The influence of age at menarche and hamstrings fatigue on knee biomechanics

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THE INFLUENCE OF AGE AT MENARCHE AND HAMSTRINGS FATIGUE ON KNEE BIOMECHANICS

A thesis submitted in partial fulfillment of the Requirements for the degree of Master of Science

By

STEPHANIE EILEEN FIELD
B.S., Furman University, 2013

2016
Wright State University
WRIGHT STATE UNIVERSITY

GRADUATE SCHOOL

November 17, 2016

I HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER MY SUPERVISION BY Stephanie Eileen Field ENTITLED The Influence of Age at Menarche and Hamstrings Fatigue on Knee Biomechanics BE ACCEPTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF Master of Science.

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ABSTRACT

Field, Stephanie Eileen. M.S. Department of Neuroscience, Cell Biology and Physiology, Wright State University, 2016. The Influence of Age at Menarche and Hamstrings Fatigue on Knee Biomechanics.

Anterior cruciate ligament (ACL) injury is 4-8 times more likely to occur in females than in males and is associated with significant morbidity. The mechanism underlying the gender disparity in non-contact ACL injuries is likely multifactorial, and may be influenced by a variety of risk factors. This study examines the influence of age at menarche and neuromuscular fatigue of the hamstrings muscles on knee biomechanics.

Twelve female athletes performed drop vertical jump (DVJ) tasks before and after a fatiguing protocol utilizing a glute-ham bench. Knee kinematics and kinetics were recorded during the contact phase of the DVJ. The results of this study found a significant decrease in knee abduction angle in the dominant leg after fatigue ($P \leq 0.05$), and a significant decrease in peak vertical ground reaction force (vGRF) after fatigue.

The findings of this study do not support our hypotheses that (1) age at menarche is correlated with greater dynamic knee valgus, (2) that knee valgus is greater after fatigue, or (3) that the effects of fatigue are exacerbated in females with earlier age at menarche. We concluded that because the glute-ham raise targeted fatigue of the lateral hamstrings (biceps femoris), the exercise caused a decrease in potentially harmful valgus
knee angles and moments. The reduction in peak vGRF after fatigue may be a result of an altered landing strategy in order to reduce impact forces and to maintain overall landing mechanics. Understanding the factors that affect knee biomechanics is critical for the development of more effective injury prevention strategies.
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For my right knee.
I. INTRODUCTION

The knee joint is the largest joint in the body, transmitting significant loads and sustaining high forces, particularly during athletic maneuvers (Nordin and Frankel, 2001). Thus, knee injuries are among the most common injuries in sports that utilize the lower limb (Price et al., 2004). The anterior cruciate ligament (ACL), which functions to stabilize the knee joint, is particularly susceptible to injury when forces at the knee joint are too large (Nordin and Frankel, 2001). The ACL can be injured via a variety of mechanisms, and there is a vast array of potential risk factors that increase the likelihood of sustaining an ACL injury. These risk factors have been the focus of many recent biomechanical and neuromuscular studies of the lower limb in order to improve preventative strategies for this devastating injury.

ACL injuries are common in sports, particularly those which involve sudden cutting movements, quick decelerations, or landing after a jump (Boden et al., 2010; Olsen et al., 2004). Rupture of the ACL is often associated with significant morbidity as a result of prolonged symptoms and increased incidence of osteoarthritis later in life (Campbell et al., 2014; Keays et al., 2010). Athletes can miss up to a year of competition while rehabilitating an ACL injury, and the fear of re-injury can be permanently detrimental to athletic performance. Additionally, the financial burden associated with ACL injuries can be significant, with conservative estimates at an average of $27,500 for
reconstructive surgery and rehabilitation costs, and even higher costs for long term treatments (Mather et al., 2013). Thus, elucidating the mechanisms and risk factors for ACL injury is critical in order to inform more effective and adaptable prevention strategies.

A gender disparity for the occurrence of ACL injuries has been documented throughout relevant literature, with females being 4-8 times more likely to experience an ACL injury than males playing the same sports (Agel et al., 2005; Arendt and Dick, 1995; Hewett et al., 2005; Hewett et al., 2006; Slauterbeck et al., 2002). Many studies have shown that female athletes often utilize high-risk biomechanics, such as cutting and landing from a jump with low knee flexion and increased valgus angles (Campbell et al., 2014; Gehring et al., 2009; Kernozek et al., 2005). The elevated occurrence of ACL injuries in females is likely multifactorial, with developmental, neuromuscular, biomechanical, and hormonal influences. Non-contact ACL injuries, or those which do not involve collisions or contact with other players or structures, comprise up to three quarters of all ACL injuries (Boden et al., 2000), and will be the focus of this study.

Knee Joint Anatomy, ACL Injuries and Mechanisms

The knee is a two-joint structure made up of the patellofemoral joint and the tibiofemoral joint, the latter of which can be further divided into medial and lateral condylar articulations. The knee is located between the two longest lever arms of the body, making it subject to high levels of loading and therefore particularly susceptible to injury. Knee motion occurs simultaneously in three planes, but most prominently in the sagittal plane, which allows for knee flexion and extension (Nordin and Frankel, 2001).
Motion in the transverse plane, or internal and external rotation, is limited, especially when the knee is extended. Motion in the frontal plane, or adduction and abduction, is also limited, particularly when the knee is extended (Nordin and Frankel, 2001). The tibiofemoral joint, which contains the ACL, is exposed to a variety of joint moments during dynamic activities. A joint moment refers to the rotational or bending action of a segment about an axis as a result of an applied force (Nordin and Frankel, 2001).

Knee ligaments are the primary stabilizers of the tibiofemoral joint, limiting motions such as anterior and posterior translation, valgus and varus angulation, and internal and external rotation (Nordin and Frankel, 2001). Anterior and posterior translation describes movement of the tibia relative to the femur within the sagittal plane (Behrens et al., 2013). Dynamic valgus refers to the combined angles and moments experienced at the knee joint in the frontal plane. It is defined as the position or motion of the distal femur toward, and the distal tibia away from, the midline of the body, while dynamic varus describes the opposite position or motion (Hewett et al., 2005). Dynamic valgus refers to a combination of motions and rotations at all three lower extremity joints, including hip adduction and internal rotation, knee abduction, tibial external rotation and anterior translation, and ankle eversion (Hewett et al., 2006). Throughout relevant literature and within this paper, the terms “knee valgus angle” and “knee valgus moment” can be used synonymously with “knee abduction angle” and “knee abduction moment”, respectively. Conversely, the terms “varus angle” and “varus moment” refer to adduction angles and moments, respectively. During dynamic sports maneuvers, excessive tibial
translation, valgus angles and moments, or tibial rotation can cause injuries to the knee joint, including rupture of the ACL.

Risk Factors for ACL Injury

Anatomical Factors

Numerous intrinsic factors have been associated with an increased risk of ACL injury. Anthropometric measurements, such as thigh length, tibia length, pelvic width, body mass index (BMI), and intercondylar notch width have been studied for potential correlations with injury occurrence (Hewett et al., 2006). General joint laxity has been of particular interest, as studies have found that females have increased general joint laxity relative to male counterparts. Joint laxity can significantly increase risk of injury by affecting sagittal knee motion (hyperextension) and frontal knee motion (valgus), both of which can strain the ACL (Boden et al., 2000; Ford et al., 2010; Hewett et al., 2005; Hewett et al., 2006; Myer et al., 2008; Uhorchak et al., 2003). Excessive anterior tibial translation relative to the femur, increased hamstring flexibility, and increased BMI are other intrinsic anatomical factors that have been associated with an increased risk of ACL injury (Boden et al., 2000; Hewett et al., 2006; Uhorchak et al., 2003; Wojtys et al., 1996).

Hormonal Factors

The influence of estrogen in females has been of great interest to researchers studying the causes and risk factors for ACL injury. Wojtys et al. (1998) observed a trend toward increased ACL injury during the ovulatory phase of the menstrual cycle when
estrogen levels are at their peak, and fewer injuries during the follicular phase when estrogen levels are lowest. High estrogen levels may decrease ligament strength, thus making the ACL more susceptible to injury (Hewett et al., 2006). Studies also suggest that estrogen affects neuromuscular function and control, altering muscle strength, anaerobic and aerobic capacity, and high-intensity endurance in female athletes (Hewett et al., 2006; Lebrun, 1994; Sarwar et al., 1996). In contrast to these findings, however, Slauterbeck et al. (2002) found that ACL injuries occurred most often during the first two days of menses when estrogen levels are low. These contradictory findings make it difficult to understand the true influence of estrogen on ACL injury risk.

**Age at Menarche**

Many studies have explored biomechanical and neuromuscular changes during pubertal maturation, which may contribute to the factors that underlie increased knee injury risk in females (Hewett et al., 2006). Recent studies have focused on age at menarche, or first menstruation, suggesting that earlier menarche can have detrimental effects on biomechanical and neuromuscular development (Froehle et al., 2016). The onset of menarche is associated with an increase in estrogen levels, which induces the cessation of accelerated pubertal bone growth (Seselj et al., 2012) and may also influence other aspects of biomechanical and neuromuscular maturation. Biomechanical maturation during puberty includes narrowing of gait base of support, which is the ratio of pelvic width to ankle spread (Sutherland et al., 1980), and which affects dynamic knee alignment (Froehle et al., 2013; Froehle et al., 2016; Vanwanseele et al., 2009). Earlier age at menarche is correlated with retention of a more immature, wider base of support,
leading to valgus dynamic knee angles and moments in the frontal plane (Froehle et al., 2016). Knee valgus upon contact with a surface is a key risk factor for ACL injury (Hewett et al., 2004), suggesting that earlier menarche increases ACL injury risk by affecting base of support and dynamic knee alignment.

In addition to anatomical factors, menarche has also been shown to influence neuromuscular development. Sensorimotor functions and neuromuscular coordination develop and mature during early adolescence, but in some cases, may undergo a regression (Quatman-Yates et al., 2012). Hewett et al. (2004) found that following the onset of pubertal growth, female athletes alter their landing biomechanics, which may be due to decreased neuromuscular control of the knee. Similarly, Ahmad et al. (2006) found that shortly after the onset of menarche, female athletes increase their quadriceps strength to a greater degree than their hamstrings strength, which can cause unsafe moments at the knee and increase risk of ACL injury. It is important to note that there is no documented evidence of a difference in rates of ACL injury between pre-pubertal males and females (Hewett et al., 2004). Ford et al. (2010), Hewett et al. (2004), and Quatman et al. (2006) all studied neuromuscular control of the knee during a jump landing throughout different stages of pubertal maturation. These studies showed that from the onset of puberty to post-pubertal stages, men and women demonstrate a divergence in neuromuscular control. During puberty, females have demonstrated decreased neuromuscular control of the knee and imbalances in muscular strength and coordination, whereas males improve balance in their quadriceps and hamstrings force production and overall neuromuscular control of the lower extremity (Ford et al., 2010; Hewett et al., 2004; Hewett et al., 2005;
Quatman et al., 2006). These findings suggest that changes in neuromuscular function and coordination coincide with the onset of puberty, but it has yet to be determined whether the timing of menarche affects this development in women.

**Biomechanical and Neuromuscular Factors**

Biomechanical and neuromuscular characteristics are among the most widely studied risk factors for ACL rupture as they more directly affect hazardous knee movements and positions. Many of these suggested risk factors have been correlated with the gender disparity predisposing females to ACL rupture more so than their male counterparts. For example, some studies suggest that differences in passive and active stability of the tibiofemoral joint could contribute to the gender disparity for ACL injury (Hughes et al., 2006). Passive stability of the knee joint depends largely on intrinsic factors, such as the laxity of ligaments and the geometry of articular surfaces. However, active stability relies on structural and mechanical factors such as patellar tendon-tibial shaft angle, muscle activity pattern, muscle reaction time, time to peak torque, and muscle stiffness (Behrens et al., 2013; Hughes et al., 2006).

Studies suggest that the hamstrings muscles act as important synergists to the ACL, helping to resist anterior tibial translation and valgus moments (Melnyk and Gollhofer, 2007). Struminger et al. (2013) found that subjects with higher activation of the hamstrings muscles experienced less valgus moments. Women have been found to have significantly slower generation of peak hamstrings muscle torque compared with men. They are considered to be "quadriceps dominant" in both strength and muscle-firing pattern (Ahmad et al., 2006). Quadriceps muscle contraction creates anterior shear force...
on the tibia relative to the femur, adding strain to the ACL without a balanced force from
the hamstrings (Ahmad et al., 2006). Co-contraction of the hamstrings muscles in
opposition to the quadriceps muscles can help protect the knee joint against high risk
movements, including anterior tibial translation and dynamic valgus (Hewett et al., 2006;
Palmieri-Smith et al., 2009). Thus, strength and activation deficits of the hamstrings can
greatly increase the risk of knee joint injury (Behrens et al., 2013; Gehring et al., 2009;
Tsepis et al., 2004).

Evidence also suggests that the position of semimembranosus and
semitendinosus, the medial hamstrings muscles, in particular helps them to resist tibial
shear forces and valgus moments by creating a varus moment (Lloyd et al., 2005;
Palmieri-Smith et al., 2008; Struminger et al., 2013). Therefore, a balanced medial-to-
lateral hamstrings strength ratio is important for injury prevention (McAllister et al.,
2014; Palmieri-Smith et al., 2008; Struminger et al., 2013). Rozzi et al. (1999) found that
females have disproportionate firing of the lateral hamstrings (biceps femoris (BF))
compared to males during deceleration of a jump landing, increasing their risk of harmful
knee motion. Myer et al. (2005) also reported a decreased ratio of medial-to-lateral
quadriceps recruitment in females. The vastus medialis (VM) quadriceps muscle tends to
have a varus moment arm, while the vastus lateralis (VL) quadriceps muscle tends to
have a valgus moment arm. Thus, lower VM recruitment in women as suggested by Myer
et al. (2005) would allow for greater valgus loads at the knee. The combined deficits of
anterior and posterior medial thigh musculature recruitment may contribute to reduced
control of frontal plane forces at the knee (Hewett et al., 2005; Hewett et al., 2006;
Palmieri-Smith et al., 2009). Palmieri-Smith et al. (2009) further supported the notion that medial-to-lateral quadriceps and hamstrings co-contraction is unbalanced in women, suggesting that this limits their ability to resist valgus loads.

Additionally, an unbalanced or low ratio of medial-to-lateral quadriceps activation has the potential to compress the lateral side of the tibiofemoral joint, open the medial joint, and thus exert shear stress on the ACL (Hewett et al., 2006). Both quadriceps and hamstrings contractions are important for joint compression, which may protect the ACL against excessive anterior translation or valgus loads. Joint compression through muscular co-contraction allows for potentially harmful loads to be absorbed mainly via articular surfaces, thus protecting the ligaments (Hewett et al., 2006). Thomas et al. (2010) suggested that the quadriceps and hamstrings absorb energy upon impact, creating a muscle dominant landing strategy during jumping and cutting maneuvers in order to protect the ACL.

In addition to factors directly affecting the position and movement of the knee, studies have found that biomechanics of the hip and ankle can have significant secondary effects on knee joint mechanics (McLean et al., 2005; Sanna and O’Connor, 2008; Zeller et al., 2003). Some hazardous knee loads are known to stem from altered control elsewhere in the lower extremity (McLean et al., 2005), and combined motions at the hip and ankle can generate high-risk positions and motions at the knee (Sanna and O’Connor, 2008). Excessive ankle eversion, hip adduction, and hip internal rotation are among the most significant of these risk factors, which can result in detrimental knee biomechanics, particularly increased valgus moments (Ford et al., 2005; Ford et al., 2010; Hewett et al.,
Excessive ankle eversion increases tibial rotation, which then increases valgus moments at the knee (Ford et al., 2005), while excessive hip adduction and internal rotation may compromise the ability of medial thigh musculature to adequately support valgus loads during dynamic movements (McLean et al., 2005). These compromising motions and loads at the hip likely stem from decreased strength and altered timing and recruitment of the gluteal muscles (Struminger et al., 2013). Gluteus maximus externally rotates, extends, and abducts the hip, and is thus important in limiting hip internal rotation and adduction during dynamic movement (Hewett et al., 2006).

Several studies have indicated that high-risk hip biomechanics may influence the increased occurrence of ACL injury in females, suggesting that females may have altered dynamic neuromuscular control of the lower extremity and may adopt altered landing and cutting strategies compared to their male counterparts (Decker et al., 2003; Hewett et al., 2005; Zazulak et al., 2005). For example, Ford et al. (2010) associated gender differences in knee valgus with altered dynamic control of the lower extremity in the frontal plane, suggesting that these kinematic differences were caused by altered contraction patterns of the hip and thigh musculature.

Fatigue

Many of the biomechanical and neuromuscular components affecting movement of the knee joint are interrelated during dynamic motion. Numerous studies have investigated the effects of fatigue on lower extremity biomechanics and neuromuscular control. Fatigue is considered to be one of the most significant risk factors for ACL
injury, exacerbating many of the other biomechanical and neuromuscular risk factors (Behrens et al., 2013; Borotikar et al., 2008; Chappell et al., 2005; Dickin et al., 2015; Gehring et al., 2009; Kernozek et al., 2008; McLean et al., 2007; Thomas et al., 2010).

Epidemiological evidence suggests that ACL injury rates are higher at the end of athletic competition when fatigue is greatest (Price et al., 2004). Fatigue is an extrinsic factor stemming from combined physiological mechanisms at both the central and peripheral levels (Kernozek et al., 2008). Fatigue has been associated with decreased force production, impaired neuromuscular coordination and precision, slower muscle reaction times, increased joint laxity, and weakened proprioceptive capabilities (Chappell et al., 2005; Gehring et al., 2009; Madigan and Pidcoe, 2003; McLean et al., 2007; Melnyk and Gollhofer, 2007; Rozzi et al., 1999; Sanna and O’Connor, 2008; Thomas et al., 2010).

Many studies suggest that fatigue may contribute directly to ACL injury risk via the promotion of high-risk neuromuscular strategies (Chappell et al., 2005; McLean et al., 2007; Thomas et al., 2010). Compromised neuromuscular function can cause alterations in lower extremity landing and cutting mechanics which are believed to increase the risk of ACL injury, particularly in female athletes (Chappell et al., 2005; Kernozek et al., 2008; McLean et al., 2007; Sanna and O’Connor, 2008; Thomas et al., 2010). These fatigue-induced changes to neuromuscular function have been associated with reduced motor activity (Melnyk and Gollhofer, 2007), as well as extended latency of muscle contraction (Rozzi et al., 1999; Wojtys et al., 2003). The ACL may endure potentially hazardous forces during landing and cutting sports movements if the musculature that
controls the knee joint does not sufficiently dissipate the associated torques and forces (Hewett et al., 2005).

Lower extremity musculature plays a crucial role in injury prevention, helping to attenuate shock and stabilize joints (Madigan and Pidcoe, 2003; Melnyk and Gollhofer, 2007; Thomas et al., 2010). The quadriceps and hamstrings muscles are functionally important for maintaining stability of the knee joint and controlling hip and knee biomechanics. Therefore, fatigue of these muscles can significantly increase the likelihood of an ACL injury (Melnyk and Gollhofer, 2007; Thomas et al., 2010). Wojtys et al. (1996) found that isolated fatigue of both quadriceps and hamstrings induced potentially hazardous increases in anterior tibial translation and longer muscle reaction times. Similarly, Chappell et al. (2005) found that fatigue caused late onset of quadriceps and hamstrings activation and early occurrence of maximal knee flexion, which could decrease shock absorption and knee stabilization during dynamic movements. The hamstrings, in particular, play a role in resisting anterior tibial translation and excessive knee motions, thus resulting in higher injury risk when they are fatigued (Behrens et al., 2013).

Fatigue of the lower limb musculature can also affect movement at the hip and ankle joints. Borotikar et al. (2008) found that lower extremity fatigue caused significant increases in initial contact (IC) hip extension and internal rotation during a landing task, as well as increases in peak stance ankle supination angles. Thomas et al. (2010) studied the effects of quadriceps and hamstrings fatigue, and also found significant increases in IC hip internal rotation. Thomas et al. (2010) found that females landed with greater hip
flexion and less abduction than males at both IC and peak vertical ground reaction force (vGRF). Coventry et al. (2006) had similar results, finding overall increases in hip and knee flexion and decreases in ankle plantarflexion at IC with fatigue. These changes in hip and ankle biomechanics with general lower limb fatigue suggests an altered landing strategy, with a possible distal-to-proximal redistribution of work as a neuromuscular protective mechanism to decrease harmful forces at the knee joint (Coventry et al., 2006; Madigan and Pidcoe, 2003). However, if the musculature acting on the hip and knee joint is fatigued, alterations to dynamic sports mechanics may induce harmful forces and movements at the knee joint.

Altered neuromuscular function as a result of fatigue can also lead to higher loading rates at the knee during landing and cutting maneuvers. Thomas et al. (2010) found that peak vGRF was larger for females than males with hamstrings and quadriceps fatigue. High loading rates, or increased vGRF during a shorter amount of time, combined with decreased lower limb neuromuscular control could lead to larger moments placed on the knee joint, thus increasing ACL injury risk (Quatman et al., 2006; Thomas et al., 2010).

Fatigue of the lower limb has also been associated with changes in overall knee joint stability. The altered neuromuscular control caused by fatigue can result in decreased joint stability, increased anterior tibial shear force, strain on the ACL, and risk of injury (Behrens et al., 2013; Chappell et al., 2005; Melnyk and Gollhofer, 2007; Rozzi et al., 1999). Wojtys et al. (1996) found significant increases in anterior tibial translation after fatigue of thigh musculature. Muscle responses were significantly slowed or
occasionally absent after fatigue of the hamstrings and quadriceps, which appeared to affect the dynamic stability of the knee joint and altered the neuromuscular response to anterior tibial translation (Wojtys et al., 1996). Wojtys et al. (1996) concluded that the increase in tibial translation was due to delayed muscle reaction times and reduced muscle firing. Chappell et al. (2005) also found that general neuromuscular fatigue caused hazardous anterior tibial shear forces during a stop jump task, especially in females. This increased tibial shear force in women may be a combined effect of decreased knee flexion angle and increased valgus moment (Chappell et al., 2005). Behrens et al. (2013) further supported these findings, suggesting that reflex onset latencies were exacerbated in women after fatigue, which corresponded to an increase in tibial translation. Based on these results, the fatigue-induced decrease in neuromuscular function with a corresponding increase in tibial translation may contribute to the higher incidence of ACL injuries in women (Behrens et al., 2013).

The combined findings from studies examining the effects of neuromuscular fatigue suggest that women may exhibit greater performance changes than men, which increase their risk of noncontact ACL injury (Kernozek et al., 2008). Kernozek et al. (2008) found that men had greater peak knee flexion angles to attenuate shock after fatigue, while women did not alter knee flexion. McLean et al. (2007) found that females exhibited greater peak valgus moments with fatigue, which occurred closer to IC as fatigue progressed. Males did not experience the significant changes in valgus moments or the timing of peak moments that were exhibited by their female counterparts. McLean et al. (2007) concluded that fatigue-induced modifications in hip and knee joint control
strategies during landing and cutting maneuvers appear more pronounced in females than in males, suggesting that neuromuscular fatigue may be a factor in the gender disparity for ACL injury.

**Hypotheses**

ACL injuries have been attributed to a variety of mechanisms and risk factors, and are often caused by compounding influences. Females have a significantly greater risk for ACL injury than males, which is likely due to a variety of factors, including developmental, hormonal, biomechanical, and neuromuscular influences. This study is particularly interested in the effects of fatigue, which has been associated with a variety of biomechanical and neuromuscular alterations during dynamic movement. We chose to fatigue the hamstrings muscles in isolation in order to evaluate their influence on dynamic activity. Our study is, to the best of our knowledge, the only study to date that evaluates knee kinetics and kinematics after fatiguing the hamstrings in isolation. Additionally, this study is interested in the effects that age at menarche has on knee biomechanics and neuromuscular control after fatigue. Many studies have found correlations between the onset of menarche and decreased neuromuscular function in females, but little is known about how the age at menarche affects this development. This study tests several hypotheses regarding the effects of menarche and muscle fatigue on knee biomechanics. First, we hypothesize that earlier age at menarche is associated with greater valgus loading of the knee. Furthermore, we theorize that valgus loading will be greater with fatigue of the hamstrings muscles. Finally, we hypothesize that the effects of fatigue will be exacerbated in females with earlier age at menarche. Altered or abnormal
neuromuscular control likely contributes to the injury mechanism in many ACL injuries. Neuromuscular risk factors may be the most modifiable (Borotikar et al., 2008), justifying ongoing research in order to inform more effective preventative strategies. Identifying populations that may be more susceptible to these neuromuscular risk factors may also help to reduce the incidence of ACL injury.
II. MATERIALS AND METHODS

Participants

Twelve healthy females (age 20.9 ± 1.7 yrs, height 166.8 ± 6.2 cm, body mass 65.3 ± 10.6 kg) participated in the study. An a priori power analysis determined that 11 subjects would be required to achieve 80% statistical power to detect significant pre/post-fatigue differences with significance set to \( \alpha = 0.05 \). Sample size estimations and power analysis relied on data from a similar fatigue study conducted by Thomas et al. (2010).

Subjects were eligible for inclusion in the study if they were at least recreationally active, participating in organized sports and/or moderate exercise at least two times per week. Eight of the participants were NCAA Division I Wright State University athletes, and all were Wright State University students. Exclusion criteria for participation included (1) a history of ACL rupture or significant knee injuries; (2) lower limb pain or injuries that might affect biomechanics at the time of participation; (3) age < 18 or > 25.

All subjects provided informed consent, and the study was approved by the Wright State University institutional review board.

Data Collection and Instrumentation

Anthropometric measurements were taken using standard methods (Lohman et al., 1988) with subjects in light clothing and socks. Measurements included mass (to nearest 0.1 kg; scale), height (to nearest 0.1 cm; stadiometer), and sitting height (to
nearest 0.1 cm; stadiometer and chair). Subischial lower limb length was calculated as height – sitting height.

Biomechanical data was collected using the Wright State University Boonshoft School of Medicine Lifespan Health Research Center’s (LHRC) three-dimensional motion capture laboratory, equipped with six high-speed Osprey cameras (Motion Analysis Corp., Santa Rosa, CA) synchronized with three force plates embedded in a 15 m walkway (two AMTI OR6-7-1000, Advanced Medical Technology, Inc., Watertown, MA; and one Kistler Type 9281B11, Kistler Instruments, Winterthur, Switzerland). The cameras captured subject motion at a rate of 120 frames per second, recording the movement of reflective markers placed on precise anatomical landmarks (Figure 1). A total of 25 reflective markers were placed on the subjects following the Helen Hayes system (Kadaba et al., 1990), including a unilateral sacral marker, and bilateral markers at the mid-acromion process, lateral humeral epicondyle, mid-wrist, anterior superior iliac spine, thigh, medial and lateral femoral epicondyles, shank, medial and lateral malleoli, heel, and head of second metatarsal. The medial femoral epicondyle and medial malleoli markers were only used during static trials and were removed for dynamic tasks. Markers were placed by the same investigator for all subjects and trials.

Six trials were performed by each participant (three per side), and any trials with poor marker recognition were removed from the data set. For each variable, participant averages across viable trials for each leg were used in the analysis. Biomechanical data was collected using Cortex 6.0 software, and was processed using MacGait 1.0 software (Motion Analysis Corp., Santa Rosa, CA). Reported moments are external moments, which were calculated using inverse dynamics and normalized for body mass. Ground
reaction forces are expressed as multiples of body weight. This study uses the following conventions for angle and moment signs: positive values refer to abduction/valgus and flexion; negative values refer to adduction/varus and extension.

**Experimental Procedure**

A pre-/post-fatigue study design was used in this investigation, allowing for control of variation among subjects. Subjects participated in two visits to LHRC for data collection. During the first visit, subjects performed unfatigued drop vertical jump (DVJ) tasks, and completed a questionnaire regarding their menstrual history, exercise habits, and knee injury history. A second visit was scheduled roughly a month after the first visit while subjects were in the same phase of their menstrual cycle as they were during the first visit. This was determined using information provided by the subject regarding their recent menstrual history. During the second visit, the DVJ task was performed after subjects completed the fatigue protocol.

**Drop Vertical Jump**

For the DVJ task, subjects were asked to step or lightly hop off of a box with a height of 31 cm so that they landed on both feet simultaneously. Immediately after landing, subjects jumped straight up with hands raised as if going for a basketball rebound. Figure 1 displays a series of images showing the DVJ task. Prior to data collection, each subject performed one or more practice jumps until the investigators were satisfied that the subject understood the instructions and landed properly on the force plate. The DVJ was done a total of six times, with a single foot landing on the force plate at a time (three trials per side).
**Fatigue Protocol**

Fatigue was induced by having subjects perform glute-ham raises on a York glute-ham bench (Figure 2). This concentric exercise focuses peripheral fatigue on the gluteus maximus and hamstrings muscles, particularly biceps femoris long head (McAllister et al., 2014). Participants were positioned on the bench so that their hips or upper thighs rested comfortably on the roller pads and their legs were approximately parallel with the ground. The subjects performed each repetition at full range of motion with arms crossed over their chest.

At the end of the first visit, subjects performed the glute-ham raises to the point of muscular failure in order to determine their maximum repetition number. During the second visit, subjects performed the glute-ham raises to 70% of their maximum number of repetitions before biomechanical data was collected. Seventy percent of maximum was used to induce sufficient fatigue without putting subjects at high risk of injury during the DVJ task.

Immediately after completing the glute-ham raises at 70% of their max, subjects were shown a Borg Rating of Perceived Exertion (RPE) 6-20 scale and were asked to give a rating for the perceived fatigue of their hamstrings muscles. To maintain fatigue levels, subjects were periodically shown the Borg RPE scale and performed body weight squats as necessary between phases of data collection to maintain their original rating throughout the data collection process. On average, subjects performed approximately 15-20 body weight squats three or four times throughout the data collection process in order to maintain fatigue levels.
**Statistical Analysis**

Motion and force data were analyzed for the contact phase of the DVJ task. Initial contact (IC) was determined as the first frame in which GRF was measured by the force plate, and toe-off (TO) was indicated as the last frame with a GRF. DVJ trials were grouped based on leg dominance, which was determined by which limb was used most frequently to lead off during walking trials.

Frontal and sagittal plane knee angles and frontal plane knee moments were analyzed at IC, TO, maximum, and minimum during the full contact phase, and at peak vGRF using paired t-tests. The peak vGRF was also analyzed via a paired t-test, with an additional sensitivity analysis to test for the effects of removing an outlier. The effects of age at menarche on knee biomechanics and measures of fatigue were analyzed via two-way repeated measures ANOVA. Statistical analysis was conducted using the JMP 12.0 and SAS 9.3 integrated suite of statistical software.
Figure 1.

(A)

(B)
Figure 1.

Drop vertical jump (DVJ) task performed three times per leg by participants. (A) displays a series of images from Cortex 6.0 software showing movement of reflective markers placed on precise anatomical landmarks and showing force vectors during the contact phase of the DVJ task. (B) shows the corresponding video images of the DVJ task.

Images are in chronological order from left to right.
Figure 2.
Figure 2.

York glute-ham bench used to fatigue the hamstrings muscles.
III. RESULTS

During the fatiguing protocol, participants performed 32.5 ± 15.6 glute-ham raises at 70% of their maximum number of repetitions, and generally indicated a 16-17 RPE on the Borg 6-20 scale after the fatiguing protocol. Knee kinematics were measured for all 12 subjects, but kinetics (GRF and knee moments) were only measured for 10 of the subjects due to technical issues. Knee kinematic and kinetic measurements are summarized in Table 1.

At multiple time points during the DVJ, the knee abduction angle was significantly different between the pre- and post-fatigue trials for the dominant limb. After fatigue, knee abduction angle was significantly lower (i.e., less valgus) at IC ($P = 0.02$), at peak vGRF ($P = 0.01$), at TO ($P = 0.01$), and at its maximum during contact ($P = 0.01$) for the dominant limb (Figure 3). Knee abduction angles were consistently close to 4° less valgus after fatigue in the dominant leg (4.1° at IC, 3.7° at peak vGRF, 4.2° at TO, and 4.4° at maximum). No significant difference was found for minimum knee abduction angle in the dominant limb ($P = 0.41$). Fatigue did not have a significant effect on knee abduction angles for the non-dominant limb at IC ($P = 0.68$), at peak vGRF ($P = 0.11$), at TO ($P = 0.52$), or at maximum ($P = 0.90$) (Figure 3). Though not statistically significant ($P = 0.07$), the minimum knee abduction angle changed from a valgus angle to a varus angle as a result of fatigue in the non-dominant limb. Average minimum knee
abduction angles were negative values in both the fatigued (-1.6° ± 3.9° for dominant limb, -2.3° ± 4.2° for non-dominant limb) and unfatigued (-0.4° ± 5.7° for dominant limb, -0.5° ± 3.4° for non-dominant limb) states, indicating that, on average, varus angles were measured during at least a portion of the contact phase of the DVJ. Figure 4 displays the knee abduction angle throughout the contact phase of the DVJ before and after fatigue for both limbs.

Knee flexion angles in the sagittal plane were unaffected by fatigue in both limbs (Table 1). No significant difference in knee flexion angle was detected at IC ($P = 0.61$ for dominant limb; $P = 0.48$ for non-dominant limb), at peak vGRF ($P = 0.27$ for dominant limb, $P = 0.80$ for non-dominant limb), at TO ($P = 0.33$ for dominant limb; $P = 0.91$ for non-dominant limb), at its maximum ($P = 0.09$ for dominant limb; $P = 0.23$ for non-dominant limb), or at its minimum ($P = 0.39$ for dominant limb, $P = 0.90$ for non-dominant limb) during contact.

Fatigue did not significantly affect knee abduction moment at IC ($P = 0.40$), at peak vGRF ($P = 0.27$), at TO ($P = 0.22$), at its maximum ($P = 0.99$), or at its minimum ($P = 0.53$) for the dominant limb. The knee abduction moment at TO for the non-dominant limb was significantly different ($P = 0.05$) with fatigue, displaying a slight varus moment (-0.02 Nm/Kg) after fatigue compared to a valgus moment (0.16 Nm/Kg) without fatigue. Knee abduction moment in the non-dominant limb was not significantly influenced by fatigue at IC ($P = 0.65$), at peak vGRF ($P = 0.41$), at its maximum ($P = 0.23$), or at its minimum ($P = 0.81$) (Table 1). Figure 5 shows the knee abduction moment throughout the contact phase of the DVJ task before and after fatigue in both limbs.
Analysis of peak vGRF showed a trend for lower peak vGRF after fatigue for the dominant leg, though it was not statistically significant (1.5 ± 0.3 times body weight unfatigued, 1.3 ± 0.3 times body weight fatigued; \( P = 0.08 \)). Fatigue did not significantly affect peak vGRF in the non-dominant leg (\( P = 0.25 \)). A sensitivity analysis to evaluate peak vGRF without an outlier revealed that the peak vGRF was significantly lower after fatigue for both the dominant limb (1.5 ± 0.3 times body weight unfatigued, 1.2 ± 0.2 times body weight fatigued; \( P = 0.01 \)) and the non-dominant limb (1.4 ± 0.3 times body weight unfatigued, 1.1 ± 0.2 times body weight fatigued; \( P = 0.04 \)). Figure 7 displays the vGRF throughout the contact phase of the DVJ before and after fatigue for the dominant and non-dominant limbs.

The timing of peak kinetic and kinematic measures was analyzed to determine whether there was a significant change between fatigue states. Negative percentage values indicate earlier peak measures after fatigue, while positive values indicate later peak measures after fatigue. For the dominant limb, there was no significant difference between fatigue states for the percent contact phase at which peak knee abduction angle occurred (-11.1% change; \( P = 0.28 \)), for peak knee abduction moment (7.4% change; \( P = 0.28 \)), for knee flexion angle (0.7% change; \( P = 0.69 \)), or for peak vGRF (1.7% change; \( P = 0.60 \)). For the non-dominant limb, peak knee flexion angle occurred significantly later in the contact phase after fatigue (3.4% change; \( P = 0.001 \)). Figure 6 displays the change in knee flexion angle throughout the contact phase between fatigue states. For the non-dominant limb, there was no significant difference between fatigue states for the timing
of peak knee abduction angle (-5.2% change; \( P = 0.66 \)), for peak knee abduction moment (13.6% change; \( P = 0.21 \)), or for peak vGRF (-0.6% change; \( P = 0.84 \)).

Age at menarche had no significant effect on knee kinetic and kinematic measures (Table 2). For the dominant limb, age at menarche did not significantly influence knee abduction angle \( (P = 0.82 \text{ at IC, } P = 0.48 \text{ at maximum, } P = 0.70 \text{ at minimum, } P = 0.49 \text{ at TO}) \), knee flexion angle \( (P = 0.79 \text{ at IC, } P = 0.84 \text{ at maximum, } P = 0.63 \text{ at minimum, } P = 0.97 \text{ at TO}) \), knee abduction moment \( (P = 0.89 \text{ at IC, } P = 0.61 \text{ at maximum, } P = 0.34 \text{ at minimum, } P = 0.65 \text{ at TO}) \), or peak vGRF \( (P = 0.96) \). In the non-dominant limb, age at menarche was not significantly correlated with knee abduction angle \( (P = 0.88 \text{ at IC, } P = 0.71 \text{ at maximum, } P = 0.26 \text{ at minimum, } P = 0.86 \text{ at TO}) \), with knee flexion angle \( (P = 0.48 \text{ at IC, } P = 0.92 \text{ at maximum, } P = 0.72 \text{ at minimum, } P = 0.14 \text{ at TO}) \), with knee abduction moment \( (P = 0.61 \text{ at IC, } P = 0.07 \text{ at maximum, } P = 0.37 \text{ at minimum, } P = 0.41 \text{ at TO}) \), or with peak vGRF \( (P = 0.41) \).

A two-way repeated measures ANOVA analyzing the age at menarche*fatigue interaction effect was used to evaluate the relationship between age at menarche and the effects of fatigue on knee kinetic and kinematic measures (Table 2). For the dominant limb, age at menarche did not significantly influence the effects of fatigue for knee abduction angle \( (P = 0.27 \text{ at IC, } P = 0.23 \text{ at maximum, } P = 0.78 \text{ at minimum, } P = 0.38 \text{ at TO}) \), for knee abduction moment \( (P = 0.46 \text{ at IC, } P = 0.27 \text{ at maximum, } P = 0.89 \text{ at minimum, } P = 0.38 \text{ at TO}) \), or for peak vGRF \( (P = 0.84) \). Though not statistically significant, age at menarche trended toward a significant interaction with the effects of fatigue on IC knee flexion angle in the dominant limb \( (P = 0.06) \); however, no interaction
was found during any other point in the contact phase for knee flexion angle ($P = 0.30$ at maximum, $P = 0.10$ at minimum, $P = 0.25$ at TO). For the non-dominant limb, age at menarche was significantly correlated with the effects of fatigue on IC knee flexion angle ($P = 0.05$), but it did not influence the effects of fatigue on knee flexion angle at any other time during the contact phase ($P = 0.82$ at maximum, $P = 0.11$ at minimum, $P = 0.13$ at TO). Age at menarche had no significant influence on the effects of fatigue for knee abduction angle ($P = 0.77$ at IC, $P = 0.91$ at maximum, $P = 0.28$ at minimum, $P = 0.72$ at TO), for knee abduction moment ($P = 0.30$ at IC, $P = 0.08$ at maximum, $P = 0.46$ at minimum, $P = 0.15$ at TO), or for peak vGRF ($P = 0.23$) in the non-dominant limb.
Table 1.

<table>
<thead>
<tr>
<th>Contact Phase Event</th>
<th>Dominant</th>
<th>Non-dominant</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre-</td>
<td>Post-</td>
</tr>
<tr>
<td>Knee Abduction Angle (°)</td>
<td>10.5 ± 5.3</td>
<td>6.4 ± 3.2</td>
</tr>
<tr>
<td>IC*</td>
<td>8.3 ± 4.6</td>
<td>4.7 ± 2.6</td>
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<tr>
<td>Peak vGRF*</td>
<td>10.1 ± 4.8</td>
<td>5.9 ± 1.6</td>
</tr>
<tr>
<td>TO*</td>
<td>13.5 ± 5.0</td>
<td>9.1 ± 2.2</td>
</tr>
<tr>
<td>Maximum*</td>
<td>-0.4 ± 5.7</td>
<td>-1.6 ± 3.9</td>
</tr>
<tr>
<td>Knee Flexion Angle (°)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IC</td>
<td>35.5 ± 9.0</td>
<td>34.3 ± 8.7</td>
</tr>
<tr>
<td>Peak vGRF</td>
<td>64.2 ± 10.1</td>
<td>68.3 ± 9.4</td>
</tr>
<tr>
<td>TO</td>
<td>32.6 ± 12.5</td>
<td>27.9 ± 8.5</td>
</tr>
<tr>
<td>Maximum</td>
<td>92.0 ± 12.0</td>
<td>97.4 ± 14.4</td>
</tr>
<tr>
<td>Minimum</td>
<td>29.3 ± 7.6</td>
<td>26.2 ± 7.8</td>
</tr>
<tr>
<td>Knee Abduction Moment (Nm/Kg)</td>
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<td></td>
</tr>
<tr>
<td>IC</td>
<td>0.1 ± 0.3</td>
<td>-0.1 ± 0.6</td>
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<tr>
<td>Peak vGRF</td>
<td>-0.3 ± 0.6</td>
<td>-0.6 ± 0.5</td>
</tr>
<tr>
<td>TO†</td>
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<td>-0.1 ± 0.5</td>
</tr>
<tr>
<td>Maximum</td>
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<td>0.8 ± 1.0</td>
</tr>
<tr>
<td>Minimum</td>
<td>-1.0 ± 0.8</td>
<td>-1.3 ± 0.9</td>
</tr>
</tbody>
</table>

Note. A negative value for knee abduction angle or moment indicates a knee adduction/varus angle or moment, respectively.

* Indicates a statistically significant difference between fatigue states for the dominant limb \( P \leq 0.05 \).

† Indicates a statistically significant difference between fatigue states for the non-dominant limb \( P \leq 0.05 \).
Table 1.
The effects of fatigue on mean (± SD) knee kinematic and kinetic variables at various time points of the DVJ contact phase for the dominant and non-dominant legs.
Table 2.

<table>
<thead>
<tr>
<th>Contact Phase Event</th>
<th>Knee Abduction Angle</th>
<th>Knee Flexion Angle</th>
<th>Knee Abduction Moment</th>
<th>vGRF</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IC</td>
<td>Maximum</td>
<td>Minimum</td>
<td>TO</td>
</tr>
<tr>
<td></td>
<td>0.82</td>
<td>0.48</td>
<td>0.79</td>
<td>0.97</td>
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<td></td>
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<td>0.71</td>
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<td></td>
<td>0.70</td>
<td>0.26</td>
<td>0.63</td>
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<td></td>
<td>0.49</td>
<td>0.86</td>
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<td>0.89</td>
<td>0.61</td>
<td>0.46</td>
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</tr>
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<td></td>
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<td>0.07</td>
<td>0.27</td>
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<td></td>
<td>0.34</td>
<td>0.37</td>
<td>0.89</td>
<td>0.65</td>
</tr>
<tr>
<td></td>
<td>0.65</td>
<td>0.41</td>
<td>0.38</td>
<td>0.96</td>
</tr>
<tr>
<td>Dominant</td>
<td>0.27</td>
<td>0.78</td>
<td>0.72</td>
<td>0.84</td>
</tr>
<tr>
<td>Non-dominant</td>
<td>0.77</td>
<td>0.28</td>
<td>0.72</td>
<td>0.23</td>
</tr>
<tr>
<td>Maximum</td>
<td>0.71</td>
<td>0.91</td>
<td>0.91</td>
<td>0.11</td>
</tr>
<tr>
<td>Minimum</td>
<td>0.26</td>
<td>0.92</td>
<td>0.27</td>
<td>0.46</td>
</tr>
<tr>
<td>vGRF</td>
<td>0.23</td>
<td>0.05</td>
<td>0.30</td>
<td>0.82</td>
</tr>
</tbody>
</table>

Note. Displayed values are P values.

* Indicates a statistically significant P value (P ≤ 0.05).
Table 2.
The effects of age at menarche ($P$ values) on overall knee kinetics and kinematics, and the influence of age at menarche ($P$ values) on the effects of fatigue on knee kinetics and kinematics during the DVJ contact phase for the dominant and non-dominant legs.
Figure 3.

Dominant

- Initial Contact *
- Peak vGRF *
- Toe-off *
- Maximum *

Non-Dominant

- Initial Contact
- Peak vGRF
- Toe-off
- Maximum
Figure 3.

Effect of fatigue on knee abduction angle (degrees) at IC, peak vGRF, TO, and maximum for the dominant and non-dominant legs. Open bars: unfatigued, filled bars: fatigued. Data are displayed as maximum, minimum, interquartile range, and median. X indicates the mean.

* denotes a significant difference between fatigue states ($P \leq 0.05$).
Figure 4.

**Dominant Limb**

- Unfatigued
- Fatigued

**Non-dominant Limb**

- Unfatigued
- Fatigued

Knee abduction angle (degrees) vs. Percent of contact phase.
**Figure 4.**

Average knee abduction angle throughout the contact phase of the DVJ task before and after fatigue for the dominant and non-dominant limbs. The solid waveforms are the mean knee abduction angle ± one standard deviation (dashed waveforms). The solid vertical lines represent IC and TO (at 0 and 100% contact phase, respectively), and the dashed vertical line represents peak vGRF.
Figure 5.

Dominant Limb

Non-dominant Limb
Figure 5.

Average knee abduction moment throughout the contact phase of the DVJ task before and after fatigue for the dominant and non-dominant limbs. The solid waveforms are the mean knee abduction moment ± one standard deviation (dashed waveforms). The solid vertical lines represent IC and TO (at 0 and 100% contact phase, respectively), and the dashed vertical line represents peak vGRF.
Figure 6.

Dominant Limb

Non-dominant Limb
Figure 6.
Average knee flexion angle throughout the contact phase of the DVJ task before and after fatigue for the dominant and non-dominant limbs. The solid waveforms are the mean knee flexion angle ± one standard deviation (dashed waveforms). The solid vertical lines represent IC and TO (at 0 and 100% contact phase, respectively), and the dashed vertical line represents peak vGRF.
Figure 7.
**Figure 7.**

Average vertical ground reaction force (vGRF) throughout the contact phase of the DVJ task before and after fatigue for the dominant and non-dominant limbs. The waveforms represent the mean vGRF (solid line) ± one standard deviation (dashed lines). The solid vertical lines represent IC and TO (at 0 and 100% contact phase, respectively), and the dashed vertical line represents peak vGRF. Ground reaction forces are expressed as a multiple of body weight.
IV. DISCUSSION

The purpose of this study is to examine the effects of hamstrings fatigue and age at menarche on knee biomechanics and to determine whether a correlation exists between age at menarche and the degree to which hamstrings fatigue affects knee biomechanics. Previous studies have suggested that fatigue can induce changes in knee biomechanics that may increase risk of ACL injury, especially in women (Borotikar et al., 2008; Chappell et al., 2005; Dickin et al., 2015; Kernozek et al., 2008; McLean et al., 2007; Padua et al., 2006; Wojtys et al., 1996). Additionally, menarche and puberty influence maturational processes involved in skeletal development and neuromuscular control of the knee (Hewett et al., 2004; Quatman-Yates et al., 2012; Seselj et al., 2012), indicating that age at menarche may influence knee biomechanics. The results of this study do not support our initial hypotheses that (1) age at menarche is associated with greater valgus loading of the knee, (2) that valgus loading is greater after hamstrings fatigue, or (3) that the effects of fatigue are exacerbated in females with earlier age at menarche. Rather than increasing with fatigue, knee valgus decreased after the fatiguing protocol within our sample, specifically in the dominant limb, and no substantial correlations were found regarding age at menarche.
Effects of Fatigue on Frontal Plane Knee Kinetics and Kinematics

In contrast to our initial hypothesis, the results of this study indicate that knee abduction angle decreased in the dominant limb after the fatiguing protocol. With fatigue, the knee abduction angle was significantly lower by an average of 4.1° at IC, peak vGRF, TO, and maximum for the dominant leg. This is contradictory to the findings of Borotikar et al. (2008), Chappell et al. (2005), Dickin et al. (2015), and McLean et al. (2007), who found an increase in knee abduction angle after fatigue. Other studies have found no significant change in knee abduction angle with fatigue (Gehring et al., 2009; Kernozek et al., 2008; Thomas et al., 2010), but we are unaware of any studies to date that have found a significant decrease in knee abduction angle after fatigue apart from the current study.

Previous studies have examined the effects of fatigue on lower limb biomechanics, many suggesting that neuromuscular fatigue may perpetuate potentially hazardous knee biomechanics which can increase the risk of ACL injury (Borotikar et al., 2008; Chappell et al., 2005; Kernozek et al., 2008; McLean et al., 2007; Padua et al., 2006). In a study by Borotikar et al. (2008), female subjects performed sets of body weight squats to the point of failure while biomechanical changes during single-leg landings were examined as fatigue progressed. Results showed a significant increase in peak knee abduction and internal rotation angles with fatigue (Borotikar et al., 2008). The results also indicated that fatigue induced changes in kinematic parameters at only 50% of maximum fatigue, suggesting that potentially hazardous lower limb positions may occur at lower levels of fatigue than previously assumed (Chappell et al., 2005; McLean
et al., 2007). A recent study by Dickin et al. (2015) fatigued female subjects with a weighted squat and jumping lunge fatigue protocol, and also found an increase in maximum knee valgus angle after fatigue during a DVJ from various heights. Similarly, McLean et al. (2007) found that female subjects experienced significant increases in peak knee abduction angles and moments during a DVJ task for both dominant and non-dominant legs after a generalized lower limb fatigue protocol. Chappell et al. (2005) found the same increases in knee abduction angle and moments during a stop jump task using a generalized fatigue protocol. It should be noted that all of these studies that found an increase in knee abduction angle with fatigue utilized a generalized thigh fatigue protocol rather than isolating a particular muscle group, such as the protocol used in the current study.

In contrast to the results of Borotikar et al. (2008), Chappell et al. (2005), Dickin et al. (2015), and McLean et al. (2007), several studies did not find significant changes in knee abduction angles or moments with fatigue. Kernozek et al. (2008) did a study using a weighted squat fatigue protocol and biomechanical analysis of a single-leg drop landing. Results of this study found increases in anterior tibial shear force after fatigue; however, women did not significantly alter knee flexion or valgus angle (Kernozek et al., 2008). Gehring et al. (2009) induced fatigue via a leg press machine, and measured lower limb biomechanics and electromyographic (EMG) muscle activity during a double-leg drop landing. Results of this study found that hamstrings and gastrocnemius muscle activity decreased with fatigue; however, knee abduction angles did not change. In a study by Thomas et al. (2010), fatigue was induced isokinetically via alternating
quadriceps and hamstring concentric contractions, and knee biomechanics were analyzed using a single-leg forward hopping task. After fatigue, subjects displayed a decrease in knee flexion and internal rotation angles at peak vGRF, but no change occurred in knee abduction angles or moments (Thomas et al., 2010). The lack of change in knee abduction angles following fatigue contradicted the initial hypotheses of the Thomas et al. (2010) study, as well as the findings of other relevant studies (Borotikar et al., 2008; Chappell et al., 2005; McLean et al., 2007). Unlike many previous studies that did find a change in frontal plane knee angles and moments, the studies by Gehring et al. (2009), Kernozek et al. (2008), Thomas et al. (2010), as well as the current study, did not use fatiguing protocols that induce significant fatigue of hip adductor or abductor musculature. Although the thigh musculature comprises the primary stabilizers of knee frontal plane motion, the hip adductors and abductors may also stabilize the knee, particularly in the frontal plane (Lloyd and Buchanan, 2001).

Muscles that cross the knee joint play an important role in maintaining safe knee kinetics and kinematics, and the activity of these muscles can affect the strains and forces experienced by the ACL (Takeda et al., 1994). Several studies have examined activity of thigh musculature and the effects of fatigue on activation patterns (Behrens et al., 2013; Myer et al., 2005; Palmieri-Smith et al., 2008; Rozzi et al., 1999; Schache et al., 2013). In general, increased quadriceps and hamstrings strength has been shown to reduce frontal plane moments at the knee, particularly during landing tasks (Hewett et al., 1996). Lloyd and Buchanan (2001) found that overall knee joint stability was increased with greater combined medial and lateral hamstrings activity. The primary action of the hamstrings
muscles occurs in the sagittal plane; however, they also control knee varus and valgus motions (Lloyd et al., 2005). Females tend to activate lateral thigh musculature more so than medial (Myer et al., 2005; Rozzi et al., 1999), particularly when landing from a jump (Rozzi et al., 1999). Greater activity of the lateral thigh musculature would tend to abduct the knee without equal loads from the medial musculature (Palmieri-Smith et al., 2008). The medial hamstrings, in particular, have a varus moment arm at the knee (Lloyd et al., 2005); however, a study by Kernozek et al. (2005) showed that women did not generate significant knee varus moments during peak knee valgus angle as compared to their male counterparts. Several studies have suggested that hamstrings activation, particularly on the medial side, may help control frontal plane knee motion and subsequent ACL loading during dynamic activity (Lloyd et al., 2005; Palmieri-Smith et al., 2008; Struminger et al., 2013). Earlier and greater medial hamstrings activation during the preparatory and loading phases of landing activities has been correlated with lower knee valgus moments (McLean et al., 2010, Palmieri-Smith et al., 2008).

Our study is, to the best of our knowledge, the only study to date that has found a significant decrease in knee abduction angle following a fatiguing protocol. Our fatiguing protocol included the use of a glute-ham bench, which has not been used in any other relevant study. Although local fatiguing methods, such as the glute-ham raise used in this study, are not realistic to most sports settings, fatiguing muscle groups in isolation allows for analysis of specific muscle group contributions to dynamic activity. The unique results of the present study may be explained by the use of the glute-ham bench, and the manner in which it isolates muscles. A study by McAllister et al. (2014) on various
hamstrings exercises found that activity of biceps femoris (BF), the lateral hamstring, was maximized during the glute-ham raise, whereas the medial semitendinosus (ST) and semimembranosus (SM) hamstrings were less active. Thus, if our fatiguing protocol disproportionately targeted the BF muscle, we would expect it to exert a lower force after fatigue. The BF muscle, specifically the long head, exhibits hip extensor and adductor moment arms, as well as knee flexor, abductor, and external rotation moment arms (Schache et al., 2013). A study by Schache et al. (2013) found that the BF consistently exhibited a knee abduction moment in the frontal plane. Conversely, ST and SM tend to generate adduction moments at the knee (Palmieri-Smith et al., 2008). Thus, we would expect lower valgus angles to be exhibited when force from the medial hamstrings is not overpowered by force from the more fatigued lateral hamstring (BF) (Palmieri-Smith et al., 2008). Because the fatiguing protocol of this study targeted the BF muscle, we believe that potentially harmful knee abduction angles were in fact reduced as a result of lower lateral hamstrings activity relative to medial hamstrings activity after fatigue. Unfortunately, we were unable to use EMG to measure muscle activity in this study to confirm changes in muscle activation with fatigue, warranting further studies on the effects of isolated muscle fatigue on dynamic movement. A better understanding of individual muscle contributions during dynamic activity may help inform more effective strengthening and injury prevention programs.

Because women tend to activate lateral thigh musculature more so than medial (Myer et al., 2005; Rozzi et al., 1999), establishing a balance between medial and lateral thigh muscle activation may reduce high risk valgus knee angles and moments that can
put strain on the ACL (Palmieri-Smith et al., 2008). Although reduced BF activation may decrease potentially hazardous knee abduction angles as suggested by the results of our study, it is important to note that the lateral hamstrings may also play an important role in reducing anterior tibial translation and thus ACL loading (Lloyd and Buchanan, 2001; Struminger et al., 2013). Melnyk and Gollhofer (2007) found a decrease in knee stability and increase in anterior tibial translation after selectively fatiguing the hamstrings. More specifically, Behrens et al. (2013) measured a negative correlation between BF activity and tibial translation after fatigue in women. Additionally, the external rotation moment created by BF helps to limit knee internal rotation, which can increase injury risk when excessive (Fujii et al., 2012). These additional roles of the BF muscle should be taken into consideration when evaluating its overall role in knee biomechanics.

**Effects of Fatigue on Peak Vertical Ground Reaction Force**

In addition to a decrease in knee abduction angles following fatigue, our study also found a significant decrease in peak vGRF after fatigue for both the dominant and non-dominant limbs when excluding an outlier from the sample. The effects of fatigue on peak vGRF have varied greatly among related studies. During a double-leg drop landing, subjects in a study by Gehring et al. (2009) showed no change in peak vGRF with fatigue, but did show a significant reduction in vGRF during several phases of the landing task. Madigan and Pidcoe (2003) found that peak vGRF was significantly decreased by an average of 12% with fatigue during a single-leg landing. Kernozek et al. (2008) found that when performing a single-leg drop landing, women had higher peak vGRF normalized to body weight than men both before and after fatigue, and they

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exhibited a trend for reduced vGRF after fatigue, but it was not statistically significant \((P = 0.08)\). Thomas et al. (2010) found no significant effect of fatigue on peak vGRF during single-leg forward hops, while Dickin et al. (2015) found an increase in peak vGRF and maximum loading rate after fatigue during a DVJ.

The magnitude of the peak vGRF determines the magnitude of the external moments experienced throughout the lower extremity during landing. Thus, a higher vGRF could increase injury risk by placing larger moments on the joints (Thomas et al., 2010). The significant decrease in peak vGRF after fatigue seen in the current study may be the result of an altered landing strategy in order to reduce impact forces. A study by Padua et al. (2006) found that subjects altered muscle activation strategies as a result of fatigue in order to maintain overall landing mechanics. Following hamstrings fatigue, it is possible that subjects in our study adopted an antagonist inhibition strategy (Padua et al., 2006) in which quadriceps activation was maintained or increased. This strategy would help maintain sagittal plane kinetics and kinematics at the knee, which was supported by the lack of change seen in overall knee flexion angles throughout the contact phase in our study. The results of the study do show, however, that the timing of peak knee flexion angle in the non-dominant limb occurred significantly later in the contact phase after the fatiguing protocol. This may be a result of an altered landing strategy in which the subjects attempt to dissipate external forces over a wider range of joint motion in order to reduce impact forces experienced at the knee joint following fatigue (Decker et al., 2003). A study by Decker et al. (2003) found that female subjects utilized a wider range of joint motion after fatigue, particularly at the hip. We did not analyze hip kinematics in this
study, but we suspect that hip flexion at IC may have been lower after fatigue. Future studies should include analysis of hip biomechanics, as well as EMG data, in order to better evaluate changes in landing mechanics and muscle activation as a result of hamstrings fatigue.

Influence of Age at Menarche

Previous studies have suggested that the developmental changes that occur during menarche may have significant effects on ACL injury risk (Ahmad et al., 2006; Froehle et al., 2016; Hewett et al., 2004; Hewett et al., 2006). In particular, neuromuscular control of the knee and subsequent biomechanical function may be affected by the maturational changes associated with menarche (Hewett et al., 2004; Quatman-Yates et al., 2012). Our study aims to better understand dynamic knee valgus alignment, which has been suggested as a key risk factor for ACL injury and which develops during puberty in females (Hewett et al., 2004; Hewett et al., 2005). The results of our study do not support our hypotheses that age at menarche is associated with greater knee valgus angles and moments during the DVJ, nor that the effects of fatigue are exacerbated in females with earlier age at menarche.

A study by Hewett et al. (2004) found that girls in late or postpubertal stages displayed greater knee valgus angles than prepubertal girls or girls in early puberty, and it was hypothesized that females experience a decrease in neuromuscular control of the knee after puberty resulting in more hazardous knee kinetics and kinematics. A more recent study by Hewett et al. (2015) also found that adult females had increased peak knee abduction moments and angles relative to maturing adolescent females. Sigward et
al. (2012) compared biomechanics during a DVJ task between males and females across maturational levels. This study found that sex differences in lower limb biomechanics were present in pre-pubertal athletes, which is contradictory to the findings of Hewett et al. (2004), which suggest that no difference exists during this developmental stage; however, Sigward et al. (2012) found that the differences between male and female biomechanics became magnified post-puberty. The rapid increase in skeletal growth associated with puberty in females in conjunction with an absence of neuromuscular adaptation may cause increased dynamic knee abduction moments and angles and subsequent increases in injury risk (Hewett et al., 2015; Holden et al., 2016).

Although there is substantial evidence that puberty and menarche affect knee biomechanics in women, there is still little research on the effects of age at menarche. In our study, age at menarche was significantly correlated with the effects of fatigue on IC knee flexion angle in the non-dominant limb, and age at menarche trended toward a significant interaction with the effects of fatigue on IC knee flexion angle in the dominant limb as well. On average, the sample exhibited slightly less knee flexion angle at IC after fatigue (1.2° less in the dominant limb, 1.5° less in the non-dominant limb), but this change was not statistically significant. The change in IC knee flexion angle varied greatly among subjects, such that some subjects exhibited less knee flexion and others more knee flexion after fatigue. The degree of change in both limbs trended with age at menarche: subjects with earlier age at menarche tended to be less flexed at IC after fatigue, while subjects with later age at menarche tended to be more flexed at IC when fatigued. These trends were not statistically significant, so no firm conclusions can be
made; however, they warrant future studies on the interaction between age at menarche and the effects of fatigue on lower limb biomechanics, particularly the potential for a stiffer landing strategy in females with earlier age at menarche.

Our results show no evidence that age at menarche directly affects knee angles and moments in this sample, and there were no other significant interactions between age at menarche and the effects of fatigue on knee biomechanics other than the IC knee flexion angle. It is possible that our sample size was not large enough to detect a significant difference in any other measured kinetics or kinematics based on age at menarche. Additionally, given that our glute-ham raise fatiguing protocol was unique and the subsequent effects of fatigue were contrary to most other studies, future studies on the effects of age at menarche and fatigue would be beneficial before any conclusions should be made.

**Influence of Leg Dominance**

When examining lower extremity biomechanics, it is important to evaluate the effects of leg dominance, as well as to differentiate the roles of each limb when defining limb dominance. For example, when kicking a ball, the mobilizing or manipulating limb is generally considered to be the dominant limb, while the stabilizing limb is considered to be the non-dominant limb (Sadeghi et al., 2000). These varying roles could influence the biomechanics of each limb individually as well as how each limb is affected by fatigue. The results of our study showed that the decreases in knee abduction angle after fatigue were significant for the dominant limb only. It is possible that subjects utilized their dominant side more so than the non-dominant side to complete the glute-ham raises,
causing greater fatigue to the BF muscle on the dominant side. Alternatively, because the dominant limb is generally the manipulating limb, it may actually be weaker and more susceptible to fatigue than the non-dominant leg which is utilized more for stability. Interestingly, the results in Table 1 show that the post-fatigue knee abduction angles for the dominant limb resemble the pre-fatigue knee abduction angles in the non-dominant limb. Because the non-dominant limb is typically the stabilizing limb during athletic maneuvers, it may exhibit more balanced medial-to-lateral hamstring activity and thus less risky biomechanics. Meanwhile, the dominant limb may exhibit more unbalanced hamstrings activity, which may explain the greater change in knee abduction angles as a result of BF fatigue. When BF is fatigued in the dominant limb, it may allow for a more even contraction force from the medial and lateral hamstrings, causing the fatigued dominant leg biomechanics to more closely resemble the unfatigued non-dominant leg biomechanics. These hypotheses cannot be confirmed by the results of the current study, and should be examined using EMG in follow-up studies.

Few studies on knee biomechanics have evaluated the effects of leg dominance on knee kinematics and kinetics, and many have only analyzed the dominant limb. A study by McLean et al. (2007) did account for limb dominance, but found that it did not affect any kinematic measures during a DVJ task. Hewett et al. (2004) found that peak valgus angle during a DVJ task was significantly lower on the non-dominant side than on the dominant side for girls in late or postpubertal stages of development. A recent study by Paz et al. (2016) did not find a difference in knee valgus angles between dominant and non-dominant limbs during a DVJ task using male subjects, suggesting that gender may
influence the effects of limb dominance. During the same study, Paz et al. (2016) did find a significant difference in knee valgus angle between limbs during a forward step-up task, suggesting that differences between limbs may also depend on the biomechanical task being performed. Thus, future studies on knee biomechanics and the effects of fatigue should account for variation in limb dominance.

**Variation in Fatiguing Protocols and Biomechanical Analyses**

Studies on the effects of fatigue on lower limb biomechanics have produced a wide array of results. Several factors may influence discrepancies between the results of the current study and those of other relevant research. In particular, the fatiguing protocols have varied tremendously among these studies, ranging from functional exercise protocols to strictly controlled, isolated fatigue conditions. Although functional fatigue protocols such as running and jumping more closely resemble conditions that are experienced in a sports setting, it can be difficult to regulate and monitor the state of fatigue. Conversely, the functional relevance of highly controlled, isolated fatigue protocols is limited as they are not generally experienced in a sports setting (Gehring et al., 2009). Additionally, isolated fatigue methods have varied greatly among studies, including concentric and eccentric exercises as well as isokinetic and isotonic exercises using a dynamometer.

Some studies suggest that functional fatigue models may result in a variety of adaptive neuromuscular strategies in order to maintain joint stability and to limit forces experienced at the joints (Gehring et al., 2009; Madigan and Pidcoe, 2003; Padua et al., 2006; Thomas et al., 2010). Different fatigue protocols can yield varying biomechanical
effects and compensation strategies. These neuromuscular characteristics and activation strategies can lead to different loading of the knee joint (Lloyd et al., 2005); thus, it may be difficult to compare results among studies with differing fatiguing protocols (Madigan and Pidcoe, 2003).

Variability in study results may also be influenced by the biomechanical tasks used to measure lower limb kinetics and kinematics. These tasks have varied greatly among studies, including both double-leg and single-leg tasks. Some tasks have involved just landing maneuvers, while others have included a jumping component as well. Some studies indicate that single-leg tasks produce more hazardous biomechanical strategies than double-leg tasks (Paz et al., 2016). Kernozek et al. (2008) chose to use a single-leg landing task because of the probable asymmetries between legs in double-leg landings (Schot et al., 1994), and because many non-contact ACL injuries that occur during landing are single-legged (Olsen et al., 2004). Other studies have used double-leg tasks, such as the DVJ used in the current study, and have still found significant changes in knee kinetics and kinematics (Hewett et al., 2004; McLean et al., 2007). Future studies on the differences between fatigue protocols and biomechanical assessment tasks may provide a better understanding of the variation in results among these studies.

**Study Limitations**

Several limitations of this study should be noted when considering the results and analyses. The fatigue protocol used was unique to our study, which likely contributed to the discrepancy between our results and the results of other studies. Because the glute-ham raise is not a widely used exercise, it is possible that there was a training bias
between subjects who had done the exercise before and subjects who had not, which
could affect the level of fatigue between subjects. Also, the results of our study may have
been influenced by the sample used, as maturity, skill level, and training can affect
neuromuscular control of the lower limb (Hewett et al., 2004). Because our study only
examined athletes and relatively active subjects, all of whom were free of prior knee
injuries, it may be that our sample was generally more effective at limiting potentially
hazardous knee biomechanics while fatigued than the population as a whole.

Additionally, it is possible that our pre- post-fatigue study design was confounded by
deterioration of fatigue effects (McLean et al., 2007). We attempted to avoid this
deterioration by having subjects perform body weight squats between phases of data
collection; however, this exercise may not have maintained the same fatigue effects that
were initially induced by the glute-ham raises. Because pre- and post-fatigue data were
collected during different visits, it is possible that marker placement between visits were
not exactly the same. We attempted to limit this variable by having the same investigator
position the markers each time, and it is therefore highly unlikely that systematic
differences in marker placement from one trial to the next explain the study’s results.

Conclusions

The findings of this study do not support our hypotheses that (1) age at menarche
is correlated with greater knee valgus angles and moments, (2) that valgus loading of the
knee is greater after fatigue, or (3) that the effects of fatigue are exacerbated in females
with earlier age at menarche. Based on the results of this study, the following conclusions
can be made:
• Fatiguing protocols that do not incorporate fatigue of hip adductors and abductors may not induce significant changes in frontal plane knee kinetics and kinematics.

• Targeted fatigue of the lateral hamstrings may decrease potentially harmful valgus knee angles and moments during landing and jumping tasks.

• Reduced peak vGRF after fatigue may be a result of an altered landing strategy in order to reduce impact forces and to maintain overall landing mechanics.

• Age at menarche did not directly influence knee valgus angles and moments and had no substantial correlations with the effects of fatigue on knee kinetics and kinematics in our study.

• Limb dominance has a significant correlation with the effects of fatigue on knee biomechanics.

• Differences in fatiguing protocols and biomechanical assessments make it difficult to compare results among studies evaluating lower limb biomechanics and the effects of fatigue.
V. REFERENCES


