 2007).

2.2 Effects of the Menstrual Cycle on the Anabolic Response Post-Exercise

Additionally, the anabolic stimulus post-exercise may be affected during the LP, as demonstrated by increased growth hormone (GH) levels in response to a resistance training bout (Nakamura, Aizawa, Imai, Kono, & Mesaki, 2011). Enea et al. (2009) observed increases in T and dehydroepiandrosterone after a prolonged exercise session and Wingate test, but no influence of menstrual cycle phase was observed among the cohort. However, the results were confounded by predetermined test timing and E and PG levels were not assayed. T concentrations are elevated during ovulation and, therefore, may play a role in strength development (Phillips, Rook, Siddle, Bruce, & Woledge, 1993; Rutherford & Jones, 1992; Sarwar, Niclos, & Rutherford, 1996; Shultz, Sander, Kirk, & Perrin, 2005). However, Elliot et al. (2003) did not note a difference in the concentration of testosterone ([T]) during the menstrual cycle and no corresponding alterations in isometric maximal voluntary contraction (MVC) in the first dorsal interosseous muscle were observed. Although unlikely, fluctuating E and PG levels may play a role in the expression of muscular strength and, therefore, agility in women; differences in E receptor mRNA levels, and higher E concentrations could

attenuate E receptor downregulation (Devries et al., 2006; Wiik et al., 2003). PG could counteract the effects of E on MVC, but in postmenopausal women where the concentration of PG is low, muscle weakness is still present (although muscle weakness can be attributed to the effects of sarcopenia) (Phillips, Rook, Siddle, Bruce, & Woledge, 1993; Rutherford & Jones, 1992; Sarwar, Niclos, & Rutherford, 1996; Shultz, Sander, Kirk, & Perrin, 2005). Elliot et al. (2003) also notes that bioavailable concentrations of free and albumin-bound E2 fractions did not change throughout the cycle even though total E2 levels fluctuated. This indicates that E remains bound to sex hormone binding globulin (SHBG) and is, therefore, unlikely to influence acute muscle strength.

2.3 Mediators of Agility

Although additional research is needed to delineate if fluctuating hormones influence substrate utilization and the anabolic milieu post-exercise, its effect on agility may be significant, especially if it affects mediators of agility. Agility has been traditionally used as an umbrella term that denotes any swift alteration in body positioning during athletic competition. Whether the deviation in body position is preplanned, influenced by auditory stimuli (firing a gun during a 100-meter sprint), or by a visual stimulus is irrelevant (Sheppard & Young, 2006a). Herein, the definition proposed by Sheppard et al. (2006a) will be utilized to describe agility, which is denoted by a swift change of direction as a result of an external stimuli. Furthermore, an emphasis is also placed on open-skill movements, which they describe as being a countermovement that is influenced by a non-rehearsed stimulus (e.g., evading an oncoming linebacker in football).

The determinants of agility are vast (strength, power, anticipation, recognition, reaction time, body composition, stature, etc.), and an explanation of each modulator of agility would be impractical. Therefore, this review will be confined to the most pertinent regulators of agility. There is some ambiguity as to whether sprinting correlates with indexes of agility (Sekulic, Spasic, Mirkov, Cavar, & Sattler, 2013; Tsitskarsis, Theoharopoulos, & Garefis, 2003). The dichotomy between Tsitskarsis et al. (2003) and Sekulic et al. (2013) with regards to the correlation between speed and agility could be the result of divergent testing modalities. Sekulic et al. (2013) utilized predetermined change of direction points (e.g., T-Test) whereas Tsitskarsis et al. (2003) employed more sport specific movements. Further, Young et al. (2002) state that test specificity influences the degree to which two measures are correlated. For example, a higher correlation between depth jump height and change of direction is present because of the similar neural discharge patterns.

In relation, strength and power influence agility more readily in sports that require changes of direction in a more confined area (e.g., racquetball court); the association is curtailed as the distance required to execute the change of direction movement is increased (e.g., soccer player trying to move past a defender over a 10 yard distance) (Sheppard et al., 2006a). Congruently, Sekulic et al. (2013) noted a strong correlation between power and agility. A more pronounced relationship was noted between leg reactive strength (stretch-shortening cycle) and change-of-direction performance, but concentric leg extension power did not demonstrate a significant relationship (Young, James, & Montgomery, 2002).

Furthermore, cognition (reaction speed, recognizing movement cues) are undervalued components of agility in the sporting community. Processing information and speed of reaction are integral in sports such as football, where an offensive player must ascertain the

opposing players speed, position, and their intersection prior to initiating a change of direction movement (Sheppard et al., 2006a). It is plausible that impairments in cognition may result in differentiated agility times during the menstrual cycle because it may affect cognitive function (Rosenberg et al., 2002; Wolf et al., 1999). Even minute deficiencies in mental processing can have drastic implications during athletic competition. For example, an inability to recognize movement cues of an opposing football player can result in an inability to evade an oncoming defender. No study has investigated if cognitive function can significantly alter agility times in open skilled tests (e.g., RAT). Likewise, no information is present as to how the menstrual cycle can impact reaction time and recognition. Rosenberg et al. (2002) only measured how the menstrual cycle influences verbal memory, which has no bearing on athletic performance. However, verbal memory, reaction time, and recognition skills constitute cognitive performance. Therefore, an impairment in verbal memory during EF may also be paralleled by impaired reaction time and recognition. Given that agility is affected by a wide array of variables, any performance decrements during the menstrual cycle in any of the aforementioned mediators of agility would impede an athlete's ability to change direction.

A more in depth review of indices of strength and neuromuscular control during the menstrual cycle is warranted; hamstring eccentric and concentric isokinetic force have been shown to correlate with timed agility tests, with a more significant correlation ($r = .58$) being noted in the former (Anderson et al., 1991). Eccentric forces generated during isokinetic testing would likely predict timed agility runs to a greater extent because eccentric movements are involved during deceleration, which is prevalent in agility tests (Sheppard et al., 2006a).

2.4 Effects of the Menstrual Cycle on the Neuromuscular System

Trials that favor an effect of menstrual cycle phase on neuromuscular control and/or indexes of strength are confounded by methodological shortcomings. In eumenorrheic subjects not on OC (n=10), a study found that isometric quadriceps MVC and handgrip strength were highest during MC, defined as day 12-18 of the menstrual cycle, was more fatigable, and had a longer half relaxation time from twitch and fused tetanus (denotes the ability of the sarcoplasmic reticulum to sequester calcium) (Sarwar et al., 1996). The cohort was tested during two full cycles, during five different phases (i.e., EF, between days 1-7; mid-follicular (MF), between days 7-12; MC, between days 12-18; ML, between days 18-21; and late luteal (LL), between days 21-32). A Jamar hydraulic hand dynamometer, conventional strength testing chair, stimulated contractions, and an adapted Burke protocol quantified grip strength, quadriceps MVC (percutaneous twitch superimposition [PTS] was utilized), contractile properties, and fatigability, respectively. Although the researchers studied 20 total subjects across two cycles, the data from this study lacks validity because the researchers did not assay hormonal levels and instead estimated cycle phase by counting backward from onset of menses. Thus, the study could have been confounded by anovulatory women and an inaccurate interpretation of cycle phase. Also, in trained (n=10) and untrained (n=12) women who were not on OC, researchers noted significant increases in muscle force values of the adductor pollicis muscle (AP) during the FP and a decline in maximal voluntary force (MVF) during ovulation (Phillips, Sanderson, Birch, Bruce, & Woledge, 1996). The trained populations were members of a rowing club, while the untrained subjects were largely sedentary. MVF measurements were ascertained by placing a transducer between the thumb and index finger and adducting the thumb for six to nine maximal contractions.

However, while the researchers detected ovulation via basal body temperature (BBT) and LH urinary kits and measured E2 levels, they failed to utilize PTS during the measurement of MVF. Consequently, they were not able to ascertain if the subjects exerted maximal effort. Moreover, a study noted that the muscle stretch reflex response of the rectus femoris was lowest during the periovulatory phase, which is an indicator of neuromuscular control (Casey, Hameed, & Dhaher, 2014). Although, as with the antecedent studies, the results are confounded by several design flaws: the authors did not utilize urinary LH strips; PTS was not incorporated; and it was not a longitudinal study (given that they assessed OC and non-OC subjects separately). Lastly, motor unit firing frequency was higher for the vastus medialis in relation to the vastus medialis oblique, and there was a phase effect such that firing rates were more pronounced in the LL phase in comparison to the EF phase (Tenan, Peng, Hackney, & Griffin, 2013). However, endogenous hormonal levels were not assayed and the authors did not use PTS during MVC contractions.

Other studies have failed to note a difference in muscular strength during the course of the menstrual cycle (Dibrezzo et al., 1991; Elliot et al., 2003; Fridén et al., 2003; Janse de Jong et al., 2001; Lebrun et al., 1995). Contemporary evidence has indicated that the neuromuscular system is not affected by the menstrual cycle (Abt, et al., 2007; Hertel, Williams, Olmsted-Kramer, Leidy, & Putukian, 2006). Excluding Hertel et al. (2006) who predicted the day of ovulation, well-conducted trials provide credence that any deviation in timed agility tests during the menstrual cycle would be independent of deviations in neuromuscular performance, across the menstrual cycle. Consequently, perturbations in T, E, and PG do not appear to affect muscular strength.

2.5 Effects of the Menstrual Cycle on Joint Laxity

Although knee sprains can be significantly influenced by a multitude of variables (i.e., sexual dimorphism in strength and morphological characteristics, etc.) (Hewett, 2000), knee joint laxity and the corresponding knee injury rate (e.g., ACL injuries) is also largely affected by endogenous fluctuations in E. Specifically, heightened physiologic levels of E2 induce reductions in fibroblast proliferation and collagen synthesis (Liu, Al-Shaikh, Panossian, Finerman, & Lane, 1997; Yu, Liu, Hatch, Panossian, & Finerman, 1999; Yu, Panossian, Hatch, Liu, & Finerman, 2001), while PG has an antagonistic effect. Thus, possible perturbations in agility performance during the menstrual cycle may also be paralleled by an increased injury risk. For example, if agility performance is hindered MC, when knee injuries are most prevalent, then strength and conditioning programs can reduce the workload and/or intensity during this isolated time period, to reduce the risk of injuries.

Trials that either failed to measure hormonal levels or conducted testing at arbitrary time points have demonstrated either increased knee joint laxity during the LP or no deviations. In a smaller sample size ($n=7$, age 26.9 ± 4.2 -years), subjects were tested during predetermined time intervals (i.e., day 1, 10-13, and 20-23). As a consequent, hormonal concentrations may be significantly varied, as peak levels may be attained at different time points (e.g., ovulation may occur on day 14 or 15). Laxity measurements were attained via the KT-200 knee arthrometer (KTNM). The results indicated that ACL laxity was greatest during phase II (i.e., days 10-13) and III (i.e., days 20-23), relative to phase I (i.e., day 1), with the highest values occurring during phase III (Heitz, Eisenman, Beck, & Walker, 1999). Similarly, in a recreationally active cohort ($n=70$, age 21.5 ± 2.6 -years), Shultz et al. (2010) noted significant differences in anterior knee laxity between the fifth and eighth luteal days,

such that they were higher during the former. The aforementioned days would correspond to about phase III in Heitz et al. (1999). Concurrently, genu recurvatum (i.e., degree of knee hyperextension) and general joint laxity were significantly higher during the fifth luteal day, relative to the first day of menses. Unsurprisingly, the magnitude of cyclical changes were non-uniform among the cohort. In contrast, others have observed no significant difference in knee joint laxity across the menstrual cycle, among adolescent subjects ($n=7$, age 16.3 ± 0.65 -years) (Eiling, Bryant, Petersen, Murphy, & Hohmann, 2007).

A few well-conducted trials have corroborated Eiling et al. (2007). In one study, 12-eumenorrheic subjects were tested during the first day of menses, near ovulation, and at day 23 of their menstrual cycle (i.e., midluteal). Luteinizing ovulation strips were utilized, to ascertain when the subjects ovulated. Furthermore, knee joint laxity was evaluated with the KTNM and radiographic measurements. The results indicated that although there were normal plasma fluctuations in T, E2, PG, LH, and follicle stimulating hormone (FSH), there was no significant difference in joint laxity between menstruation, ovulation, and midluteal phases (Van Lunen, Roberts, Branch, & Dowling, 2003). Likewise, others have noted no phase effect on knee joint laxity, but found that increases in knee joint laxity were associated with high knee joint loads which may precipitate injury (Park, Stefanyshyn, Ramage, Hart, & Ronsky, 2009a).

Dichotomously, other well-designed trials have favored an effect of phase on joint laxity. For instance, not only was knee joint laxity greater at ovulation, relative to the LP, but it was paralleled by increased knee joint loads (Park, Stefanyshyn, Ramage, Hart, & Ronsky, 2009b). The aforesaid study recruited 25-subjects with a mean age of 22.7 ± 3.5 -years. Moreover, in a similar cohort, although no differences were found in knee stiffness across the

menstrual cycle, knee laxity was highest during ovulation and the LP (Deie, Sakamaki, Sumen, Urabe, & Ikuta, 2002). In relation, others have noted both increases in knee laxity and reductions in stiffness during ovulation (Park, Stefanyshyn, Loitz-Ramage, Hart, & Ronsky, 2009c). Others have substantiated these findings by stating that anterior knee laxity, general joint laxity, and genu recurvatum were potentiated during the early luteal (EL) phase (Shultz et al., 2011).

The best empirical trials have confirmed that knee laxity is affected by the menstrual cycle, as they measured knee laxity and endocrine parameters daily. For example, 22-eumenorrheic females and 20-males were tested on the KTNM (Shultz et al., 2005). Serum E2, PG, and T were ascertained daily across one complete cycle for females, whereas the males were tested once per week for 4-weeks. Due to the divergent cycle lengths, the data was superimposed to when there was a similar hormonal milieu for all the female subjects; thus, allowing for the comparison of joint laxity and stiffness across similar hormonal concentrations and not during specific days of the menstrual cycle (e.g., comparing joint laxity during day 11 of the menstrual cycle when E levels may vary significantly across each subject). Although there were no differences in knee stiffness across or between the sexes, on average, females displayed greater laxity during the entirety of the menstrual cycle, relative to males. However, the values were not statistically significant during menses. Furthermore, within females joint laxity was greatest during ovulation, EL, and LL, with the highest values occurring during ovulation and EL (Shultz et al., 2005). Thus, the increased laxity in females was paralleled by the surge in E during MC and PG during EL. Congruently, in a similar design, nonathletic subjects (n=25, age 18 to 30-years) were tested daily while also conducting hormonal assays for E2, PG, and T (Shultz, Kirk, Johnson, Sander, & Perrin,

2004). Including the effect of time, the aggregate of E2, PG, and T and their interactions explained 63.3% of the variability in knee joint laxity. On average, the association was greater when variations in the hormonal environment were compared with alterations in knee laxity, which were taken 3-4 days after the assays. Thus, a latency period existed between the fluctuations in the hormonal milieu and variations in knee joint laxity (Shultz et al., 2004). Concurrently, knee laxity that is heightened during ovulation or the EL phase may necessitate lower and higher concentration of E and PG at the outset of the menstrual cycle, respectively; this improves the sensitivity of the collagen tissues to peak E and T levels (Shultz, Gansneder, Sander, Kirk, & Perrin, 2006).

Therefore, there does appear to be increases in knee joint laxity during ovulation and/or shortly thereafter, which is significantly mediated by perturbations in endogenous hormones. Further, relative to males, laxity is heightened in the frontal (varus/valgus) and transverse (internal/external rotation) planes, while no difference is noted in the sagittal plane (flexion/extension) (Shultz, Pye, Montgomery, & Schmitz, 2012c). More so, the aforementioned were correlated with lower-limb lean mass (LLLM). However, there seemed to be significant inter-individual variability in the magnitude and timing of hormonal fluctuations and subsequently specifically, when laxity is altered. A delayed response to peak hormonal levels and/or the degree to which tissues are responsive to increases in endocrine parameters are the most plausible factors that explain the inter-individual increases in knee joint laxity. Elucidating when a subject will experience increases in knee joint laxity is possible when assays and knee laxity measurements are conducted daily.

2.6 Effects of the Menstrual Cycle on Joint Stiffness

Similar to joint laxity, stiffness has also been demonstrated to be affected by the menstrual cycle; in comparison to men, this is especially evident in the frontal and transverse planes and is associated with LLLM (Shultz et al., 2012c). In part, this has been demonstrated to be mediated by perturbations in hormones, as there was a negative correlation between E2 and ACL stiffness ($r=-0.80$), whereas PG and estriol ($r=0.70$, $r=0.66$, respectively) were positively correlated with ACL stiffness (Romani, Patrie, Curl, & Flaws, 2003). The aforesaid occurred during ovulation. Park et al. (2009a) observed significant reductions in knee stiffness during ovulation, in relation to the LP. Musculotendinous stiffness was significantly lower during the ovulatory phase, in relation to the first day of menses and day seven of the FP (Eiling et al., 2007). Moreover, Shultz et al. (2011) noted that varus-valgus stiffness was lower in females and was significantly reduced during the EL phase, in comparison to menses. The aforementioned information is pertinent because attenuations in stiffness may predispose female athletes to injurious situations (Eiling et al., 2007). Furthermore, stiffness is reduced during the same time interval that laxity is increased, which would accenuate an athletes risk to injuries. Interestingly, stiffness and laxity are perdominately affected in the frontal and transverse planes (Shultz et al., 2012c). Ergo, adduction, abduction, and rotational kinematic movement patterns should be limited during MC.

2.7 Effects of the Menstrual Cycle on the Prevelance of Injuries

Empirical evidence fails to demonstrate an increased prevelance of injuries during MC, when laxity and stiffness are increased and attenuated, respectively. In a prosepctive

study, male and female handball players were followed for one year, and although females sustained more ACL injuries (23 females vs. 5 males), they occurred at divergent time points during their menstrual cycle (Myklebust, Maehlum, Holm, & Bahr, 1998). Of the 23 injuries, all were of the non-contact variety and occurred while running at full speed or during cutting maneuvers (Myklebust et al., 1998). Specifically, five occurred during the menstrual phase, two in the follicular phase, one in the EL phase, and nine in the LL phase. However, hormonal assays were not conducted and the phases were defined arbitrarily. Moreover, eight-women were on OC. Therefore, interpretation is limited. In relation, a retrospective study observed a greater incidence during ovulation (Wojtys, Huston, Lindenfeld, Hewett, & Greenfield, 1998).

2.8 Oral Contraceptives and Athletic Performance

In a similar manner to eumenorrheic non-OC adherent subjects, there has been conflicting data with regards to the effects of OC on a plethora of physiological parameters. Exogenous administration of synthetic hormones, in the form of a mono or triphasic OC regimen, has been demonstrated to homogenize hormonal levels, such that there are non-significant differences across the phases of the menstrual cycle (Bell, et al., 2011; Redman & Weatherby, 2004). Moreover, circulating endogenous hormonal levels are significantly attenuated, relative to non-OC users (Devries et al., 2006). Therefore, the aforesaid hormonal milieu may negate the effects of the menstrual cycle, as hormonal levels remain fairly static. However, synthetic exogenous hormones are elevated (Ekenros, Hirschberg, Heijne, & Fridén, 2013), which are able to exert the same physiological stimulus as non-synthetic isoforms. Subsequently, elucidating how synthetic hormones influence athletic performance is needed.

As such, evidence that substantiates the notion that OC negate the effects of the menstrual cycle are abundant. Bell et al. (2011) noted non-significant changes in muscle stiffness and hamstring neuromechanics. Concurrently, Sarwar et al. (1996) and Phillips et al. (1996) observed no cyclical changes in MVC and relaxation times from twitch and fused tetanus; Ekenros et al. (2013) documented no variations in muscle strength and power, in subjects on triphasic OC. Given that the menstrual cycle (i.e., non-OC) does not impact indices of strength, it is unlikely that OC influence strength across the menstrual cycle. Similarly, triphasic OC had no measurable effect on substrate utilization (Jacobs et al., 2005). However, in women on OC, anaerobic performance was significantly greater on days 26-28 of three consecutive menstrual cycles (Redman & Weatherby, 2004). Additional insight into the effects of exogenous hormones in anaerobic-based sports is needed.

CHAPTER III

METHODS

The trial received IRB approval and prior to the outset, subjects signed the informed consent form and completed the AHA/ACSM symptoms and menstrual cycle history questionnaire (See Appendix A-D). Prerequisites included a healthy body weight (BMI <25 kg/m²), normal menstrual cycle length, no oral contraceptive use within the prior six-months, and no orthopedic limitations.

3.1 Study Design

A causal-comparative study that investigated the effects of the menstrual cycle (independent variable) on agility (dependent variable) recruited eumenorrheic subjects (n=10) from Cleveland State University, to participate in a two-month study. Prior to the outset of the trial, subjects were familiarized with home-based urinary LH kits. During one month prior to and during the month of agility testing (i.e., second month) they completed the symptoms questionnaire, which consisted of menstrual cycle-related symptoms (e.g., breast tenderness, fluid retention, etc.). On a daily basis, the subjects

input on the questionnaire whether they experienced the aforementioned symptoms. Also, during the allotted two-months on days 7-20 of their menstrual cycle, they utilized urinary LH kits to indirectly estimate the time of ovulation. LH kits more accurately predict ovulation, relative to BBT (Behre, 2001; Guermandi, et al., 2001). Ovulation occurs roughly 18-hours after the LH surge (Widmaier et al., 2011).

All subjects completed testing during EF on days one through four of menses. Subsequent testing during MC was specific to each individual, as the estimated day of ovulation was predicated on the presence of a positive LH strip. During each session, the subjects were verbally and visually (i.e., the test administrator conducted each test) familiarized with the testing protocols. To improve objectivity, the administrator tested each subject. The subjects were instructed to wear the same shoes during each testing session, and the test administrator wore the same attire throughout the trial. Subjects were instructed to refrain from exercising 24 hours prior to the day of testing because the hormonal milieu can fluctuate after exercise (Frankovich & Lebrun, 2000). Similarly, subjects abstained from alcohol and caffeine intake during the allotted time. On entry into the lab, height and weight were measured on a wall-mounted stadiometer and physician scale, respectively. Afterwards, each subject conducted a dynamic warm-up for 10-minutes, which consisted of five-minutes of light jogging and five-minutes of dynamic stretching, before the agility tests. The dynamic stretches were comprised of grapevines, shuffles, forward leg swings, and an external hip rotation stretch. Afterwards, they completed the T-Test and RAT sequentially; the order of which was counterbalanced. Congruently, to control for any order effects, counterbalancing was utilized when testing women during the two phases of the menstrual cycle. Testing was conducted in an indoor

testing center to minimize the effects of air density and wind velocity (McArdle, Katch, & Katch, 2009).

3.2 Reactive Agility Test

The RAT was selected for its ability to measure changes of direction and cognitive factors (e.g., anticipation, decision making speed, and decision making accuracy) (Sheppard, Young, Doyle, Sheppard, & Newton, 2006b). The RAT encompasses motor movements that are not pre-planned (i.e., open skills), which distinguishes this protocol from traditional closed skill agility tests (e.g., T-Test; movements that are preplanned). Therefore, ascertaining whether the menstrual cycle can impair cognitive function is more readily accomplished with open-skilled tests than closed skill tests. Test retest intra class coefficient (ICC) for the RAT was 0.878 (Sheppard et al., 2006b). Similarly, reliability testing demonstrated a strong correlation between pre- and post-agility run times that were separated by seven-days ($r=0.91$), which indicates no learning effect (Veale, Pearce, & Carlson, 2010).

During the RAT (See Appendix E), the subjects emulated the sport specific movement patterns of the tester. The teste administrator was situated in proximity to a motion sensor while the subjects stood five-meters directly across. Directly parallel to the subject and the tester (five-meters to the left and right) were two timing gates. The test commenced once the tester initiated the movement and stepped away from the sensor. The subjects then reacted to the movement patterns of the test administrator and executed the movements to the best of their ability. The time was stopped when the subject passed the timing beam on the left or the right. For example, when the test administrator moved

forward and to the left, the subject would have, likewise, performed the same movement patterns until the beam on the left was triggered. As presented by Sheppard et al. (2006b), each movement was executed twice and the fastest time for each movement pattern was utilized. The movement patterns were comprised of:

- 1) Step forward with the right foot and change direction to the left.
- 2) Step forward with the left foot and change direction to the right.
- 3) Step forward with the right foot, then left, and change direction to the right.
- 4) Step forward with the left foot, then right, and change direction to the left.

The test administrator, however, was instructed to perform all of the aforesaid movements in each testing session. The order of which was randomized for each session.

3.3 T-Test

In contrast, the T-Test utilizes four cones that are dispersed in the shape of a “T” (See Appendix F). A motion sensor was placed near the right foot of each subject and a timing gate was placed 0.56-meters behind the subject. The subject initiated the test at cone A and sprinted to cone B, which was located 10-yards ahead and touched the base of the cone with their right hand. Thereafter, they shuffled five-yards to the left and, congruently, touched the base of cone C with their left hand. Afterwards, they shuffled to the right 10-yards and touched the base of cone D with their right hand. Once they initiated contact with cone D, they shuffled back to cone B and touched the base of the cone with their left hand. Finally, the subject backpedaled from cone B to cone A, and the time was stopped when the subject passed the timing gate. Criteria for disqualification

included failing to touch the base of the cone, overlapping the feet while shuffling, and failure to maintain a neutral head position throughout.

3.4 Statistical Analysis

Inferential statistics were used to assess treatment differences due to menstrual cycle phase on agility run times. Level of significance was set at $p < 0.05$. A per-protocol (PP) and intent-to-treat analysis (ITT) were conducted; ITT analysis included all subjects in the analysis post-randomization, while the PP analysis omits subjects who committed protocol violations. Differences between continuous variables were analyzed with a paired samples t-test and expressed in mean values (\pm standard deviation [SD]). Data was analyzed via Microsoft Excel (version 15.0.497.1003).

CHAPTER IV

RESULTS

Due to protocol deviations, an ITT and PP analysis were conducted. The latter resulted in the exclusion of two subjects. Specifically, subject seven was tested in a divergent setting, whilst the eighth wore different shoes during MC and EF. As depicted in figure one and table one, the PP analysis demonstrated that there was a significant difference in T-Test agility times between MC and EF (12.56 ± 0.90 -s vs. 12.3 ± 0.89 -s, $p=0.007$), while the ITT analysis did not note a significant difference (12.76 ± 1.15 -s vs. 12.89 ± 1.65 -s, $p=0.623$). For more robust conclusions to be drawn, mutual agreement, or at least a degree of similarity, between the forms of analysis should be apparent. This was observed with the RAT however; with both analyses noting non-significant differences.

As shown in table one, for PP and ITT analysis, no differences were noted in RAT agility times between EF and MC for any of the movements. Specifically, PP median values for RAT maneuvers 1-4 were not different across the phases.

For ten-subjects (BMI 22.4 ± 1.4 -kg/m²; weight 61.3 ± 5.6 -kg; age 24.2 ± 5 -years; height 165.58 ± 8.29 -centimeters), there was no significant difference ($p=0.49$) in weight

between EF (61.2 ± 5.5 -kg) and MC (61.4 ± 6 -kg). The mean cycle length was 28 ± 3 -days, and subjects experienced a LH surge on day 15 ± 2.8 . The mean LP length was 12.6 ± 3.4 -days. Age of menarche occurred at 13 ± 2.3 -years.

Table 1. The Effects of the Menstrual Cycle on Agility Times Attained on the RAT and T-Test

Protocol	Mid-Cycle	Early Follicular Phase	p-value
T-Test (P)	12.56 ± 0.90 -s	12.3 ± 0.89 -s	0.007
T-Test (I)	12.76 ± 1.15 -s	12.89 ± 1.65 -s	0.623
RAT 1 (P)	1.87 ± 0.090 -s	1.91 ± 0.13 -s	0.350
RAT 2 (P)	$1.97 \pm .015$ -s	1.96 ± 0.11 -s	0.807
RAT 3 (P)	1.99 ± 0.86 -s	2.04 ± 0.17 -s	0.374
RAT 4 (P)	2.15 ± 0.22 -s	2.06 ± 0.18 -s	0.258
RAT 1 (I)	1.92 ± 0.16 -s	1.95 ± 0.18 -s	0.438
RAT 2 (I)	2.01 ± 0.21 -s	2 ± 0.16 -s	0.616
RAT 3 (I)	2.05 ± 0.16 -s	2.05 ± 0.18 -s	1
RAT 4 (I)	2.18 ± 0.23 -s	2.11 ± 0.23 -s	0.295

I= intent-to-treat analysis (n=10); P=per-protocol analysis (n=8). p-values: Paired samples t-test. Values expressed with means and standard deviations. RAT, reactive agility test.

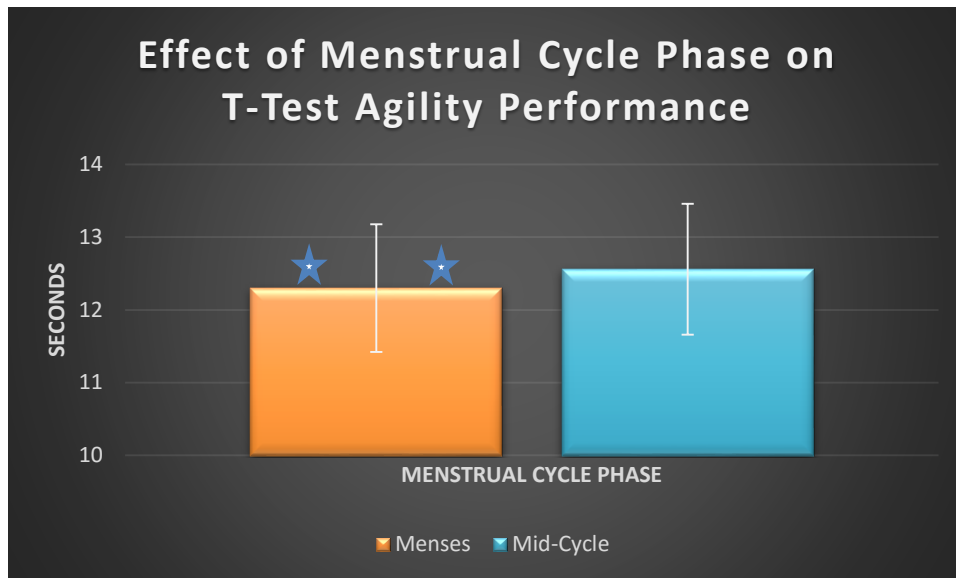


Figure 1. During a per-protocol analysis (n=8), a paired samples t-test was conducted to elucidate if there was an effect of menstrual cycle phase on agility performance. As depicted, subjects were significantly faster during menses (12.3 ± 0.89 -seconds), relative to mid-cycle (12.56 ± 0.90 -seconds). Values expressed as means and SD. ★=significant difference ($p=0.007$).

CHAPTER V

DISCUSSION

Given that subjects were significantly faster during menses in comparison to mid-cycle, the hypothesis was refuted. As such, the principal and novel outcome from this trial was that agility performance may be influenced by the menstrual cycle during agility tests that do not require mental processing. To the knowledge of the author, no empirical trial has investigated how the menstrual cycle influences agility. The results affirm that mental processing, when executing athletic maneuvers, may not be affected by fluctuations in E; given that agility times did not differ between RAT values attained during menses versus MC. This is in contrast to the noted effect on cognitive function at rest (Rosenberg et al., 2002; Wolf et al., 1999). The discrepant finding may be due to the diverse testing environment; the current trial being in an athletic setting, while that of the aforesaid was in the rested state. Moreover, the protocols were on the opposite ends of the spectrum, with respect to assessing cognitive function; Wolf et al. (1999) and Rosenberg et al. (2002) studied how verbal memory was influenced, while the present study elucidated if mental processing during agility drills was affected.

A possible explanation for the lack of a significant finding between agility times attained during MC versus EF during the RAT relates to the fact that it requires only one or two change of direction movements. In relation, the T-Test consists of four change-of-direction movements, along with accelerating, decelerating, shuffling, and back-pedaling motions. However, the neuromuscular system has not been noted to be affected by the menstrual cycle, which makes it unlikely that perturbations in E had an effect on strength, power production, deceleration, or acceleration (Dibrezzo et al., 1991; Elliott et al., 2003; Friden et al., 2003; Janse de Jong et al., 2001; Lebrun et al., 1995). Thus, the aforesaid did not likely contribute to deviations in T-Test times. Body weight also had a negligible contribution to agility times, as there were non-significant differences between EF and MC.

With respect to the endocrine system, potentiated levels of E could have hindered athletic performance while performing change of direction movements, as indicated by the T-Test. Dichotomously, PG may not have played a significant role, given that concentrations remain low during EF and MC, which is when agility testing was conducted. This would indicate that PG would not have been able to antagonize the actions of E (Romani et al., 2003), but sensitizing collagen to E during the MC surge may require requisite low and high levels of E and PG during menses, respectively (Shultz et al., 2006).

Corroborating the link between the menstrual cycle, knee laxity, and altered biomechanical patterns are made by several empirical trials: the menstrual cycle has been noted to affect knee laxity, which has been demonstrated to alter biomechanical patterns and possibly have a subsequent effect on agility performance (Shultz et al., 2012a; Shultz

et al., 2005). Indeed, the aggregate of E2, PG, and T explained 63% of the variation in laxity (Shultz et al., 2004). Subsequently, the association between laxity, an altered biomechanical profile, and agility is multifaceted. It is likely that a myriad of hormones and other innate factors contributed to the variance in agility performance. There is likely to be large inter-individual differences, given that the level and timing of surges in circulating endogenous hormones are widely different between subjects. Consequently, only a general time point can be pinpointed as to when laxity, biomechanical patterns, and agility performance appears to be most negatively affected: MC. This would correspond to ovulation and the early segments of the LP. However, it is equally likely that this may extend into the LL phase.

With respect to athletes, curtailing their exercise regimen during the aforesaid time points may be necessary, given that stiffness is also attenuated during MC (Shultz et al., 2012c). Therefore, for optimal reductions in injury risk and improved athletic performance, athletic competitions, if possible, may need to be conducted during the EF and LL phase. Congruently, intensity and/or volume of exercise may need to be curtailed during MC, as laxity and stiffness are heightened and attenuated, respectively. A deterrent to injurious situations would be to increase muscle mass (Shultz et al., 2012c), overall muscular strength, and correcting ipsilateral and contralateral strength deficits. This would not only entail prescribing the typical sport specific movement exercises, but also single-joint exercises (e.g., knee curls, hip abduction exercises, etc.), which are needed to correct muscular imbalances.

LP suppression (a truncated LP) is a possible marker of infertility and consequently attenuated levels of ovarian hormones (PCASRM, 2012). As indicated by

the results, there was no indication of luteal phase suppression and the subjects were noted to have normal menstrual function. The mean LP length was 12-days, but individuals with LP suppression usually have a LP length of < 10-days.

5.1 Limitations

The principal limitation was the omission of hormonal assays. Furthermore, conducting agility drills during ovulation is difficult to isolate because it represents a physiologically condensed time interval. More specifically, its duration is limited as it only peaks for approximately three days (Landgren, Undén, & Diczfalusy, 1980). Ultradian rhythm variations in hormones could skew effects of the menstrual cycle phase on agility times. Lastly, increased E levels may not immediately influence physiological parameters (Phillips et al. 1996). A finite sample size (n=8) limits the ability to make broad generalizations.

5.2 Future Considerations and Conclusions

There does appear to be an effect of the menstrual cycle on agility. Specifically, agility performance is hindered during MC, when the corresponding surges in E occur. This is likely the result of increases in joint laxity and its associated alterations in biomechanical patterns. Future trials should conduct hormonal assays and agility trials daily; measuring the effects of the menstrual cycle by only conducting trials during 2-3 time points during the cycle only gives a small representation of how fluctuations in endogenous hormones influence agility. As is frequently conducted, two to three snapshots of perturbations in hormones and the congruent effects on athletic performance are not sufficient to make conclusive inferences.

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APPENDIX

APPENDIX A

IRB APPROVAL

IRB protocol #30082-KUL

Kimberly A. Neuendorf <k.neuendorf@comcast.net>

Fri, May 2, 2014 at 1:14
PM

To: Karim Kheniser <kheniser.3@gmail.com>, e.kullman@csuohio.edu

Cc: Sheila M Patterson <s.m.patterson@csuohio.edu>, Bernidean R Strong
<b.r.strong@csuohio.edu>, k.neuendorf@comcast.net

Dear Researchers Kullman and Kheniser,

Thank you for your revisions, conveyed to me on 4/30/14 and 5/1/14 (the latter including a Recruiting Flyer that supplants the one sent on 4/30/14). The revised application, Informed Consent Form, and Recruiting Flyer satisfy all of our concerns. You may consider this email to be an initial approval of your protocol #30082-KUL, as expedited category 4. A hard copy confirmation will follow.

Best wishes for success in your research endeavors.

Sincerely,

Kim Neuendorf, Ph.D.

APPENDIX B

INFORMED CONSENT



INFORMED CONSENT FOR PARTICIPATION

Effects of Menstrual Cycle on Agility Performance

Introduction

Thank you for considering participation in this project. Professor Emily Kullman, faculty in the Department of Health and Performance, and Mr. Karim Kheniser, Graduate Student in the Exercise Science Program, Department of Health and Performance at Cleveland State University are inviting you to participate in a research study to be conducted in the Human Performance Laboratory at Cleveland State University, which is located in room PE59. The study is being conducted to complete Mr. Kheniser's master's thesis.

The purpose of this study is to observe the effects of menstrual cycle, and particularly estrogen levels, on agility performance.

There is some evidence to suggest that estrogen affects joint laxity, which may have negative influences on joint mobility, and subsequent agility performance during the days of the menstrual cycle when estrogen levels are low. This, in turn, may result in impaired sports performance, or potential injury.

By determining if low estrogen levels does in fact cause a decline in sports performance, and potentially put the athlete at greater risk for injury, we can begin to develop specific training programs aimed at improving agility performance throughout the menstrual cycle; thus, providing better recommendations regarding injury prevention among female athletes.

You will be asked to complete two exercise sessions as part of this study at the Cleveland

State University Human Performance Laboratory. Each session will consist of a brief warm-up, followed by two separate agility tests. One test will be the T-Test, which consists of a T-shaped running and shuffling pattern on a pre-determined course. The second test is the reactive agility test (RAT), during which you will be emulating the movement pattern of the tester, and we will require you to do this 4 times.

Procedures

Prior to completion of the agility tests, you will be asked to monitor your menstrual cycle for one complete cycle using a questionnaire related to your symptoms, as well as a home-based ovulation test kit, which monitors urinary excretion of Luteinizing Hormone (LH). The ovulation kits will be provided to you by the study team to complete in the convenience of your own home. We will use these tests to establish proper timing of the agility tests during your next cycle.

Once your menstrual cycle patterns have been determined, we will establish testing dates to correspond with low-estrogen, and high-estrogen time points during your following menstrual cycle. The low-estrogen trial will be performed 2-4 days after the start of your menses. The high-estrogen trial will be performed approximately 2-3 days before your established ovulation date based on the previous month's ovulation test kit and questionnaires. You will also be asked to come to the HPL for familiarization with the agility testing procedures prior to your study testing dates. Each HPL visit will require ~1 hour of your time, for a total of 3 hours throughout the study.

For each of the two study testing dates, you will be asked to come to the HPL, where a venous blood sample will be obtained to verify estrogen levels. Following the blood draw, you will perform a brief warm up consisting of 5 minutes of light jogging, and 5 minutes of dynamic stretching. A warm-up will be followed by the agility testing protocols, including the T-Test and the RAT. These tests will be performed in random order. The T-Test consists of a T-shaped running and shuffling pattern on a pre-determined course, and you will perform this test one time. The second test is the reactive agility test (RAT), during which you will be emulating the movement pattern of the tester, and we will require you to do this 4 times. Both of these tests will be timed to monitor agility performance, with the goal of completing these tests in the fastest time possible.

Risks and Discomforts

Risks associated with this study include muscle soreness from the completed agility tests. There may be joint discomfort at the ankle, knee, and hip due to the abrupt movements of the agility tests. There may be some discomfort in the arm in which the blood sample is taken from. Due to the nature of the agility tests, there is risk of tripping or falling. As a result of a fall, the attendant risks include: bone fractures, torn ligaments, muscle strains, joint sprains, bruises or joint dislocations. Furthermore, although each of these tests lasts ~10-20 seconds, an all-out effort will be required and in rare instances could lead to fainting, abnormal blood pressure, fatal heart rhythms, stroke or heart attack.

Benefits

The indirect benefits of the study are to help your understanding of how menstrual cycle hormone fluctuations may play a role in exercise performance. There are no guaranteed direct benefits, but this study may help you in knowing how to maximize your fitness level as well as educate you on the roles of hormones in athletic performance.

Confidentiality

To protect your privacy, your name will not be used in any document of the project. A number will be assigned to the subject in place of a name. The information, however, may be used for a statistical or scientific purpose with your right of privacy retained. Professor Emily Kullman and Karim Kheniser will be the only witnesses of the information being presented. Data will be stored in the Human Performance Lab PE60B in a locked filing cabinet.

Participation

I understand that participation in this project is voluntary and that I have the right to withdraw at any time with no consequence. I attest and verify that I have no known health problems that could prevent me from successfully participating in the interval protocol treadmill test. If I have any questions about the procedures I can contact Professor Emily Kullman at (216) 687-4854 or Mr. Karim Kheniser at (440) 796-0056.

I understand that if I have any questions about my rights as a participant, I can contact Cleveland State University’s Review Board at (216) 687-3630.

Participant Acknowledgement

The procedure, purposes, known discomforts and risks, possible benefits to me and to others have been explained to me. I have read the consent form or it has been read to me, and I understand it. I agree to participate in this program. I have been given a copy of this consent form.

Signature: _____

Date: _____

Witness: _____

Date: _____

APPENDIX C

SCREENING QUESTIONNAIRE

Name:

Date:

Age:

DOB:

At what age did you first begin menstruating?

Are you currently taking any medications? If yes, please note the name of the medication.

Section 1	Yes	No
1. Is your menstrual cycle generally shorter than 26 days?		
2. Is your menstrual cycle generally longer than 31 days?		
3. Do you experience irregular bleeding during your cycle?		
4. Do you ever miss periods or have changes in the frequency or duration of your menstrual cycle?		
5. Does stress make your menstrual cycle length more irregular?		
6. Has chronic stress, increasing age or exercise changed the regularity of your period or stopped your period altogether?		
7. Do you experience sharp, stabbing, or dull aching pains mid-cycle which is worse when you apply pressure?		

Section 2	Yes	No
1. Do your periods last less than 3 days?		
2. Do your periods last more than 5 days?		

Section 3	Yes	No
1. Have you been medically diagnosed with endometriosis?		
2. Have you been medically diagnosed with uterine fibroids?		
3. Have you been medically diagnosed with ovarian cysts?		
4. Have you been medically diagnosed with pelvic adhesions or masses?		
5. Have you been medically diagnosed with polycystic ovarian syndrome?		
6. Do you experience very heavy periods and/or acne?		

APPENDIX D

AHA/ACSM QUESTIONNAIRE

Asses your health needs by marking all true statements.

History: You have had:

- a heart attack
- heart surgery
- cardiac catheterization
- coronary angioplasty (PTCA)
- pacemaker/implantable cardiac defibrillator/rhythm disturbance
- heart valve disease
- heart failure
- heart transplantation
- congenital heart disease

Name:

Date:

Symptoms:

- You experienced chest discomfort with exertion.
- You experience unreasonable breathlessness.
- You experience dizziness, fainting, blackouts.
- You take heart medications.

Other health issues:

- You have musculoskeletal problems.
- You have concerns about the safety of exercise.
- You take prescription medication(s).
- You are pregnant.

If you marked any of the statements in this section, consult your healthcare provider before engaging in exercise. You may need to use a facility with a medically qualified staff.

Cardiovascular Risk Factors:

- You are a man older than 45 years.
- You are a woman older than 55 years or you have had a hysterectomy or you are post-menopausal.
- You smoke.
- Your blood pressure is $> 140/90$.
- You don't know your blood pressure.
- You take blood pressure medication.
- Your blood cholesterol level is > 240 mg/dl.
- You don't know your cholesterol level.

You have a close blood relative who had a heart attack before age 55 (father or brother) or age 65 (mother or sister).

You are physically inactive (ie, you get < 30 minutes of physical activity on at least 3 days per week).

You are > 20 pounds overweight.

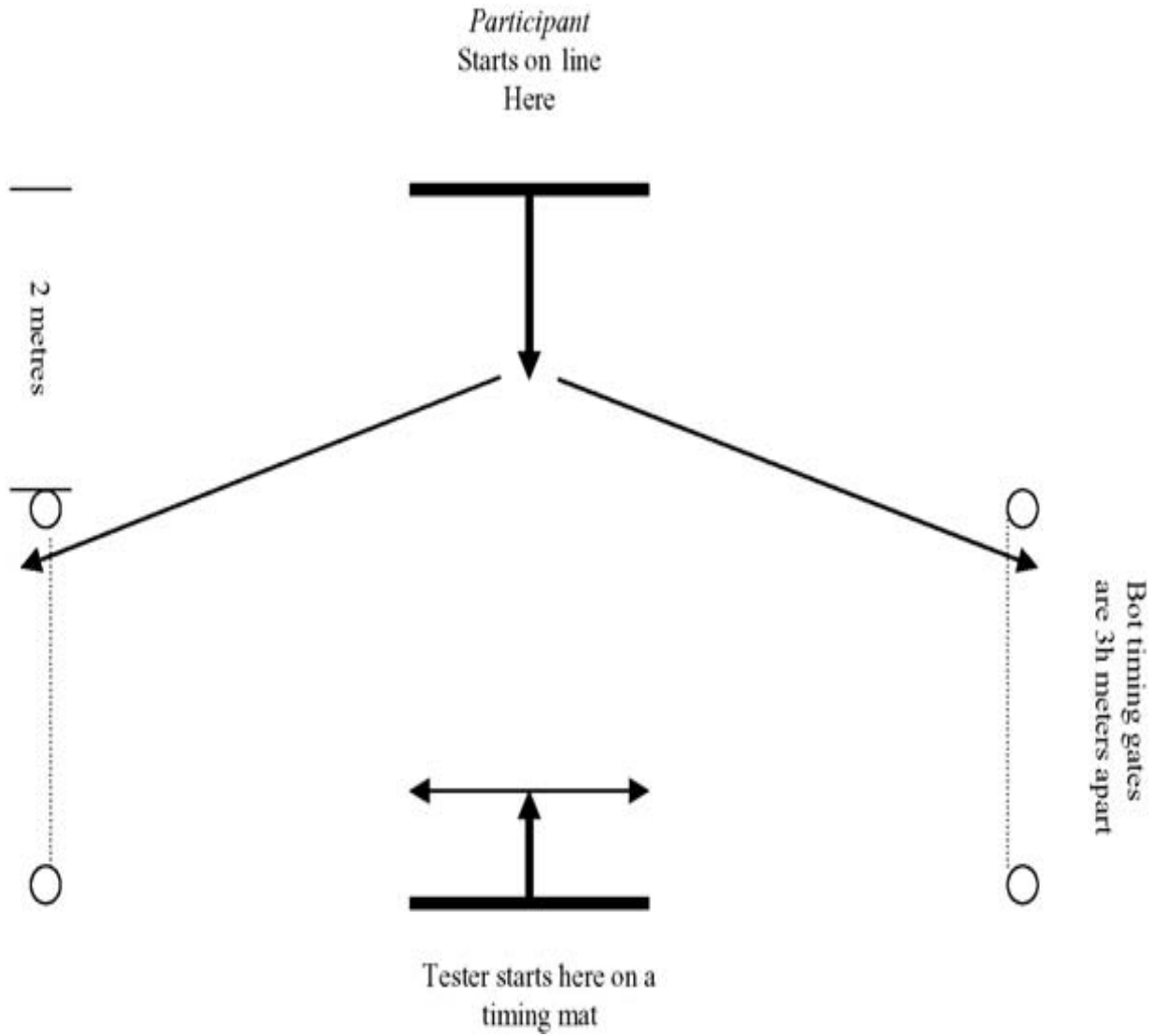
If you marked 2 or more of the statements in this section, consult your healthcare provider before engaging in exercise. You might benefit by using a facility with a professionally qualified exercise staff to guide your exercise program.

None of the above is true.

You should be able to exercise safely without consulting your healthcare provider in almost any facility that meets your exercise program needs.

APPENDIX E

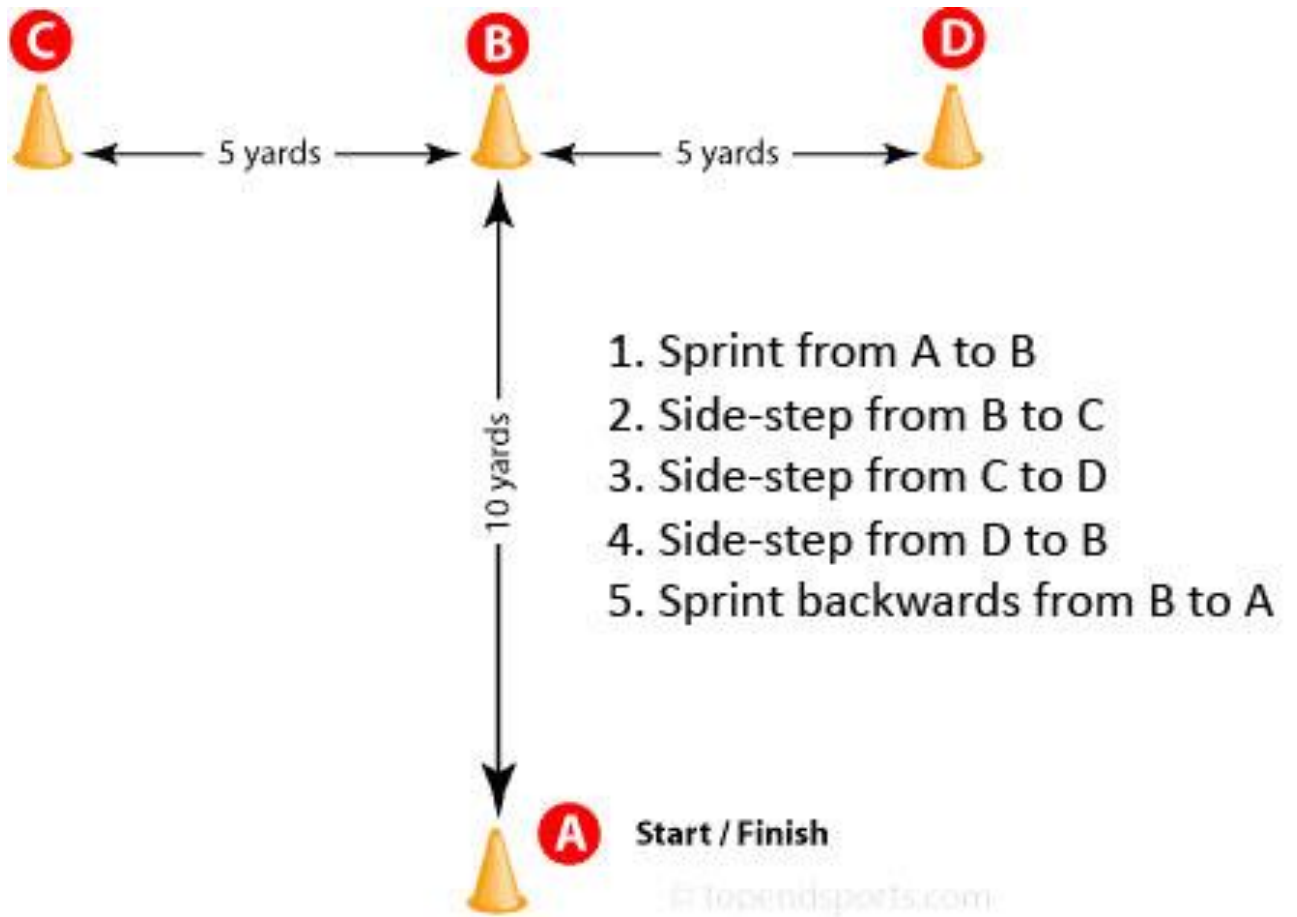
REACTIVE AGILITY TEST



(Sheppard et al., 2006b)

APPENDIX F

T-TEST



(Brady, 2013)

APPENDIX G

ABBREVIATIONS

ACL, anterior cruciate ligament;
E2, estradiol;
E, estrogen;
PG, progesterone;
T, testosterone;
GH, growth hormone;
EF, early follicular phase;
MC, mid-cycle;
RAT, reactive agility test;
LH, luteinizing hormone;
OC, oral contraceptives;
MVC, maximal voluntary contraction;
ML, mid-luteal phase;
FP, follicular phase,
LP, luteal phase;
PP, per-protocol;
ITT, intent-to-treat analysis;
LL, late luteal;
PTS, percutaneous twitch superimposition;
AP, adductor pollicis muscle;
MVF, maximal voluntary force;
KTNM, KT-200 knee arthrometer;
Vo₂max, maximal oxygen consumption;
ICC, intra class coefficient