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Noncontact anterior cruciate ligament (ACL) injuries commonly occur upon initial foot contact (IC) with the ground during single-leg cutting or jump-landing maneuvers. Because these injuries occur in the absence of physical contact with another player or object, it is believed that some of these injuries may be avoided through intervention strategies aimed to target modifiable injury risk factors. In this regard, hamstring musculo-articular stiffness (K_{HAM}) may play a critical role in protecting the ACL during functional athletic movements by helping resist biomechanical characteristics indicative of ACL loading, such as proximal tibia anterior shear force (PTASF), anterior tibial translation (ATT), and anterior tibial acceleration (ATA). However, current evidence regarding the influence of K_{HAM} on knee joint biomechanics is limited to studies of nonweight bearing perturbations and double-leg landing tasks, which may not adequately represent the single-leg landing situations in which noncontact injuries commonly occur. Additionally, males and females have been included in the same analyses without accounting for between-sex differences that may confound reported relationships. Thus, the purposes of this study were to: 1) compare the neuromuscular and biomechanical demands of a double-leg stop-jump (DLSJ) task to that of a single-leg stop-jump (SLSJ) task in males and females; 2) determine, within each sex, the extent to which K_{HAM} predicts ACL-loading characteristics during a SLSJ, after controlling for initial body positioning (i.e. trunk center-of-mass position and hip and knee flexion angles at IC); and 3) examine the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics collectively predict ACL-loading characteristics during a SLSJ. Eighty healthy, physically-active, males (n = 40) and females (n = 40) completed a 5-min warmup, were measured for anterior knee laxity (AKL), quadriceps and hamstring maximal voluntary isometric contractions (MVIC), and K_{HAM}, and then performed the DLSJ and SLSJ tasks, during

which biomechanical and neuromuscular activation data were collected. Compared to the DLSJ, males and females performed the SLSJ with a more posterior trunk center-of-mass position (P < .001) and smaller knee-flexion angles (P < .001) at IC, less knee-flexion excursion (P = .038), greater ground reaction forces (P < .001), knee-extension moments (P = .033), and PTASF (P < .001).001), and less ATT (P = .007). Compared to men, women performed both tasks with smaller knee-flexion angles at IC (P = .047), less hip-flexion excursion (P = .006), slower hip-flexion velocities (P = .040), smaller hip-extension moments (P < .001), and greater ATT (P = .006); however, compared to the DLSJ, females performed the SLSJ with a greater reduction in hipflexion velocity (P < .001) and a smaller increase in hip-extension moment (P < .001) than males. Irrespective of sex, individuals with greater amounts of AKL performed the SLSJ with a greater increase in PTASF compared to individuals with lesser AKL (P < .001). After controlling for initial body positioning, K_{HAM} was not a predictor of ACL-loading characteristics during the SLSJ in either sex. These results indicate that performing a stop-jump task on a single leg elicits characteristics associated with increased ligamentous loading and a landing posture that is more representative of what has been observed during injurious situations, and that the demands placed on the body during the SLSJ are greater for females compared to males. Thus, researchers are encouraged to use tasks that more closely mimic the conditions in which noncontact ACL injuries commonly occur, and employ sex-specific analyses, in future work. Additionally, although individuals with greater K_{HAM} have previously been reported to display biomechanical characteristics indicative of lesser ACL loading during non-weight bearing perturbations and double-leg jump-landings, K_{HAM} was not found to be a significant predictor of ACL-loading characteristics in either sex during the SLSJ in the current study. While these conflicting findings may indicate that the hamstrings ability to resist sagittal-plane ACL loading characteristics is negated when landing on a single leg, due to a more upright landing style, future studies are

needed to further elucidate the functional role of the hamstrings in resisting sagittal-place ACL loading characteristics when landing on a single leg in a more upright position.

THE INFLUENCE OF HAMSTRING MUSCULO-ARTICULAR STIFFNESS ON BIOMECHANICAL FACTORS INDICATIVE OF ANTERIOR CRUCIATE LIGAMENT LOADING

by

Justin Phillip Waxman

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by	Approved by
e Chair	Committee Cha



To Beth, Paul, and Jordan:

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APPROVAL PAGE

Committee Chair _____

This dissertation written by JUSTIN PHILLIP WAXMAN has been approved by the following committee of the Faculty of The Graduate School at The University of North Carolina at Greensboro.

	Dr. Sandra J. Shultz
Committee Members	
	Dr. Randy J. Schmitz
	Dr. J. Troy Blackburn
	Dr. Christopher K. Rhea
	Dr. Robert J. Henson
	Di. Robert J. Henson
Date of Acceptance by Committee	
Date of Final Oral Examination	
Date of I mai of at Enammation	

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CHAPTER I

INTRODUCTION

Noncontact anterior cruciate ligament (ACL) injuries often occur as the relatively extended knee (< 30° flexion) transitions from non-weight bearing to weight bearing upon initial ground contact during cutting and jump-landing maneuvers (Boden, Dean, Feagin, & Garrett, 2000; Koga et al., 2010; Krosshaug et al., 2007; Olsen, Myklebust, Engebretsen, & Bahr, 2004). Although the precise mechanism(s) of injury likely involves a combination of anterior tibial translation, knee valgus, and internal tibial rotation; anterior tibial translation represents the most direct loading mechanism to the ACL (Butler, Noyes, & Grood, 1980; Markolf et al., 1995). Research demonstrates that anterior tibial translation naturally occurs as the knee transitions from non-weight bearing to weight bearing in a relatively extended position (<30° flexion) (Fleming, Renstrom, Beynnon, et al., 2001; Torzilli, Deng, & Warren, 1994). This occurs for two reasons. First, ground reaction forces induce a compressive axial load that acts through the posterior- and inferiorly-directed slope of the tibial plateau to cause anterior tibial acceleration and proximal tibia anterior shear force, and thus anterior tibial translation (McLean, S. G., Lucey, Rohrer, & Brandon, 2010; McLean, S. G. et al., 2011; Meyer & Haut, 2005; Schmitz, Kim, & Shultz, 2010; Torzilli et al., 1994). Second, these ground reaction forces also produce an external knee flexion moment that must be counteracted by a quadriceps-generated internal knee extension moment to stabilize the knee and control the downward acceleration of the body (Blackburn & Padua, 2009; Yu, Lin, & Garrett, 2006). At more extended knee angles, contraction of the quadriceps muscles act through the anteriorly-oriented patellar tendon to create additional proximal tibia anterior

shear and anterior tibial translation, further loading the ACL (DeMorat, Weinhold, Blackburn, Chudik, & Garrett, 2004; Li et al., 1999; Withrow, Huston, Wojtys, & Ashton-Miller, 2006).

The hamstring muscles function antagonistically to the quadriceps, and proximal tibia anterior shear loading, by resisting anterior and rotary tibiofemoral motion (Victor, Labey, Wong, Innocenti, & Bellemans, 2010). Therefore, it seems intuitive that functional athletic tasks which require considerable quadriceps activation, such as landing from a jump, should be accompanied by adequate co-contraction of the hamstring muscles to resist proximal anterior tibial shear force and consequently minimize ACL loading. To this end, several in-vivo (Baratta et al., 1988; Beynnon et al., 1995; Solomonow et al., 1987), in-vitro (Draganich & Vahey, 1990; Li et al., 1999; MacWilliams, Wilson, DesJardins, Romero, & Chao, 1999; Victor et al., 2010; Withrow et al., 2006), and musculoskeletal modeling studies (Imran & O'Connor, 1998; Kellis, 1998; Pandy & Shelburne, 1997) demonstrate that adequate co-contraction of the hamstrings can effectively reduce the net anterior shear forces placed on the tibia, subsequently reducing ACL loading. Specifically, these studies have demonstrated that peak anterior tibial translation and ACL loading occur when the knee is in 15-30° of flexion and an isolated quadriceps contraction is applied (Beynnon & Fleming, 1998; Beynnon et al., 1995; Draganich & Vahey, 1990; Fujiya, Kousa, Fleming, Churchill, & Beynnon, 2011); and that when a hamstring contraction is then applied, anterior tibial translation and ACL loading are reduced at knee flexion angles greater than 10-15° (Draganich & Vahey, 1990; MacWilliams et al., 1999; Pandy & Shelburne, 1997; Withrow et al., 2006; Withrow, Huston, Wojtys, & Ashton-Miller, 2008). However, due to the inherent difficulties associated with measuring muscle forces and ACL loading *in-vivo*, the demonstrated effects of hamstring co-contraction on ACL loading have been limited to cadaver models and musculoskeletal modeling simulation studies, or in-vivo during isometric kneeextension exercises. Therefore, the true extent to which the hamstrings effectively reduce ACL loading during functional athletic tasks remains unknown.

It is well accepted that preparatory muscle activation occurs in anticipation of initial ground contact during functional athletic tasks such as landing from a jump. This preparatory muscle activation increases overall joint stiffness and is thought to enhance functional knee stability (Bryant, Creaby, Newton, & Steele, 2008; McNair & Marshall, 1994; Swanik, Lephart, Swanik, Stone, & Fu, 2004). Because noncontact ACL injuries occur within the first 10 to 50 milliseconds following initial ground contact (Koga et al., 2010; Krosshaug et al., 2007), any imbalance or delay in preparatory muscle activation may lead to improper limb positioning and higher ACL loading, increasing the risk of injury. In this regard, a property of the hamstring muscles that may play a critical role in helping resist the biomechanical factors reported to contribute to ACL loading (i.e. proximal tibia anterior shear force, anterior tibial acceleration, anterior tibial translation) is hamstring musculo-articular stiffness. Hamstring musculo-articular stiffness ($K_{\rm HAM}$) is a modifiable neuromechanical property that quantifies the resistance of the hamstring musculo-articular unit to lengthening in response to an applied load (Blackburn & Norcross, 2014). Research demonstrates that K_{HAM} is positively related to neuromuscular activation levels (Ditroilo, Watsford, & De Vito, 2011; Jennings & Seedhom, 1998). Therefore, it is theorized that for a given load, relatively stiffer hamstrings will permit a smaller change in length compared to more compliant hamstrings, thus limiting tibiofemoral joint motion and the biomechanical factors that contribute to ACL loading.

There is currently a small, but growing body of literature to support the theory that K_{HAM} may play a critical role in ACL loading by helping control tibiofemoral motion. Research demonstrates that ACL-deficient individuals with higher levels of K_{HAM} possess greater functional knee stability than more compliant individuals (McNair, Wood, & Marshall, 1992), which

suggests that K_{HAM} may help supplement the stability roles of the native ACL. In addition, healthy (uninjured) individuals with higher levels of K_{HAM} are shown to display less anterior tibial translation (Blackburn, Norcross, & Padua, 2011) and proximal tibia anterior shear force (Blackburn, Norcross, Cannon, & Zinder, 2013) during controlled perturbations and double-leg landing tasks, respectively. Further, females display less K_{HAM} (Blackburn, Bell, Norcross, Hudson, & Kimsey, 2009; Blackburn & Pamukoff, 2014; Blackburn, Riemann, Padua, & Guskiewicz, 2004; Granata, Wilson, & Padua, 2002), perform dynamic landing tasks with greater proximal anterior tibial shear force (Chappell, Yu, Kirkendall, & Garrett, 2002; Sell et al., 2007; Yu et al., 2006), and are at substantially greater risk of experiencing a noncontact ACL injury (Arendt, E. & Dick, 1995), compared to similarly trained males. Thus, it appears that insufficient K_{HAM} may influence an individual's functional knee stability and risk for noncontact ACL injury.

Statement of Problem

Although a direct link between K_{HAM} and noncontact ACL injury risk has yet to be established, there is evidence to suggest that higher levels of K_{HAM} may protect the ACL from deleterious loading during the early phase of landing, the time at which such injuries are reported to occur (Blackburn et al., 2013; Blackburn et al., 2011). However, current evidence regarding the influence of K_{HAM} on knee joint biomechanics is limited to studies of non-weight bearing perturbations (Blackburn et al., 2011) and double-leg drop-jump landings (Blackburn et al., 2013). Research demonstrates that noncontact ACL injuries most often occur when cutting or landing on a single leg (Boden et al., 2000; Boden, Torg, Knowles, & Hewett, 2009; Hewett, Torg, & Boden, 2009; Koga et al., 2010; Olsen et al., 2004), and that large asymmetries in weight-distribution are present when these injuries occur during double-leg landings (Hewett et al., 2009; Olsen et al., 2004). Additionally, laboratory-based studies show that during single-leg

landing tasks, individuals land with larger ground reaction forces and internal knee extension moments, smaller hip and knee flexion angles and slower hip and knee flexion angular velocities at initial ground contact, and greater proximal tibia anterior shear force, compared to double-leg landing tasks (Pappas, Hagins, Sheikhzadeh, Nordin, & Rose, 2007; Wang, I. L., Wang, & Wang, 2015; Wang, L. I., 2011; Yeow, Lee, & Goh, 2010). Thus, open-kinetic-chain perturbations and double-leg jump-landings may not adequately represent the situations in which noncontact ACL injuries commonly occur. There are also methodological differences in the way that K_{HAM} has been assessed in previous work. For example, some studies have assessed K_{HAM} by standardizing the assessment load as a percentage of an individual's body mass, whereas other have standardized the assessment load as a percentage of an individual's maximal isometric hamstring torque. Because K_{HAM} is influenced by neuromuscular activation levels (Ditroilo et al., 2011; Jennings & Seedhom, 1998), the ability to make comparisons between studies that have used different methods of standardizing the applied load is limited. Furthermore, the influence of K_{HAM} on measures of ACL loading has been established with males and females in the same statistical analyses without equal sex-stratification. This is despite females having less K_{HAM} (Blackburn et al., 2009; Blackburn & Pamukoff, 2014; Blackburn et al., 2004; Granata et al., 2002), and displaying a more posterior center of mass position (DiStefano, Padua, Prentice, Blackburn, & Keras, 2005; Yu et al., 2006), less hip and knee flexion (Schmitz, Kulas, Perrin, Riemann, & Shultz, 2007), higher quadriceps and lower hamstring muscle activation (Malinzak, Colby, Kirkendall, Yu, & Garrett, 2001), and greater posterior ground reaction forces and knee extensor moments (Schmitz et al., 2007), during functional athletic tasks than males. This creates a problem, because the combination of peak posterior ground reaction force, knee extensor moment, knee flexion angle, quadriceps muscle activation, and sex, has been shown to account for 86.1% of the variance in proximal tibia anterior shear force during a vertical stop-jump task

(Sell et al., 2007). Therefore, the true extent to which K_{HAM} is associated with biomechanical factors that directly influence ACL loading (i.e. proximal tibia anterior shear force, anterior tibial translation, and anterior tibial acceleration) once other sex dependent factors are accounted for during functional landing tasks remains unknown.

Objectives and Hypotheses

The primary objective of this study was to determine, within each sex, the extent to which K_{HAM} predicts biomechanical factors indicative of sagittal plane ACL loading during a functional single-leg stop-jump landing task. This was accomplished through the following aims and hypotheses:

Aim 1: To examine the effects of landing type (double-leg/single-leg) and sex (male/female) on neuromuscular (i.e. preparatory muscle activation of the quadriceps and hamstring muscles) and biomechanical variables (i.e. trunk center of mass position and hip and knee flexion angles at initial ground contact; trunk center of mass position and hip and knee flexion excursions; average hip and knee flexion angular velocities throughout landing; and peak posterior and vertical ground reaction forces, peak knee extensor moment, peak proximal tibia anterior shear force, peak anterior tibial acceleration, and peak anterior tibial translation throughout landing) during a stop-jump landing task.

Hypothesis 1a: Compared to the double-leg stop-jump, the single leg stop-jump will elicit a more upright landing posture (i.e. a more posteriorly-oriented trunk center of mass position and less hip and knee flexion at initial ground contact), slower hip and knee flexion angular velocities, smaller trunk center of mass position and hip and knee flexion excursions, larger posterior and vertical ground reaction forces and knee extensor

moments, greater preparatory muscle activation, and biomechanical factors indicative of greater ACL loading (i.e. greater peak proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation).

Hypothesis 1b: During both tasks, females will display a more upright landing posture (i.e. a more posteriorly-oriented trunk center of mass position and less hip and knee flexion at initial ground contact), slower hip and knee flexion angular velocities, trunk center of mass position and hip and knee flexion excursions, larger posterior and vertical ground reaction forces and knee extensor moments, greater preparatory muscle activation, and biomechanical factors indicative of greater ACL loading (i.e. greater peak proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation), compared to males.

Aim 2: To determine, within each sex, the extent to which K_{HAM} predicts biomechanical factors indicative of sagittal plane ACL loading (i.e. peak proximal tibia anterior shear force, peak anterior tibial acceleration, and peak anterior tibial translation) during a single-leg stop-jump landing task; and whether the extent to which K_{HAM} predicts biomechanical factors indicative of sagittal plane ACL loading is influenced by the way in which K_{HAM} is measured (i.e. assigning the applied load as a fixed percentage of body mass or as a fixed percentage of maximal voluntary isometric torque).

Hypothesis 2a: After controlling for body positioning at initial ground contact (i.e. trunk center of mass position and hip and knee flexion angles), higher K_{HAM} values will be predictive of biomechanical characteristics indicative of lower sagittal plane ACL loading during landing (i.e. less proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation) within each sex.

Hypothesis 2b: The extent to which K_{HAM} predicts biomechanical factors indicative of sagittal plane ACL loading will be dependent on the method by which K_{HAM} is measured (i.e. assigning the applied load as a fixed percentage of body mass or as a fixed percentage of maximal voluntary isometric torque). Specifically, it is hypothesized that K_{HAM} will be more predictive of biomechanical factors indicative of sagittal plane ACL loading when K_{HAM} is assessed using an applied load standardized as a percentage of peak isometric torque versus a percentage of body mass.

Aim 3: To determine the extent to which K_{HAM} independently predicts biomechanical factors indicative of sagittal plane ACL loading (i.e. proximal tibia anterior shear force, anterior tibial translation, and anterior tibial acceleration) during a single-leg stop-jump landing, once other known neuromuscular and biomechanical characteristics are accounted for. These neuromuscular and biomechanical characteristics include preparatory quadriceps muscle activation, peak posterior ground reaction force, knee flexion angle at the time of peak posterior ground reaction force, and knee extensor (internal) moment at the time of peak posterior ground reaction force.

Hypothesis 3a: The linear combination of peak posterior ground reaction force, knee extensor moment, knee flexion angle, preparatory quadriceps muscle activation, and sex, will be highly predictive of biomechanical factors indicative of sagittal plane ACL loading. This hypothesis is based on the previous work of Sell et al (Sell et al., 2007), who demonstrated greater preparatory quadriceps muscle activation, peak posterior ground reaction force, external knee flexion moment, and knee flexion angle, and sex (being female), significantly predicted greater proximal tibia anterior shear force during a double-leg stop jump landing task.

Hypothesis 3b: Hamstring musculo-articular stiffness (K_{HAM}) will be a significant independent predictor in the final regression model when added to the pool of possible predictors, with higher K_{HAM} being predictive of biomechanical characteristics indicative of lower sagittal plane ACL loading (i.e. less proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation).

Limitations and Assumptions

- 1. All participants exerted maximal effort during all experimental testing procedures.
- 2. The Phase Space IMPULSE three-dimensional motion tracking system and Bertec force platforms are valid and reliable devices for kinematic and kinetic measurements, respectively.
- The representation of the foot, shank, thigh, and trunk, as rigid segments, are accurate depictions of the motion occurring during athletic movements.
- 4. Inverse dynamics calculations are representative of the total moments occurring at the joint.
- Electromyography analysis by way of the surface electrodes (sEMG), using the Delsys
 Trigno system, is a valid and reliable device for the assessment of neuromuscular activation timing and amplitude.
- 6. The neuromuscular activity (i.e. sEMG amplitude) obtained at each muscle site is representative of the total activity throughout the entire muscle.
- The functional landing tasks used in this study (i.e. single-leg and double-leg stop-jump landing) adequately simulate the situations in which non-contact ACL injuries commonly occur.
- 8. The in-vivo assessment of hamstring musculo-articular stiffness (K_{HAM}) via free-oscillation results in a global stiffness measurement, which includes contributions from the hamstring muscle-tendon unit(s), skin, ligament(s), bone(s), and articular joint capsule.

- 9. Hamstring musculo-articular stiffness (K_{HAM}) is effectively represented by a spring-mass model.
- 10. Results from this study are most generalizable to healthy, highly-active, college-aged males and females, who regularly participate in activities that involve running, cutting, jumping, and landing (e.g. basketball, soccer, tennis, rugby, and volleyball), and caution should be taken when attempting to generalize these results to other populations.
- 11. Biomechanical assessments were performed in a standard laboratory setting, which may elicit different kinematic and kinetic measurements than what would be observed during actual practice and competition.
- **12.** Due to the *in-vivo* nature of this study, it is not possible to measure anterior cruciate ligament (ACL) loading directly.

Delimitations

- Participant recruitment was limited to healthy, highly-active, college-aged males and females, who regularly participated in activities that involved running, cutting, jumping, and landing (e.g. basketball, soccer, tennis, rugby, and volleyball).
- 2. Participants were considered healthy, as defined by: 1) no history of injury to the anterior or posterior cruciate ligaments (ACL and PCL, respectively), the medial or lateral collateral ligaments (MCL and LCL, respectively), or the medial or lateral menisci, 2) no history of lower extremity surgery, 3) no history of lower-extremity injury within 6 months prior to recruitment, and 4) no known medical conditions that would affect their connective tissue or vestibular system.

- 3. Participants were considered highly-active, as defined by engaging in greater than the equivalent of 300 minutes of moderate-intensity physical activity per week (ACSM's Health-Related Physical Fitness Assessment Manual, 2013).
- 4. In order to control for potential effects of cycling hormones on knee joint laxity, stiffness, and landing biomechanics, all testing for eumenorrheic female participants were constrained to the follicular phase of their menstrual cycle when hormones are most stable and at their nadirs (Days 1-8 following self-reported onset of menstrual bleeding).
- 5. Anterior knee laxity has been reported to influence anterior tibial translation during weight acceptance (Shultz, S. J., Shimokochi, et al., 2006). It has also been reported that characteristics of the load-displacement curve (anterior knee laxity and stiffness) influence knee anterior shear forces during double-leg drop-jump landings (Schmitz, Sauret, & Shultz, 2013). Therefore, anterior knee laxity was collected in order to account for passive restraint characteristics that potentially influence stop-jump landing biomechanics.
- 6. It was expected that all participants would be able to successfully and consistently complete all experimental testing procedures following familiarization.
- 7. All participants were standardized shoes and clothing during the experimental testing session, and all biomechanical landing assessments were performed on the same laboratory surface.

Operational Definitions

Anterior Knee Laxity (AKL): The anterior displacement (mm) of the tibia relative to the femur when subjected to an anterior-directed load of 133 N, applied to the posterior proximal tibia, with the knee placed in $25 \pm 5^{\circ}$ of flexion.

<u>Center of Pressure (COP)</u>: The planar point location of the vertical ground reaction force vector. <u>Center of Mass (COM)</u>: The planar point location about which the body's mass is equally distributed.

Double-Leg Stop Jump (DLSJ) Task: A task which involves a double-leg horizontal jump onto two force platforms from a distance equal to 40% of the participant's standing height, immediately followed by a maximum vertical jump and subsequent double-leg landing. Hamstring Musculo-Articular Stiffness (K_{HAM}): An in-vivo measure of active stiffness that quantifies the resistance of the hamstring musculo-articular unit(s) to lengthening in response to an applied load. Specifically, K_{HAM} is assessed via the free-oscillation technique, whereby the leg is modeled as a single-degree-of-freedom mass-spring system, and the damping effect that the hamstring impose on oscillatory flexion-extension at the knee joint is quantified, following a brief manual perturbation. The derived value of K_{HAM} is then normalized by being divided by the participant's body mass (expressed in units of $N \cdot m^{-1} \cdot kg^{-1}$).

Healthy: An individual with: 1) no history of injury to their anterior or posterior cruciate ligaments (ACL and PCL, respectively), their medial or lateral collateral ligaments (MCL and LCL, respectively), or their medial or lateral menisci, 2) no history of lower extremity surgery, 3) no history of lower-extremity injury within 6 months of recruitment, and 4) no known medical conditions that would affect their connective tissue or vestibular system.

<u>Initial Ground Contact (IC)</u>: The point in time when the vertical ground reaction force exceeds 10 newtons (N).

<u>Landing Phase</u>: The time-interval from initial contact to maximal descent during each landing task.

Maximal Descent: The time point at which the participant's center of mass (CoM) reaches its lowest position.

Maximal Voluntary Isometric Contraction (MVIC) Peak RMS sEMG Amplitude: The peak RMS sEMG amplitude obtained from each individual quadriceps (vastus medialis and vastus lateralis) and hamstring muscle (semitendinosus and biceps femoris) during a 3-second maximal-effort isometric contraction with the hip and knee fixed in 30° of flexion, averaged across three trials.

Maximal Voluntary Isometric Contraction (MVIC) Peak Torque: The peak torque produced by the quadriceps and hamstring muscles during 5-second maximal-effort isometric quadriceps and hamstring contractions, respectively.

<u>Muscle Pre-Activation</u>: The mean RMS sEMG amplitude obtained from a given muscle 150 milliseconds prior to initial ground contact, normalized to the MVIC peak RMS sEMG amplitude from the same muscle, and expressed as a percentage of MVIC (%MVIC).

<u>Single-Leg Stop Jump (SLSJ) Task</u>: A task which involves a single-leg horizontal jump onto a force platform from a distance equal to 40% of the participant's standing height, immediately followed by a maximal single-leg vertical jump and subsequent single-leg landing.

Independent and Dependent Variables

<u>Anterior Tibial Acceleration (ATA)</u>: The peak anterior acceleration (m/s²) of the proximal tibia recorded during the landing phase.

<u>Anterior Tibial Translation (ATT)</u>: The peak anterior displacement (mm) of the proximal tibia relative to the femur during the landing phase.

<u>Average Hip Flexion Angular Velocity (HFV)</u>: The sagittal plane angular velocity (°/s) of the femur relative to the pelvis averaged across the landing phase.

<u>Average Knee Flexion Angular Velocity (KFV)</u>: The sagittal plane angular velocity (°/s) of the tibia relative to the femur averaged across the landing phase.

<u>Hamstring Muscle Pre-Activation (HAM_{PRE})</u>: A composite average of the muscle pre-activation of the medial and lateral hamstrings (semitendinosus and biceps femoris, respectively), expressed as a percentage of MVIC (%MVIC).

<u>Hamstring Musculo-articular Stiffness - Body Mass (K_{HAM}_BM)</u>: The K_{HAM} value obtained when assessed using an applied load equal to 10% of the participant's body mass (expressed in units of $N \cdot m^{-1} \cdot kg^{-1}$).

<u>Hamstring Musculo-articular Stiffness - MVIC (K_{HAM} MVIC)</u>: The K_{HAM} value obtained when assessed using an applied load equal to 30% of the participant's MVIC peak hamstring torque (expressed in units of $N \cdot m^{-1} \cdot kg^{-1}$).

<u>Hip Flexion Excursion (HF_{EXC})</u>: The sagittal plane angle (°) of the femur relative to the pelvis at peak minus the sagittal plane angle (°) of the femur relative to the pelvis at initial contact.

<u>Initial Hip Flexion Angle (HF_{IC})</u>: The sagittal plane angle ($^{\circ}$) of the femur relative to the pelvis at initial contact.

Initial Knee Flexion Angle (KF_{IC}): The sagittal plane angle (°) of the tibia relative to the femur at initial contact.

<u>Initial Trunk COM Position (TrunkCOM_{IC}):</u> The anterior-posterior distance (cm) of the trunk's center of mass (COM) relative to the center of pressure (COP) at initial contact.

Knee Extension Moment at Peak Posterior Ground Reaction Force (KEM_{PKpGRF}): The internal moment acting about the medial-lateral axis of the knee joint at the time of peak posterior ground reaction force, normalized to the product of body height and weight ($N \cdot m \cdot BW^{-1} \cdot Ht^{-1}$).

Knee Flexion Angle at Peak Posterior Ground Reaction Force (KF_{PKpGRF}): The sagittal plane angle (°) of the tibia relative to the femur at the time of peak posterior ground reaction force.

<u>Knee Flexion Excursion (KF_{EXC}):</u> The sagittal plane angle (°) of the tibia relative to the femur at peak minus the sagittal plane angle (°) of the tibia relative to the femur at initial contact.

<u>Landing Type:</u> Single-leg versus double-leg stop-jump landing tasks.

<u>Peak Hip Extension Moment (KEM_{PEAK}):</u> The peak internal moment acting about the mediallateral axis of the hip joint, normalized to the product of body height and weight ($N \cdot m \cdot BW^{-1} \cdot Ht^{-1}$).

<u>Peak Knee Extension Moment (KEM_{PEAK})</u>: The peak internal moment acting about the medial-lateral axis of the knee joint, normalized to the product of body height and weight ($N \cdot m \cdot BW^{-1} \cdot Ht^{-1}$).

<u>Peak Posterior Ground Reaction Force (pGRF_{PEAK}):</u> The peak ground reaction force in the posterior direction, recorded during the landing phase, normalized to body weight (BW).

<u>Proximal Tibia Anterior Shear Force (PTASF)</u>: The peak net anterior shear force at the proximal tibia during the landing phase, normalized to body weight (BW).

<u>Peak Vertical Ground Reaction Force (vGRF_{PEAK}):</u> The peak ground reaction force in the vertical direction, recorded during the landing phase, normalized to body weight (BW).

Quadriceps Muscle Pre-Activation (QUAD_{PRE}): A composite average of the muscle pre-activation of the medial and lateral quadriceps (vastus medialis and vastus lateralis, respectively), expressed as a percentage of MVIC (%MVIC).

<u>Sex</u>: The sex of the participant (male or female).

<u>Trunk COM Position Excursion (TrunkCOM_{EXC}):</u> The peak anterior-posterior distance (cm) of the trunk's center of mass (COM), relative to the center of pressure (COP), during the landing phase minus the anterior-posterior distance of the trunk's COM, relative to the COP, at initial contact.

CHAPTER II

REVIEW OF THE LITERATURE

Introduction

Anterior cruciate ligament (ACL) injuries are estimated to affect more than 100,000 individuals annually in the United States, with the majority of these injuries occurring in young athletes between 15 and 25 years of age (Griffin et al., 2006; Majewski, Susanne, & Klaus, 2006; Prodromos, Han, Rogowski, Joyce, & Shi, 2007). These injuries are accompanied by high financial costs due to surgical reconstruction and rehabilitation (Brophy, Wright, & Matava, 2009; Mather et al., 2013), and can often result in a number of undesirable consequences, including long-term disability and the early development of knee osteoarthritis, an increased risk of re-injury, and a reduced likelihood of returning to pre-injury levels of competition (Ardern, Taylor, Feller, Whitehead, & Webster, 2015; Lohmander, Ostenberg, Englund, & Roos, 2004; Neuman et al., 2008; Oiestad, Holm, Engebretsen, & Risberg, 2011; Wright et al., 2007). Because over two thirds of all ACL injuries are noncontact in nature, in that they occur in the absence of physical contact with another individual or object (Boden et al., 2000; Ferretti, Papandrea, Conteduca, & Mariani, 1992; Krosshaug et al., 2007), it is thought that some of these injuries may be prevented through targeted intervention strategies. As such, identifying modifiable factors that contribute to noncontact ACL injury risk has been a major focus of research over the past 15 years. However, the most appropriate risk factors to be targeted through injury prevention efforts have yet to be fully elucidated.

The purpose of the following review of literature is to support the theoretical framework that hamstring musculo-articular stiffness (K_{HAM}) may play a critical role in ACL loading, and

thus noncontact ACL injury risk, by helping control tibiofemoral motion during functional athletic tasks that are representative of the situations in which noncontact ACL injuries commonly occur. Specifically, this review aims to present and summarize what is currently known about the mechanism(s) of noncontact ACL injury, the factors that contribute to dynamic knee stability and ACL loading, and the potential role of K_{HAM} in contributing to ACL loading.

Mechanism(s) of Noncontact Anterior Cruciate Ligament Injury

For ethical reasons, *in-vivo* measurements of ACL loading to failure are not possible to obtain. Consequently, current evidence regarding the potential mechanism(s) by which noncontact ACL injury occurs has largely been limited to retrospective interviews with ACL-injured individuals and video analyses of actual injuries recorded during training (practice) or competition (games or matches). Such investigations have used this information to characterize the situations in which noncontact injuries most commonly occur, and to subsequently propose the potential mechanism(s) of injury. However, retrospective interviews and video analyses are limited due to the fact that the precise mechanism(s) of injury likely involves a complex interaction between muscle forces, external forces, and joint contact forces (Ali & Rouhi, 2010), which cannot be obtained from such methods. Therefore, *in-vitro* and *in-vivo* studies have also been conducted to gain a better understanding of the knee joint positions that load the ACL and thereby place the ligament at increased risk for injury.

Anterior Cruciate Ligament Structure and Function

Prior to discussing the proposed mechanism(s) of injury and what is currently known about ACL loading, it is important for first have a general understanding of the structural anatomy and function of the ACL. The knee joint is the largest and possibly the most complex

synovial joint in the human body, with three bony articulations (i.e. patella-femoral, medial tibiofemoral, and lateral tibio-femoral) and six degrees-of-freedom (i.e. flexion-extension, internalexternal rotation, varus-valgus angulation, anterior-posterior translation, medial-lateral translation/shift, and compression and distraction) (Woo, Debski, Withrow, & Janaushek, 1999). Collectively, the ligaments of the knee help passively restrain excessive joint motion in order to maintain knee stability (Noyes, Grood, Butler, & Malek, 1980). As described by Arnoczky (Arnoczky, 1983), the ACL originates on the posterior aspect of the medial surface of the lateral femoral condyle, and then travels anteriorly, medially, and distally across the knee joint as it passes from the femur to its insertion site on the tibia, just lateral and anterior to the tibial spine. The ACL itself consists of two distinct bundles; the anteromedial bundle, which originates on the proximal aspect of the femoral attachment and inserts on the anteromedial aspect of the tibial attachment; and the posterolateral bundle, which originates on the proximal aspect of the femoral attachment and inserts on the posterolateral aspect of the tibial attachment (Arnoczky, 1983). In terms of knee joint function, the ACL (both the anteromedial and posterolateral bundles) has been shown to resist anterior tibial translation (i.e. anterior translation of the tibia relative to the femur), internal tibial rotation (i.e. internal rotation of the tibia relative to the femur), and hyperextension of the tibiofemoral joint (< 0° knee flexion) (Ahmed, Burke, Duncan, & Chan, 1992; Ahmed, Hyder, Burke, & Chan, 1987; Butler et al., 1980; Markolf et al., 1995; Vahey & Draganich, 1991). When the knee joint is fully extended ($\sim 0^{\circ}$ knee flexion), the posterolateral bundle is taut while the anteromedial bundle is relatively slack; however, as the knee begins to flex, the femoral attachment of the ACL becomes more horizontally aligned, which causes the anteromedial bundle to tighten and the posterolateral bundle to loosen (Girgis, Marshall, & Monajem, 1975). Together, the presence of these two distinct bundles allows for different portions of the ACL to remain taut throughout the full joint range of motion (Welsh, 1980); and it is clinically important to understand that the ACL is under tension prior to the application of any external loading.

Retrospective Interview

In an early effort to gain an understanding of the potential mechanism(s) of noncontact ACL injury, several researchers conducted retrospective interviews on ACL-injured individuals (Boden et al., 2000; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; McNair, Marshall, & Matheson, 1990; Olsen et al., 2004). From these studies, it has been reported that approximately 70% of all ACL injuries occur in noncontact situations, and that such injuries are more likely to occur during competition than during practice or training (Boden et al., 2000; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; McNair et al., 1990; Olsen et al., 2004). In addition, it has been found that noncontact ACL injuries typically occur during movements that involve a sudden deceleration of the body, with or without a change in direction, such as when cutting to quickly evade an opponent (Faunø & Wulff Jakobsen, 2006; Olsen et al., 2004) or when landing from a jump on one or two legs (Boden et al., 2000; Ferretti et al., 1992; Olsen et al., 2004). Further, these studies have been able to gain a general understanding of the injured individuals' body positioning at the time of injury. The most common traits described among injured individuals are that their knee was in a relatively extended position (between 20° flexion and full extension), and that the foot of their injured leg was in contact with the ground, at the time of injury (Boden et al., 2000; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; McNair et al., 1990; Olsen et al., 2004). In contrast, there has been much more variability reported in terms of frontal and transverse plane knee motions at the time of injury. Specifically, some individuals reported injury to occur with either internal or external rotation of the tibia with the knee relatively extended (McNair et al., 1990), varus (i.e., knee adduction) or valgus (i.e., knee abduction) collapse with

the knee relatively extended (Boden et al., 2000), or a combination varus/valgus collapse and internal/external rotation of the tibia with the knee relatively extended (Ferretti et al., 1992). Although these studies provide valuable initial insight to the potential mechanisms involved in noncontact ACL injury, they are limited by the fact that they solely rely on the ability of the injured individual to recall the situations in which their injury occurred. It has been pointed out by Krosshaug and colleagues (Krosshaug, Andersen, Olsen, Myklebust, & Bahr, 2005) that even in the event that an athlete is able to describe the injury situation, the athlete's description may be influenced by what they were told by others witnessing the event (e.g., coaches, parents, teammates).

Video Analyses

In an effort to more objectively examine the potential mechanism(s) of noncontact ACL injury, some studies have performed descriptive analyses on video recordings of actual injuries (Boden et al., 2000; Boden et al., 2009; Cochrane, Lloyd, Buttfield, Seward, & McGivern, 2007; Koga et al., 2010; Krosshaug et al., 2007; Olsen et al., 2004). In agreement with the findings of retrospective interviews, these video analyses have also reported that noncontact ACL injuries most commonly occurred during movements that involved a sudden deceleration of the body, with or without a change in direction, such as cutting and jump-landing maneuvers. For example, Boden et al (Boden et al., 2000) analyzed video recordings of 23 ACL injuries from American football, soccer, basketball, and volleyball, and reported that sharp decelerations with or without a change of direction accounted for 67% of all injuries analyzed, while single- and double-leg landings accounted for the remaining 20% and 13%, respectively. However, findings from other studies on more homogenous populations tend to suggest that the type of movement that most commonly results in noncontact ACL injury may differ by sport. In a video analysis of ACL

injuries in team handball, it has been reported injuries most commonly occurred during singleand double-leg plant-and-cut (or side-cutting) maneuvers, followed by single-leg landings and
sharp decelerations on a single-leg without a change of direction (Olsen et al., 2004). Similar
findings were found by Cochrane et al (Cochrane et al., 2007) in an analysis of noncontact
injuries in Australian rules football. In contrast, two other studies analyzing ACL injuries in
basketball reported that 60% to 87% of all injuries occurred during single- and double-leg jump
landings (Boden et al., 2009; Krosshaug et al., 2007), with single-leg landings accounting for up
to 90% of all jump landing injuries (Boden et al., 2009). Furthermore, it is worth noting that even
though double-leg plant-and-cut and jump landing injuries are commonly reported, it has been
demonstrated that such injuries often involve large between-limb asymmetries in bodyweight
distribution, with the injured limb bearing a majority of the weight (65% to 100%) at the time of
injury (Olsen et al., 2004). Thus, double-leg landings may actually be more representative of
single-leg landings.

Findings from video analyses have also largely been in agreement with retrospective interview studies in terms of body positioning at the time of injury. The most common characteristics shared by these studies are that injuries occurred upon initial foot contact with the ground with the knee in a relatively extended position (range, 5°-30° knee flexion) (Boden et al., 2000; Boden et al., 2009; Krosshaug et al., 2007; Olsen et al., 2004). However, there has been a lack of agreement with regard to frontal and transverse plane knee motion at the time of injury, which may also be due to between-sport differences in the movements that most often result in injury. When analyzing frontal and transverse plane knee motions during injuries that occurred during a side-cut maneuver, Olsen et al (Olsen et al., 2004) proposed that the mechanism of injury involved knee valgus (5-20°), combined with either internal or external rotation (15° internal - 10° external) of the tibia relative to the femur, with the knee near extension (5-20° knee

flexion). Cochrane et al (Cochrane et al., 2007) later reported that injuries resulting from a sidecut involved knee valgus and internal rotation of the tibia, with the knee in less than 30° degrees of flexion, but argued that knee valgus and internal rotation could occur either in combination or exclusively. When analyzing frontal and transverse plane knee motions during sharp decelerations and jump landings, some studies have proposed that the mechanism of injury involves knee valgus (3-15°) and external rotation (5-15°) of the tibia, with the knee near extension (5-27° knee flexion) (Krosshaug et al., 2007; Olsen et al., 2004), while others have proposed that such injuries involve pure knee valgus (Boden et al., 2000) or knee varus (Cochrane et al., 2007) on a relatively extended knee. Adding further insight to the potential mechanism(s) of injury, Boden et al (Boden et al., 2009) performed a video analysis which compared kinematic characteristics between injured athletes and uninjured controls performing similar maneuvers and found that, at initial ground contact, injured athletes landed with their ankles in less plantar-flexion than controls, which resulted in injured athletes tending to land flatfooted or on their rear-foot whereas uninjured athletes tended to land on their fore-foot or a combination of their fore-foot and mid-foot. However, there were no significant differences in knee flexion or valgus angles between injured and uninjured athletes (Boden et al., 2009).

Although the findings from video analyses have provided valuable insight to potential mechanisms of injury, findings from such studies have been limited to simple visual inspection, and the accuracy of this method for determining joint kinematics during actual injury situations has been shown to have poor accuracy even among experienced researchers (Krosshaug et al., 2007). In an effort to improve on the limitations of previous video analyses, Koga and colleagues (Koga et al., 2010) developed a model-based image matching technique, which allowed them to create skeletal models and extract joint kinematics from video recordings of 10 noncontact ACL injury situations in women's team handball and basketball. Using this model-based image

matching technique, it was found that, at initial ground contact, athletes tended to land with the knee relatively extended (23 $^{\circ}$ knee flexion), with 0° of knee valgus and the tibia externally rotated 5°(Koga et al., 2010). It was also found that 40 milliseconds after initial ground contact, knee flexion increased by 24°, knee valgus increased by 12° and the tibia internally rotated 8° (Koga et al., 2010). Then, from 40 to 300 milliseconds after initial ground contact, the tibia was reported to externally rotate 17°. The authors interpreted these findings to suggest that valgus loading may be a key factor in the mechanism of injury, and that knee valgus and internal tibial rotation are coupled motions (Koga et al., 2010). Koga et al (Koga et al., 2010) then combined their findings with those of previous video analyses, and those of *in-vitro* studies that will be discussed shortly, to propose a more robust potential mechanism of injury. Specifically, it was proposed that: 1) when valgus loading is applied, the medial collateral ligament becomes taut and lateral joint compression occurs; 2) this compressive load, as well as the anterior tibial shear force caused by quadriceps contraction, causes a displacement of the femur relative to the tibia (i.e. anterior tibial translation), where the lateral femoral condyle shifts posteriorly and the tibia translates anteriorly and rotates internally (i.e. internal tibial rotation), resulting in ACL rupture within the first 40 milliseconds following initial ground contact; 3) after the ACL is torn, the primary restraint to anterior translation of the tibia is gone, which causes the medial femoral condyle to also be displaced posteriorly and ultimately results in external rotation of the tibia relative to the femur (Figure 2.1) (Koga et al., 2010). Thus, the suspected external tibial rotation at the time of injury reported in some of the video analyses previously discussed (Krosshaug et al., 2007; Olsen et al., 2004) may have been observed after ACL rupture had already occurred.

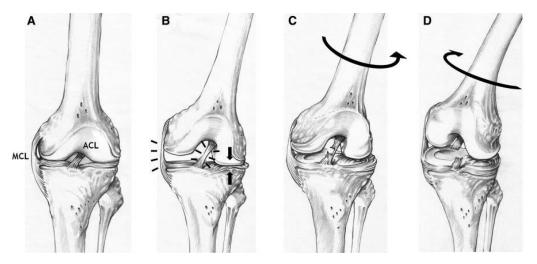


Figure 2.1. Proposed Mechanism of Injury Described by Koga et al (Koga et al., 2010). (A) unloaded knee; (B) when valgus loading is applied, the medial collateral ligament becomes taut and lateral compression occurs; (C) compressive load and quadriceps contraction causes displacement of femur relative to tibia, where lateral femoral condyle shifts posteriorly and the tibia translates anteriorly and rotates internally, resulting in ACL rupture; (D) After ACL rupture, primary restraint to anterior tibial translation is gone, causing the medial femoral condyle to also be displaced posteriorly and resulting in external rotation of tibia.

In-Vitro and In-Vivo Studies

The retrospective interviews and video analyses previously discussed have been able to provide valuable information regarding the type of movements that most often result in noncontact ACL injury, the timing of injury, and potential injury mechanisms. However, such studies have been unable to determine whether the knee joint kinematics observed at the time of injury are causative of the injury itself, or if the observed kinematics resulted from the injury. This is due to the fact that the precise mechanism(s) of injury likely involves a complex interaction between muscle forces, external forces, and joint contact forces (Ali & Rouhi, 2010), which cannot be obtained from such methods. What is currently known about the mechanism(s) of injury is also limited by the fact that ethical reasons prevent laboratory-based studies from loading the ACL to failure *in-vivo*. As such, the current body of knowledge on ACL loading and

injury risk is based on submaximal ACL loading *in-vivo*, maximal and submaximal ACL loading *in-vitro*, and computer simulation (musculoskeletal modeling) studies.

Early *in-vitro* studies have examined the effects of externally applied loads (external forces) on ACL loading by applying loads both in isolation and in combination. Such studies have demonstrated that when the knee is relatively extended (> 30° flexion) and an anterior-directed load is applied to the proximal tibia, more than 80% of the anterior-directed load is transferred to the ACL (Sakane et al., 1997; Takai, Woo, Livesay, Adams, & Fu, 1993; Woo et al., 1999). It has also been demonstrated that the load experienced by the ACL can often exceed the applied anterior tibial load as the knee approaches full extension (Markolf et al., 1995; Takai et al., 1993). Similarly, it has been reported that the ACL provides more than 80% of the total passive restraint to anterior tibial translation when the knee in fixed in 30° of flexion (Butler et al., 1980). Together these findings indicate that the ACL acts as the primary passive restraint to anterior-directed loads and motion when the knee is in a relatively extended position.

Other *in-vitro* studies have examined the strain or load response of the ACL during isolated internal-external tibial rotation and varus-valgus angulation. With the exception of one study (Berns, Hull, & Patterson, 1992), it has consistently been demonstrated that the ACL is loaded (or strained) when an internal tibial torque is applied to a relatively extended knee (Ahmed et al., 1987; Bach & Hull, 1998; Beynnon et al., 1995; Markolf et al., 1995; Markolf, Gorek, Kabo, & Shapiro, 1990). In contrast, only one study has reported that external tibial torque loads the ACL (Markolf et al., 1990), while others have reported no effect (Ahmed et al., 1987; Bach & Hull, 1998; Berns et al., 1992; Beynnon et al., 1995). Only two *in-vitro* studies have attempted to measure ACL loading in response to isolated varus-valgus angulation, and these studies have produced mixed results (Berns et al., 1992; Markolf et al., 1990). In terms of combined loading states, Markolf et al (Markolf et al., 1995) demonstrated that ACL loading was greatest when

internal tibial torque was combined with anterior tibial shear force and the knee was at full extension and hyperextension, and when a valgus moment was combined with anterior tibial shear force and the knee was in more than 10° of flexion.

Although the studies referenced above have provided valuable insight to the multi-planar role of the ACL in maintaining knee joint stability, it is important to note that such injuries have assessed ACL loading under non-weight bearing conditions, and often at static knee flexion angles. Given that retrospective studies have demonstrated that noncontact ACL injuries occur within the first 40 to 50 milliseconds following initial ground contact during cutting and landing maneuvers (Koga et al., 2010; Krosshaug et al., 2007), these findings indicate that such injuries occur as the knee initially transitions from non-weight bearing to weight bearing. As the knee transitions from non-weight bearing to weight bearing upon initial ground contact, resultant ground reaction forces induce compressive axial loading at the knee joint. As such, other studies have investigated knee joint kinematics and ACL loading in response to applied axial compressive knee joint loading in order to more closely simulate the compressive loads that the weight bearing knee experiences during dynamic tasks such as landing from a jump.

Using intact unconstrained (without simulated muscle forces) cadaveric knees, it has been demonstrated that axial compressive loading naturally results in anterior tibial translation, internal tibial rotation, and knee valgus, and that these combined motions load the ACL (Markolf, Jackson, Foster, & McAllister, 2014). Fleming et al (Fleming, Renstrom, Beynnon, et al., 2001) examined ACL strain *in-vivo* in response to isolated anterior-posterior shear forces, internal-external tibial torques, and varus-valgus moments, under both weight bearing and non-weight bearing conditions. In the non-weight bearing knee, ACL strain was reported to increase in response to anterior shear force and internal tibial torque, but not external tibial torque or varus-valgus moments (Fleming, Renstrom, Beynnon, et al., 2001). Anterior shear and internal tibial

torque were also reported to increase ACL strain in the weight bearing knee, and these strain values were significantly greater than those observed in non-weight bearing (Fleming, Renstrom, Beynnon, et al., 2001). Additionally, although ACL strain values were significantly greater during weight bearing compared to non-weight bearing, the application of varus-valgus moments did not affect ACL strain values; thus, the significant increase in ACL strain was thought to be caused by compressive loading and the response of the knee joint musculature (Fleming, Renstrom, Beynnon, et al., 2001). In a separate *in-vitro* study Torzilli et al. (Torzilli et al., 1994) examined sagittal plane knee joint translations in response to individual and combined axial compressive loading and simulated quadriceps forces at fixed knee angles of 0°, 15°, 30°, 45°, and 90° flexion. At each knee flexion angle, anterior-posterior translations were measured in response to an anterior-posterior tibial force of 100 N after axial compressive loads of 0, 111, 222, 333, or 444 N, and a quadriceps forces of 0 or 133 N, were applied. Similar to the findings of Markolf et al., 2014), Torzilli et al., 1994) found that when axial compressive loads and quadriceps forces were applied separately or in combination, anterior tibial translation naturally occurred before any external anterior-posterior tibial forces were applied. Specifically, the application of a 133 N quadriceps force resulted in significant anterior tibial translation at 15°, 30°, and 45° of knee flexion, with the greatest amount of translation occurring at 30°; however, no translation occurred at 0° or 90° (Torzilli et al., 1994). When axial compressive loads were applied, anterior tibial translation increased with increasing compressive loads in a nearly linear fashion; and these translations were significant at 15°, 30°, 45°, and 90° of knee flexion, with the greatest amount of translation occurring at 45° (Torzilli et al., 1994). Finally, the combination of quadriceps force and compressive axial loading also increased anterior tibial translation, but this translation was only significant at lower compressive loads. The effects of isolated quadriceps loading on anterior tibial translation and ACL loading have

also been demonstrated in other *in-vitro* work, with reported increases in anterior tibial translation (Victor et al., 2010) and ACL strain (Fujiya et al., 2011; Li et al., 1999) between full extension and 30° to 60° of knee flexion.

From a mechanistic standpoint, it has been explained that anterior tibial translation occurs for two reasons. First, ground reaction forces induce a compressive axial load that acts through the posteriorly- and inferiorly-directed slope of the tibial plateau, which causes the axial compressive load to have an anteriorly directed force component at the proximal tibia (Torzilli et al., 1994). This proximal tibia anterior shear force is said to cause the femoral condyles to "slide" down the posteriorly- and inferiorly-directed slope of the tibial plateau, which then accelerates the tibia anteriorly and ultimately results in anterior tibial translation (McLean et al., 2011, 2010; Meyer & Haut, 2005; Schmitz et al., 2010; Torzilli et al., 1994). Although the absolute magnitude of these ground reaction forces does not directly represent the load experienced by the ACL, they are positively associated with proximal tibia anterior shear force (Sell et al., 2007; Yu et al., 2006), anterior tibial acceleration (McLean et al., 2011; McNair & Marshall, 1994) and anterior tibial translation (Schmitz et al., 2010; Torzilli et al., 1994), which directly contribute to ACL loading (K. L. Markolf et al., 1995; McLean et al., 2011; Shelburne, Pandy, Anderson, & Torry, 2004; Shelburne, Pandy, & Torry, 2004; Vahey & Draganich, 1991). Second, these ground reaction forces also produce an external knee flexion moment that must be counteracted by a quadriceps-generated internal knee extension moment to help stabilize the knee and control the downward acceleration of the body upon weight acceptance (J T Blackburn & Padua, 2008; Yu et al., 2006). When the knee is positioned in less than 65-70° of knee flexion, the quadriceps line of pull (line of action) is directed anteriorly, resulting in a force component directed perpendicular to the tibiofemoral joint surfaces (i.e. compressive force component) and a force component directed anteriorly (i.e. anterior shear component) (Draganich, Andriacchi, & Andersson, 1987). Although

the anterior shear component may be relatively small when compared to the compressive force component, depending on the knee flexion angle, it is reported to be large enough to produce additional anterior tibial translation and further load the ACL (B D Beynnon et al., 1995; DeMorat et al., 2004; Li et al., 1999; Withrow et al., 2006). For example, in the absence of any externally applied tibial force, the addition of a quadriceps load has been shown to significantly increase the force placed on the ACL at knee flexion angles less than 50° (Bruce D. Beynnon & Fleming, 1998; DeMorat et al., 2004; Fujiya et al., 2011; Li et al., 1999; Keith L Markolf, O'Neill, Jackson, & McAllister, 2004; Pandy & Shelburne, 1997). Additionally, if left unopposed, forceful contraction of the quadriceps with the knee relatively extended has been demonstrated to produce enough ACL strain to result in ACL rupture (DeMorat et al., 2004).

Movement of a limb segment typically involves some degree of agonist-antagonist cocontraction in order to help stabilize the joint (Draganich, Jaeger, & Kralj, 1989). The hamstring
muscles function antagonistically to the quadriceps and therefore function synergistically with the
ACL (Baratta et al., 1988; Solomonow et al., 1987). Cadaver research has demonstrated that the
hamstring muscles attachments on the posterior aspects of the proximal tibia and fibula
mechanically provide this muscle group with the ability to resist anterior and rotary tibiofemoral
motion (Victor et al., 2010). In this regard, several *in-vivo* (Baratta et al., 1988; B D Beynnon et
al., 1995; Solomonow et al., 1987), *in-vitro* (Draganich & Vahey, 1990; Li et al., 1999;
MacWilliams et al., 1999; Keith L Markolf et al., 2004; Victor et al., 2010; Withrow et al., 2006,
2008), and musculoskeletal modeling studies (Biscarini et al., 2014; Biscarini, Botti, &
Pettorossi, 2013; Imran & O'Connor, 1998; Kellis & Baltzopoulos, 1999; Pandy & Shelburne,
1997) have demonstrated that adequate co-contraction of the hamstring muscles can effectively
enhance knee joint stability by reducing anterior tibial translation and net proximal tibia anterior
shear forces, thereby reducing ACL loading. In general, such studies have demonstrated that

isolated quadriceps forces produce peak anterior tibial translation and ACL loading between 15° and 30° knee flexion, and that the addition of applied hamstring forces can effectively reduce anterior tibial translation, shear forces, and ACL loading at knee flexion angles greater than 10°-15° (Draganich & Vahey, 1990; Li et al., 1999; MacWilliams et al., 1999; Keith L Markolf et al., 2004; Pandy & Shelburne, 1997; Withrow et al., 2006, 2008). Mechanistically, the noted reduction in anterior tibial translation and anterior tibial shear force when hamstring cocontraction is applied has been described as follows. Similar to the quadriceps, contraction of the hamstring muscles results in two force components: one component directed perpendicular to the tibiofemoral joint surfaces (i.e. compressive force component) and one component directed posteriorly (i.e. posterior shear component) (Pandy & Shelburne, 1997). This compressive force component of the hamstrings acts to provide tibiofemoral compression, increasing the stability of the knee through increased joint stiffness (Baratta et al., 1988; Solomonow et al., 1987). At the same time, the posterior shear component produces a posteriorly-directed pull on the proximal tibia which reduces the net anterior shear force and thus ACL strain, thereby protecting the ACL (B D Beynnon et al., 1995; More et al., 1993; Solomonow et al., 1987). Although relatively small at more extended knee angles, this posterior shear component increases as the knee flexion angle increases due to the increased angle between the tendons of the hamstring muscles and the long axis of the tibia (Pandy & Shelburne, 1997).

Summary

The findings presented in this section collectively demonstrate that approximately 70% of all ACL injuries are noncontact in nature, and that such injuries are more likely to occur during competition than during training (B P Boden et al., 2000; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; McNair et al., 1990; Olsen et al., 2004). In addition, it is widely accepted that

noncontact ACL injuries typically occur during athletic movements that involve a sudden deceleration of the body, with or without a change in direction, such as when performing a sidecut or when landing on a single leg (B P Boden et al., 2000; Barry P Boden et al., 2009; Cochrane et al., 2007; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; Koga et al., 2010; Tron Krosshaug et al., 2007; Olsen et al., 2004). There is also overall agreement that such injuries occur within the first 10 to 50 milliseconds of initial foot contact with the ground (Koga et al., 2010; Tron Krosshaug et al., 2007) and that the knee is in a relatively extended position (i.e. < 30° flexion) at the time of injury (B P Boden et al., 2000; Barry P Boden et al., 2009; Cochrane et al., 2007; Faunø & Wulff Jakobsen, 2006; Ferretti et al., 1992; Koga et al., 2010; Tron Krosshaug et al., 2007; McNair et al., 1990; Olsen et al., 2004). In contrast, there has been less agreement between studies in terms of frontal and transverse plane knee motion at the time of injury, and this may be due to the noted limitations of retrospective interviews and two-dimensional video analyses as well as potential differences in knee joint motions between side-cutting maneuvers and jump landings. Additionally, given that all of the studies presented in this section report some type of secondary joint motion (internal-external tibial rotation, varus-valgus angulation), it is likely that such injuries do not occur in a single anatomical plane. However, it is well accepted that anterior tibial translation via proximal tibia anterior shear force is the most direct ACL loading mechanism (Butler et al., 1980; K L Markolf et al., 1990). Thus, any factors that are able to effectively protect the ACL from deleterious loading in the sagittal plane may be able to effectively reduce noncontact ACL injury risk. In this regard, adequate co-contraction of the hamstring muscles is thought to play a critical role in limiting the force experienced by the ACL. However, due to the inherent difficulties associated with measuring muscle forces and ACL loading in-vivo, the effect of hamstring co-contraction on ACL loading has been limited to cadaver studies, musculoskeletal modeling simulations, or during isometric knee-extension

exercises *in-vivo*. Therefore, the true extent to which the hamstrings are able to effectively limit ACL loading during functional athletic tasks commonly associated with noncontact ACL injury remains unknown.

Dynamic Knee Stability during Functional Athletic Tasks

Dynamic knee stability is defined as the ability of the knee joint to remain stable when subjected to rapidly applied loads (Williams, Chmielewski, Rudolph, Buchanan, & Snyder-Mackler, 2001), which is accomplished through a complementary relationship between passive (static) and active (dynamic) restraint mechanisms (Johansson & Sjolander, 1993; Lew, Lweis, & Craig, 1993). Passive restraint components include the bony geometry of the knee joint, ligaments, the joint capsule, cartilage, and friction (Johansson & Sjolander, 1993; Lew et al., 1993), whereas dynamic restraint components arise from neuromuscular control of the skeletal muscle(s) that cross the joint (Riemann & Lephart, 2002). Specifically, neuromuscular control refers to the activation of dynamic restraints (the muscles) that occurs in preparation for (i.e. preparatory muscle activation), and in response to (i.e. reflexive and reactive muscle activation), joint motion and loading for the purpose of maintaining and or restoring dynamic knee stability (Myers & Lephart, 2000). The dynamic restraint system relies on both feed-forward (preparatory) and feed-back (reflexive and reactive) motor control strategies in order to anticipate or react to joint motion and loading (Riemann & Lephart, 2002). Together, these feed-forward and feedback motor control strategies govern the instantaneous and continuously changing levels of dynamic restraint in order to protect the capsuloligamentous structures (passive restraints) from deleterious loading and maintain dynamic knee stability (Swanik, Lephart, Giannantonio, & Fu, 1997). Feed-forward control is responsible for planning and/or preprogramming muscle(s) activation levels in order to act as a "stress shield" for the articular structures in anticipation of

joint loading, and is based on learned experiences from the past. Conversely, feed-back control regulates motor control through a number of reflexive pathways that continuously modify muscle activity in order to accommodate unanticipated events (Swanik et al., 1997).

As previously discussed, noncontact ACL injuries are reported to occur within 50 milliseconds of initial ground contact during cutting and landing maneuvers (Koga et al., 2010; Tron Krosshaug et al., 2007). Given that feed-back motor control strategies are shown to have a latency of approximately 100 milliseconds (Dyhre-Poulsen & Krogsgaard, 2000), it is likely that such injuries occur too rapidly for feed-back control strategies to effectively stabilize the knee joint and protect the ACL (T E Hewett et al., 2009; Hurd, Chmielewski, & Snyder-Mackler, 2006). Fortunately, however, it has been demonstrated that athletes are able to adopt preparatory neuromuscular control strategies in anticipation of knee joint loading (Cowling, Steele, & McNair, 2003), and this preparatory muscle activation has been demonstrated to increase overall joint stiffness and enhance dynamic knee stability (Bryant et al., 2008; McNair & Marshall, 1994; Swanik et al., 2004). As such, any imbalance or delay in preparatory neuromuscular activation can lead to improper limb positioning at initial ground contact and place high loads on the passive joint restraints, potentially increasing noncontact ACL injury risk. To this end, female athletes are at a substantially greater risk of experiencing noncontact ACL injuries compared to similarly trained males (Agel, Arendt, & Bershadsky, 2005; El A Arendt, Agel, & Dick, 1999; T E Hewett, Lindenfeld, Riccobene, & Noyes, 1999), and it is thought that this increased injury-risk in females may be due to between-sex differences in neuromuscular control strategies that place these individuals in positions that are associated with increased ACL loading during the time at which ACL injuries are reported to occur. Therefore, a number of controlled laboratory studies have examined between-sex differences in neuromuscular and biomechanical characteristics during a variety of movements, such as side-cutting and cross-over cutting, straight running with

quick decelerations, double- and single-leg drop vertical jumps and drop landings, and double- and single-leg stop-jump landing tasks, in order to better understand the factors that potentially explain the higher incidence of noncontact ACL injuries in female populations. The remainder of this section aims to highlight what is currently known about the influence of neuromuscular and biomechanical characteristics on biomechanical factors indicative of ACL loading during functional athletic tasks, the effects of landing type (i.e. single- versus double-leg landing tasks) on these characteristics, and between-sex differences therein.

Influence of Neuromuscular and Biomechanical Characteristics on ACL Loading

From a purely mechanistic standpoint, ACL injury occurs when the stress placed on the ligament exceeds its failure strength (Slauterbeck, Hickox, Beynnon, & Hardy, 2006). Although the absolute magnitude of stress/strain experienced by the ACL is difficult to measure *in-vivo* during functional athletic movements, previous studies have demonstrated that proximal tibia anterior shear force represents the most direct ACL loading mechanism (Butler et al., 1980; K L Markolf et al., 1990). Therefore proximal tibia anterior shear force is often used as an indicator of ACL loading in controlled laboratory experiments because it can be estimated via inverse dynamics (Sell et al., 2007). It is important to note however, that proximal tibia anterior shear force, as calculated through inverse dynamics, is a resultant force vector that includes contributions from all of the passive and active restraint mechanisms acting at the knee joint and does not directly represent the shear forces transmitted to the ACL.

There are several commonly measured neuromuscular and biomechanical factors that have been reported to be predictive of proximal tibia anterior shear force, such as posterior and vertical ground reaction forces, trunk, hip, and knee joint kinematics, joint resultant moments, and activation of the musculature surrounding the knee joint. Specifically, Yu et al. (Yu et al., 2006)

first examined relationships between select lower-extremity kinematic and kinetic variables during double-leg stop-jump landings in recreationally active men and women, and found that slower hip and knee flexion angular velocities at initial ground contact, smaller knee flexion angles at the instant of peak proximal tibia anterior shear force, and smaller peak knee flexion angles during landing, were all individually predictive of greater peak posterior and vertical ground reaction forces. Yu et al (Yu et al., 2006) also found that greater peak posterior and vertical ground reaction forces were positively associated with greater proximal tibia anterior shear forces and peak knee extensor moments, and that greater peak knee extensor moments were highly correlated with greater proximal tibia anterior shear force during landing. Sell et al (Sell et al., 2007) later expanded on the findings of Yu et al., 2006) by attempting to determine whether a select combination of neuromuscular and biomechanical characteristics could significantly predict peak proximal tibia anterior shear force during a double-leg stop-jump task. In this study, it was demonstrated that the linear combination of peak posterior ground reaction force, knee extensor moment and knee flexion angle at the instant of peak posterior ground reaction force, preparatory muscle activation of the quadriceps (vastus lateralis), and sex, was able to predict 86.1% of the variance in peak proximal tibia anterior shear force, with greater posterior ground reaction forces, knee extensor moments, and quadriceps activation, being female, and smaller knee flexion angles predicting greater proximal tibia anterior shear (Sell et al., 2007). Similar findings have also been reported during double-leg drop-jump landings, where it was found that the linear combination of sex, hip and knee flexion excursion, knee extensor moment, quadriceps and hamstring peak torque, and pre- and post-activation of the quadriceps and hamstring muscles, explained 56.5% of the variance in peak proximal tibia anterior shear force (Shultz, Nguyen, Leonard, & Schmitz, 2009). In general, Shultz et al (Shultz et al., 2009) found that, independent of sex, individuals who displayed less hip flexion excursion, greater knee flexion excursion, greater knee extensor moments, and greater quadriceps muscle activation at landing, experienced greater proximal tibia anterior shear force during landing. Furthermore, Gheidi et al (Gheidi, Sadeghi, Moghadam, Tabatabaei, & Kernozek, 2014) recently examined kinematic and kinetic predictors of proximal tibia anterior shear force during single-leg drop landings in elite female basketball and volleyball players, and it was reported that the combination of greater peak knee extensor moments and smaller peak knee flexion angles explained 30.6% of the variance in peak proximal tibia anterior shear force.

Effect of Landing-Type on Neuromuscular and Biomechanical Characteristics

It is consistently reported in the literature that noncontact ACL injuries most often occur when cutting or landing on a single leg (B P Boden et al., 2000; Barry P Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004). It has also been reported that large between-limb asymmetries in weight-distribution are present when such injuries occur during double-leg landings, and thus may actually be more representative of single-leg landings (T E Hewett et al., 2009; Olsen et al., 2004). Surprisingly, however, double-leg drop-jump and drop-landing tasks are predominantly used as a model to investigate noncontact ACL injury risk, and few studies have attempted to objectively quantify differences in neuromuscular and biomechanical characteristics between double-leg and single-leg landing tasks.

Pappas et al. (Pappas et al., 2007) investigated differences in kinematic, kinetic, and neuromuscular characteristics elicited by double-leg and single-leg drop landings in healthy college-aged men and women and found that almost all variables examined were affected by the type of landing performed (i.e. single-leg vs double-leg). Specifically, compared to double-leg drop landings, single-leg landings elicited significantly less knee flexion at initial ground contact, less peak knee flexion, greater hip adduction and knee valgus, and greater neuromuscular

activation of the quadriceps, hamstrings, and gastrocnemius musculature, whereas peak vertical ground reaction force was not statistically different (Pappas et al., 2007). In a separate study, Yeow et al (Yeow et al., 2010) examined differences in knee joint kinematics and energetics between two different drop-landing heights (0.3 meters and 0.6 meters) and between double- and single-leg landings. Compared to double-leg landings, single-leg landings were reported to elicit greater peak vertical ground reaction forces at both landing heights, smaller knee flexion angles and less knee flexion angular velocities at both landing heights, and less joint power and eccentric work at both landing heights; altogether, such findings were suggested to indicate that individuals were able to respond more effectively to larger impact forces in terms of knee joint kinematics and energetics during double-leg landings, which allowed for better shock absorption and thus may indicate a reduced risk of sustaining injury compared to single-leg landings (Yeow et al., 2010). Similar differences have also been observed for double- and single-leg stop-jump landing tasks. Specifically, Wang et al (Wang, 2011) examined differences in lower-extremity kinematics and kinetics, and ground reaction forces, in elite male volleyball players and demonstrated that the single-leg stop-jump elicited significantly smaller hip and knee flexion angles and angular velocities at initial ground contact, smaller peak hip and knee flexion angles during landing, greater peak posterior and vertical ground reaction forces, greater peak knee extensor and knee valgus moments, and greater peak proximal tibia anterior shear forces, compared to the doubleleg stop-jump task (Wang, 2011).

Between-Sex Differences in Neuromuscular and Biomechanical Characteristics

There have been several investigations on between-sex differences in neuromuscular and biomechanical characteristics during both double-leg and single-leg landing tasks and during side-cut maneuvers. Malinzak et al. (Malinzak et al., 2001) demonstrated that female recreational

athletes displayed smaller knee flexion angles, greater knee valgus angles, increased quadriceps activation, and decreased hamstring activation during the stance phase of running and cutting tasks compared to their male counterparts. Although Sigward and Powers (Sigward & Powers, 2006) also found that females displayed greater quadriceps activation than males during the stance phase of a side-cut maneuver, males and females were reported to display no differences in hamstring activation. Two other studies have also demonstrated that females display greater preparatory quadriceps muscle activation during both unanticipated (Landry, McKean, Hubley-Kozey, Stanish, & Deluzio, 2009) and anticipated (Zazulak et al., 2005) side-cut maneuvers, but that hamstring activation was similar between sex (Landry et al., 2009). When examining between-sex differences during double-leg drop landing tasks, it has been reported by Shultz et al (Shultz et al., 2009) that females display greater hip and knee flexion excursions, greater peak knee extensor moments, and greater quadriceps and hamstring activation, both before and after landing, compared to males. However, no between-sex differences were observed in hip and knee flexion angles at initial ground contact or in peak proximal tibia anterior shear force (Shultz et al., 2009). The finding that males and females display similar hip and knee flexion angles at initial ground contact has also been observed during single-leg drop landings (Schmitz et al., 2007). However, Schmitz et al. (Schmitz et al., 2007) also found that females displayed significantly less hip and knee angular excursions than males, which directly contradicts the findings of Shultz et al (Shultz et al., 2009), who reported greater hip and knee flexion excursions in females during double-leg drop landings. Schmitz et al. (Schmitz et al., 2007) further demonstrated that females performed single-leg drop landings with significantly slower knee and hip flexion angular velocities and greater peak vertical ground reaction forces than males. Females have also been reported to display significantly greater peak knee valgus angles and vertical ground reaction

forces compared to males during both double-leg and single-leg drop-landing tasks (Pappas et al., 2007).

When reviewing the literature that has specifically examined between-sex differences in neuromuscular and biomechanical characteristics during stop-jump landing tasks, there appears to be much greater consistency among studies. For example, Chappell et al (Jonathan D Chappell et al., 2002) examined between-sex differences during double-leg stop-jump landings and found that females displayed significantly greater peak proximal tibia anterior shear forces, greater knee extensor moments, and greater knee valgus moments, compared to males. Similarly, Yu et al (Yu et al., 2006) demonstrated that females performed double-leg stop-jump landings with smaller hip and knee flexion angles and a slower hip flexion angular velocity at initial ground contact, smaller knee flexion angles at the instant of peak proximal tibia anterior shear force, less peak knee flexion, and greater peak posterior and vertical ground reaction forces and peak knee extensor moments, compared to males. Given that males and females display significantly different landing patterns, it was later hypothesized by Chappell et al (J. D. Chappell, Creighton, Giuliani, Yu, & Garrett, 2007) that such between-sex differences could be due to differences in the strategies that males and females employ in preparation for landing. As such, Chappell et al (J. D. Chappell et al., 2007) conducted a study aimed to identify differences in movement patterns during the pre-landing phase of a double-leg stop-jump landing that might affect ACL loading parameters following initial ground contact. Although males and females displayed similar kinematics at the start of the task, females were found to land with less hip and knee flexion, more internal tibial and hip rotation, and more hip abduction compared to males (J. D. Chappell et al., 2007). In terms of neuromuscular activation, males and females were found to display similar quadriceps activation patterns, with a distinct increase in activation approximately 50 milliseconds prior to landing; however, the magnitude of quadriceps activation was found to be

significantly greater in females. Neuromuscular activation of the hamstring muscles was also demonstrated to gradually increase prior to landing in both sexes; however, the magnitude of hamstring activation was found to be significantly greater in females (J. D. Chappell et al., 2007).

Summary

Taken together, the findings presented in this section clearly demonstrate that men and women employ different neuromuscular control strategies when performing functional athletic movements. Specifically, previous studies demonstrate that females display greater neuromuscular activation of the quadriceps both prior to and during landing, which may or may not always be accompanied by greater hamstring activation (J. D. Chappell et al., 2007; Landry, McKean, Hubley-Kozey, Stanish, & Deluzio, 2007; Landry et al., 2009; Malinzak et al., 2001; Shultz et al., 2009; Sigward & Powers, 2006; Zazulak et al., 2005). Females are also generally reported to display smaller knee (J. D. Chappell et al., 2007; Malinzak et al., 2001; Yu et al., 2006) and hip flexion angles (Yu et al., 2006), smaller hip and knee angular excursions and slower angular velocities (Schmitz et al., 2007; Yu et al., 2006), greater knee extensor moments (Jonathan D Chappell et al., 2002; Shultz et al., 2009; Yu et al., 2006), greater knee valgus angles (Pappas et al., 2007) and valgus moments (Jonathan D Chappell et al., 2002), greater peak posterior (Yu et al., 2006) and vertical ground reaction forces (Pappas et al., 2007; Schmitz et al., 2007; Yu et al., 2006), and greater proximal tibia anterior shear force (Jonathan D Chappell et al., 2002; Yu et al., 2006), compared to similarly trained males. These between-sex differences in neuromuscular and biomechanical characteristics during functional athletic movements have collectively been considered to help explain, at least in part, the reason that females are at increased risk of experiencing noncontact ACL injury compared to males (Griffin et al., 2006; Shimokochi & Shultz, 2008).

Although the findings presented in this section clearly demonstrate that males and females perform functional athletic movements differently, this section also provides sufficient evidence to suggest that single-leg tasks elicit dramatically different neuromuscular and biomechanical responses. Specifically, single-leg tasks tend to elicit smaller hip and knee flexion angles (Pappas et al., 2007; Wang, 2011; Yeow et al., 2010), slower hip and knee flexion angular velocities (Wang, 2011; Yeow et al., 2010), greater knee extensor moments (Wang, 2011), greater peak posterior (Wang, 2011) and vertical ground reaction forces (Pappas et al., 2007; Wang, 2011; Yeow et al., 2010), greater knee valgus angles (Pappas et al., 2007) and valgus moments (Wang, 2011), and greater peak proximal tibia anterior shear (Wang, 2011), compared to double-leg tasks. Additionally, single-leg landings have been reported to elicit increased neuromuscular activation of the quadriceps, hamstring, and gastrocnemius musculature compared to double-leg landings (Pappas et al., 2007). These findings, along with the fact that noncontact ACL injuries most often occur during single-leg cutting and jump landing tasks (B P Boden et al., 2000; Barry P Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), suggests that using a double-leg landing task as a model to study ACL loading and injury risk may not adequately represent the situations in which such injuries actually occur. In this regard, noncontact ACL injuries are reported to involve a horizontal deceleration component (B P Boden et al., 2000; Barry P Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), which is absent during the drop jump and drop landings tasks commonly used in controlled laboratory studies. In contrast, stopjump landing tasks do involve a horizontal deceleration component and thus may be more appropriate, especially when acknowledging that posterior ground reaction force has been reported to be predictive of proximal tibia anterior shear force (Sell et al., 2007; Yu et al., 2006).

Finally, this review has only identified four studies that have attempted to determine the neuromuscular and biomechanical characteristics that influence proximal tibia anterior shear

force. The results of these studies are difficult to compare due to differences in sample characteristics and differences in the task employed. For example, Yu et al (Yu et al., 2006) and Sell et al (Sell et al., 2007) examined double-leg stop-jumps in a mixed sample of males and females. Conversely, Shultz et al (Shultz et al., 2009) examined a double-leg drop landing in a mixed sample of males and females whereas Gheidi et al (Gheidi et al., 2014) examined single-leg drop landings in females only. Additionally, the findings of both Yu et al (Yu et al., 2006) and Sell et al (Sell et al., 2007) indicate that peak posterior ground reaction force is positively associated with peak proximal tibia anterior shear force. However, as mentioned previously, double-leg and single-leg drop landing tasks do not produce posterior ground reaction forces of the same magnitude (if at all) as stop-jump tasks, which limits comparison between studies. Furthermore, it remains unknown whether the factors that are predictive of peak proximal tibial anterior shear forces during double-leg stop-jump tasks are similar for single-leg stop-jump tasks. Future studies examining the influence of such factors on proximal tibia anterior shear are warranted, and sex-stratified statistical models are encouraged.

Hamstring Musculo-Articular Stiffness

A property of the hamstring musculature that may potentially impact ACL loading and noncontact ACL injury risk is called musculo-articular stiffness. Stiffness (K) describes the relationship between an applied load and the amount of elastic deformation that occurs within a given structure, and is mechanically defined as the ratio of change in force to change in muscle length (Ditroilo, Watsford, Murphy, & De Vito, 2011). The rationale behind the idea that hamstring musculo-articular stiffness (K_{HAM}) potentially impacts ACL loading and injury risk is based on the following: First, anterior tibial translation naturally occurs as the knee initially transitions from non-weight bearing to weight bearing, and this motion has been shown to load

the ACL (Torzilli et al., 1994); Second, numerous cadaveric and musculoskeletal modeling studies have demonstrated that simulated hamstring forces reduce anterior tibial translation (Li et al., 1999), ACL strain (Withrow et al., 2008), and ACL loading (Li et al., 1999; Keith L Markolf et al., 2004), and similar effects of hamstring contraction on ACL strain have also been demonstrated *in-vivo* (B D Beynnon et al., 1995); Third, anterior tibial translation is thought to produce a tensile force on the hamstring muscles as well as the secondary ligamentous and capsular restraints (McNair et al., 1992). Therefore, given the mechanical definition of stiffness, it is theorized that for a given proximal tibia anterior shear force, relatively stiffer hamstrings will permit a smaller change in length compared to more compliant (i.e., less stiff) hamstrings, thus limiting anterior tibial translation and ACL loading, potentially reducing noncontact ACL injury risk. The purpose of this section is to present and summarize the current body of knowledge on *K*_{HAM}.

Measurement of Hamstring Musculo-Articular Stiffness

Stiffness (K) can be described from the macroscopic level of the whole body all the way down to the microscopic level of a single muscle fiber, and can be assessed under both passive and active conditions. Although a number of *in-vitro* and *in-vivo* stiffness measures exist, the following sections focus strictly on the assessment of hamstring musculo-articular stiffness. Hamstring musculo-articular stiffness (K_{HAM}) is an *in-vivo* measure of stiffness, which is assessed with the hamstring musculature actively contracted via the free-oscillation technique. The K_{HAM} value obtained from the free-oscillation technique represents a global measure of stiffness, which includes contributions from the muscle-tendon unit, skin, ligaments, and articular joint capsule (Ditroilo, Watsford, Murphy, et al., 2011). Previous studies using the free-oscillation technique have referred to this outcome measure of stiffness using various terms, such as 'stiffness of the

series elastic component' (G J Wilson, Wood, & Elliott, 1991), 'muscle tendon stiffness' or 'musculotendinous stiffness' (Greg J Wilson et al., 1994), 'muscle stiffness' (McNair et al., 1992), 'active stiffness' (J. Troy Blackburn, Padua, Riemann, & Guskiewicz, 2004; J.Troy Blackburn et al., 2004; K. P. Granata, Wilson, Massimini, & Gabriel, 2004), 'effective stiffness' (Kevin P. Granata, Wilson, & Padua, 2002), 'muscle viscoelasticity' (Fukashiro, Noda, & Shibayama, 2001), or 'structural stiffness' (J. Troy Blackburn, Padua, Weinhold, & Guskiewicz, 2006). Even though the muscle-tendon unit is shown to be the primary contributor towards the global stiffness value obtained under active conditions (J. Troy Blackburn, Padua, & Guskiewicz, 2008; J. Troy Blackburn et al., 2006; K. P. Granata et al., 2004; Kevin P. Granata et al., 2002), it has been suggested that future studies adopt the term 'musculo-articular stiffness' when the free-oscillation technique is used because it is thought to better represent the comprehensive nature of the measure (Ditroilo, Watsford, Murphy, et al., 2011).

The free-oscillation technique is based on the frequency response of a perturbed system, and relies on modeling the system under consideration as a damped harmonic oscillator, which consists of a spring, a mass, and a viscous damping force; when the system is acted upon by an external force, it then begins to oscillate (Symon, 1971). This technique was first introduced for use in the human body by Cavagna (Cavagna, 1970) in an attempt to estimate the amount of elastic energy stored in contracted human musculature, and the general assessment procedures and experimental set-up were later modified by McNair et al (McNair et al., 1992) to specifically assess the hamstring musculature. When using the free-oscillation technique to measure K_{HAM} , the lower-extremity is modeled as a single-degree-of-freedom mass-spring system, with a damping element, and the problem of distinguishing between individual muscles is typically avoided by assuming that a single equivalent muscle acts to flex or extend the knee (Shorten, 1987). Figure 2.2 displays the single-degree-of-freedom mass-spring model originally presented by McNair et

al (McNair et al., 1992), where the hamstring muscle-tendon unit(s) is represented as a massless linear spring (with a damping element), and the lower leg (i.e. shank and foot segment) and externally applied load are represented as the inertial mass.

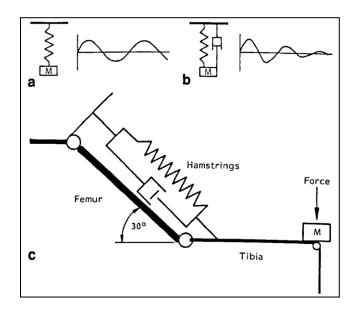


Figure 2.2. Models of a Single-Degree-of-Freedom Mass-Spring System: (A) Undamped; (B) with a Viscous Damping Component; and (C) as the Hamstrings Were Modeled by McNair et al. To the right of A and B are representative oscillations associated with the respective models when they are perturbed from their equilibrium position.

Because the muscle-tendon unit has been shown to exhibit both elastic and viscous properties, such as stress, relaxation, creep, hysteresis, and strain-rate dependence, this method of modeling the leg as a single-degree-of-freedom mass-spring system with a viscous damping element has been granted construct validity (McHugh, Magnusson, Gleim, & Nicholas, 1992; Taylor, Dalton, Seaber, & Garrett, 1990). When a perturbation is applied to the loaded system, the system begins to oscillate, and these oscillations are then rapidly dampened due to the viscoelastic properties of the muscle-tendon complex (Shorten, 1987). This damped oscillatory motion is captured via an accelerometer and is later processed using a second-order linear

equation, which considers the frequency of oscillation and the damping coefficient. Figure 2.3 depicts a representative time-series of the accelerometer data typically recorded during such procedures, where the time and acceleration interval between the first and second oscillatory peaks is then used to calculate the damped frequency and coefficient of damping, respectively (McNair et al., 1992). With this information, and knowledge of the applied load, K_{HAM} can then be calculated using the equation:

$$K_{HAM} = 4\pi^2 m f^2 + c^2/4m \qquad (Equation 1)$$

where K_{HAM} is the stiffness of the hamstrings (N·m⁻¹), m is the total system mass [mass of shank and foot segment + applied load (kg)], f is the damped frequency of oscillation, and c is the coefficient of damping. The coefficient of damping (c) is calculated from a knowledge of the natural frequency of oscillation (ω_n), the damping factor (ζ), and the total system mass (m) (McNair et al., 1992):

$$c = 2m\zeta\omega_n$$
 (Equation 2)

First, the amount of damping must be obtained from the change in oscillation amplitudes during one complete cycle, which has been expressed as:

$$\delta = \ln \frac{x_1}{x_2} \tag{Equation 3}$$

where δ is the logarithmic decrement. With knowledge of δ , the damping factor (ζ) may then be calculated:

$$\zeta = \frac{\delta}{\sqrt{(2\pi)^2 + \delta^2}}$$
 (Equation 4)

Finally, the natural frequency of oscillation (ω_n) is:

$$\omega_n = \frac{\omega_d}{\sqrt{1 - \zeta^2}}$$
 (Equation 5)

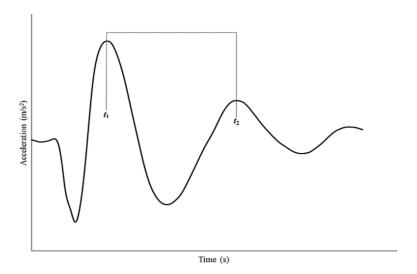


Figure 2.3. Example Accelerometer Time-Series Data Obtained During the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. t_1 and t_2 represent the time points at which the first two oscillatory peaks occur; these time points are then used to calculate the damped frequency of oscillation.

Although several studies have continued to use (Equation 1 to calculate K_{HAM} (Ditroilo, Watsford, Murphy, et al., 2011; Kevin P. Granata et al., 2002; Swanik et al., 2004; Watsford et al., 2010), others have opted to eliminate the damping coefficient from their calculations since the contribution of damping coefficient to the overall K_{HAM} value has been demonstrated to be less than 5% in one study (Jennings & Seedhom, 1998) and less than 1% in another (J.Troy Blackburn et al., 2004). Therefore, K_{HAM} has been more commonly calculated using the following equation, which has been modified to exclude the damping coefficient:

$$K_{HAM} = 4\pi^2 m f^2$$
 (Equation 6)

It should be pointed out that the assumption of linearity has been called into question. Specifically, Coveney and colleagues (Coveney, Hunter, & Spriggs, 2001) examined the time periods between the first and second oscillatory cycles (i.e. the time from the first oscillatory peak to the second oscillatory peak vs. the time from the second oscillatory peak to the third oscillatory peak) and found that the time period from the first to second oscillatory cycle had decreased, which ultimately caused an increase in stiffness due to an increase in the frequency of oscillation. Thus, it was concluded that the stiffness data obtained via free-oscillation exhibit nonlinear characteristics (Coveney et al., 2001). Despite these findings however, most studies that have adopted the free-oscillation technique have used the linear model to describe the damped oscillations because it is easier to use, and their stiffness calculations have been based solely on the first cycle of oscillations (Ditroilo, Watsford, & De Vito, 2011). Although the use of this linear model has been granted construct validity (Ditroilo, Watsford, & De Vito, 2011), nonlinear behavior has only been explored for the musculo-articular stiffness of the ankle plantar-flexor musculature, and we are unaware of any investigations that have studied this behavior for K_{HAM} . Based on such findings, it appears that use of the linear model is appropriate when investigators are solely interested in the initial response of the system following a perturbation. However, given the current evidence suggesting the potential for nonlinearity in the system, future studies interested in more than the initial response of the system are encouraged to explore the use of a nonlinear model instead. Additional studies on this topic appear warranted.

Experimental Apparatus and Procedures. Hamstring musculo-articular stiffness (K_{HAM}) is assessed with the participant positioned prone, with the trunk and thigh supported and the shank and foot segments free to move (McNair et al., 1992). A load is then attached proximal to the ankle joint, using either a DeLorme-type boot or cuff-style ankle weights, and care taken to ensure that the ankle is fixed in a neutral position. An accelerometer is then secured to the area of

the calcaneus, with the recording axis of the accelerometer aligned perpendicular to the lower leg. Once this experimental setup has been completed, investigator then passively positions the participant's lower leg until the knee is placed at the desired flexion angle, and the participant is required to hold the weight of their shank and foot segment, and the applied load, in this position via isometric hamstring contraction (Figure 3). Shortly following contraction of the hamstring musculature, a brief downward manual perturbation is applied to initiate oscillatory extension-flexion at the knee joint, and the ensuing damped oscillations are recorded via the accelerometer (Figure 2.4) (McNair et al., 1992). Although all of the studies that have investigated $K_{\rm HAM}$ share similarities in their experimental apparatus and procedures, such as adopting a prone assessment position with the hip and knee in some degree of flexion, there are a number of differences that exist between studies in terms of: 1) the degree to which the hip and knee are flexed prior to the perturbation, 2) the way in which the applied load is determined, 3) the way in which the perturbation is administered and the magnitude of the perturbation, 4) instructions provided to the participants, and 5) the use of surface electromyography (sEMG). Such differences are discussed throughout the remainder of this section.

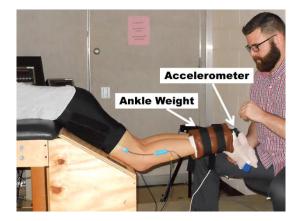


Figure 2.4. Instrumentation and Participant Positioning for the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment.

Participant Positioning. It is consistently reported in the literature that K_{HAM} is assessed with the trunk and thigh supported in 30° of hip flexion (J Troy Blackburn, Bell, Norcross, Hudson, & Engstrom, 2009; J. Troy Blackburn et al., 2013, 2011, 2004; J. Troy Blackburn & Norcross, 2014; J. Troy Blackburn & Pamukoff, 2014; Jennings & Seedhom, 1998; McNair et al., 1992; Waxman, Schmitz, & Shultz, 2015), with only one study reporting an alternative hip flexion angle of 45° (Kevin P. Granata et al., 2002). However, greater variability exists with regard to knee positioning. Most often, K_{HAM} is assessed with the lower leg positioned parallel to the ground and perpendicular to the effects of gravity, placing the knee at a relative flexion of 30° (D R Bell et al., 2012; J Troy Blackburn et al., 2009; J. Troy Blackburn et al., 2009, 2013, 2011, 2004; J. Troy Blackburn & Pamukoff, 2014; J.Troy Blackburn et al., 2004; Jennings & Seedhom, 1998; McNair et al., 1992; Swanik et al., 2004; Waxman et al., 2015). In contrast, other studies have assessed $K_{\rm HAM}$ with the knee positioned in 45° (Kevin P. Granata et al., 2002) or 80° of knee flexion (Ditroilo, Watsford, & De Vito, 2011; Ditroilo, Watsford, Murphy, & De Vito, 2013; Watsford et al., 2010). To this end, only two studies have provided a general rationale for the chosen assessment position. For 30° of hip and knee flexion, McNair et al (McNair et al., 1992) stated that this position was chosen because it closely mimics the angular position of the hip and knee joints at the time of initial ground contact during gait, which is when episodes of the knee "giving way" generally occur. Alternatively, Watsford et al. (Watsford et al., 2010) assessed $K_{\rm HAM}$ with the hip and knee in 30° and 80° of flexion, respectively, and stated that this position was chosen because it is representative of the latter part of the swing phase of gait during running, which is when the hamstring musculature is placed under high eccentric tension and hamstring injuries are thought to occur. Thus, it appears that the research question at hand (e.g. hamstring vs. knee injury/function) may influence the chosen hip and knee flexion angles for the $K_{\rm HAM}$ assessment.

Assessing K_{HAM} in a prone position, with the hip and knee in slight flexion, is reported to be the most ecologically valid method for two reasons. First, adopting a prone position allows researchers to approximate the functional length-tension relationship of the hamstring musculature during sprinting where, at initial ground contact, the hip and knee are in roughly 30-45° of flexion (Mann & Sprague, 1980; Stanton & Purdham, 1989). Second, during gait activities, the hamstrings are contracting concentrically at the knee joint and eccentrically at the hip joint, which can be simulated in the prone testing position (Worrell, Denegar, Armstrong, & Perrin, 1990). Because all of the studies included in this review have used a hip flexion angle of 30-45°, it appears that this may allow room for comparison. However, the wider range of knee-flexion angles (30° to 80°) used throughout the literature creates some concern due to length-tension relationships in human skeletal muscle. The moment arms of the hamstrings are reported to be greatest at 45° of knee flexion (Smidt, 1973); however, others have demonstrated that, when positioned prone with the hip in 30° of flexion, the hamstrings produce peak torque at approximately 30° of knee flexion (Barr & Duncan, 1988). Similar findings have been reported with the hip in a neutral position, where the hamstrings were found to produce greater torque at 30° of knee flexion, compared to 60°, and that torque production continued to decrease as the knee was flexed to 90° (Worrell et al., 2001). In addition, a wide range of variability in neuromuscular activation (i.e. normalized surface electromyography amplitude) has been observed across knee joint angles between full extension and 90° of flexion, and this variability in neuromuscular activation appears to be a key factor affecting torque-angle relationships (Worrell et al., 2001). Taken together, these findings provide support for assessing the hamstring musculature in a prone position, with the hip and knee slightly flexed, but also indicates that knee flexion angle does have an effect on both neuromuscular activation and torque production.

Therefore, caution should be taken when attempting to compare findings between studies that have assessed K_{HAM} using different hip and knee flexion angles.

Stiffness-Load Relationship and Assignment of Applied Load. The final equation used to calculate K_{HAM} ($K_{\text{HAM}} = 4\pi^2 m f^2$) implies that if the frequency of oscillation (f) were to remain constant, changing the total system mass (m; i.e. mass of the shank and foot segment + applied load) by increasing or decreasing the applied load would result in a corresponding increase or decrease in the calculated K_{HAM} value. However, there has been some debate over the linearity of this stiffness-load relationship, and the findings of such work are presented throughout the remainder of this section.

Assessing musculo-articular stiffness using a range of applied loads allows one to determine the relationship between stiffness and the applied moment (Shorten, 1987). This relationship was first examined in the ankle extensor muscles (plantar-flexors) by assessing stiffness under eight different loads (Shorten, 1987). It was reported that stiffness increased in a curvilinear fashion as the applied load increased, with the slope of the relationship being steep at low loads and then beginning to plateau at higher loads (Shorten, 1987). This relationship has been explained by experiments on the isolated muscle-tendon unit, where individual contributions of the series elastic components, parallel elastic components, and contractile components, to the overall stiffness value obtained, were reported to be dependent on the assessment load applied (Morgan, 1977). Within the single-degree-of-freedom mass-spring model, series elastic and parallel elastic components are represented as being constant (with the parallel elastic component being negligible), and the stiffness of the contractile component is thought to be proportional to the applied load (Morgan, 1977). In support of this theory, the stiffness of the series elastic component is reported to be the primary contributor to the overall musculo-articular stiffness value at low loads; and as muscle activation increases due to an increasing load, stiffness of the

contractile component increases linearly until the applied load becomes equal to that of maximal isometric tension, at which point the stiffness of the contractile component becomes similar to that of the series elastic component (Ditroilo, Watsford, & De Vito, 2011; McNair & Stanley, 1996; McNair et al., 1992; Shorten, 1987). This increase in stiffness of the contractile component is thought to be predominantly due to the activation of more muscle fibers (i.e., increased crossbridge formation). Reports of this curvilinear stiffness-load relationship have been noted for a variety of musculature, and for a more in-depth review of such findings, the reader is referred to Ditroilo et al (Ditroilo, Watsford, Murphy, et al., 2011).

In contrast to reports of a curvilinear stiffness-load relationship, two investigations on $K_{\rm HAM}$ have reported the stiffness-load relationship to be linear. Jennings and Seedhom (Jennings & Seedhom, 1998) assessed K_{HAM} using four assessment loads (15-25%, 30%, 45%, and 60% MVIC torque) and found the stiffness-load relationship to be linear. The authors stated that although their findings contradicted the nonlinear relationship demonstrated by McNair et al (McNair et al., 1992), who used only three assessment loads, the inclusion of an additional assessment load allowed a 33% greater confidence in the accuracy of the best fit line (Jennings & Seedhom, 1998). Granata et al (K. P. Granata, Padua, & Wilson, 2002) also reported a linear relationship when assessing K_{HAM} using multiple applied loads (0 kg, 6 kg, and 20% of MVIC torque). It has been demonstrated that intrinsic stiffness, arising purely from the mechanical properties of the structures involved, increases linearly with the applied load and thus neuromuscular effort, whereas reflexive stiffness, arising from a change in neuromuscular activation resulting from a reflexive response, is maximal at low loads and then decreases as the applied load in increased (Mirbagheri, Barbeau, & Kearney, 2000; Sinkjaer, Toft, Andreassen, & Hornemann, 1988). Therefore, it seems intuitive that total stiffness (the sum of intrinsic and reflexive stiffness) would fit a second-order polynomial curve (Sinkjaer et al., 1988). However, it has been argued that because K_{HAM} is assessed within the mid-range of neuromuscular activation, this assessment is able to capture purely intrinsic stiffness without any reflexive stiffness contribution, and this would explain the linear stiffness-load relationship observed (Jennings & Seedhom, 1998).

In the published literature, $K_{\rm HAM}$ has generally been assessed using multiple (3 to 5) loads ranging from 20% to 60% MVIC torque (Ditroilo, Watsford, & De Vito, 2011; Ditroilo et al., 2013; Kevin P. Granata et al., 2002; Jennings & Seedhom, 1998; McNair et al., 1992), or by using a single assessment load corresponding to either 10% of the participant's body mass (D R Bell et al., 2012; David R. Bell et al., 2011; David R Bell et al., 2009; J Troy Blackburn et al., 2009; J. Troy Blackburn et al., 2009, 2011, 2004; J.Troy Blackburn et al., 2004; Waxman et al., 2015) or 45% of the participants MVIC torque (J. Troy Blackburn et al., 2013; J. Troy Blackburn & Norcross, 2014; J. Troy Blackburn & Pamukoff, 2014; Swanik et al., 2004; Watsford et al., 2010). From the information presented on the stiffness-load relationship above, it can be reasoned that the measurement of $K_{\rm HAM}$ obtained under a specific loading condition reflects the level of stiffness that the surrounding joint structures display under that specific level of tension. Therefore, it has been suggested that the choice of an assessment load should be justified by the particular research question at hand, and that caution should be exercised when attempting to compare findings between studies using different loads (Ditroilo, Watsford, Murphy, et al., 2011). Because the hamstrings are reported to be activated approximately 30% MVIC during the stance phase of gate (Ciccotti, Kerlan, Perry, & Pink, 1994), it would appear that researchers interested in the influence of K_{HAM} on knee joint stability during the stance phase of gait would want to assess K_{HAM} using a load that evokes a similar neuromuscular response. Conversely, higher assessment loads may be more representative of the level of neuromuscular activation required during high-intensity athletic maneuvers (e.g. landing from a jump), and may therefore be more

relevant when researchers are interested in the influence of K_{HAM} on knee joint stability in athletic populations.

Based on the findings above, it seems intuitive that researchers interested in examining the extent to which K_{HAM} contributes to biomechanical factors that directly influence ACL loading should be encouraged to use higher assessment loads. However, it has been noted that although higher assessment loads may be desirable, some participants may be unable to tolerate such loads (Ditroilo, Watsford, & De Vito, 2011), which would increase the overall variability in the measure. Although additional research is needed, it may be the case that higher assessment loads simply aren't feasible in certain populations of interest. Therefore, there may be a certain trade-off between the assessment load used and the quality of the data collected. Thus, researchers should evaluate the physical status of their population of interest, as well as their experimental design and research question, before deciding on an assessment load in future work.

Perturbation Magnitude. Applying a perturbation to the system (i.e. lower-extremity) and recording the ensuing damped oscillations is inherent in the free-oscillation technique. Published literature on K_{HAM} regularly characterize the perturbation as 'a brief downward push' manually applied to the posterior aspect of the calcaneus, and state that the application of the perturbation should be sufficiently gentle in order to prevent bursts of neuromuscular activation as a result of eliciting a reflexive response. Some authors have reported the magnitude of the applied perturbation to be in the order of 100-150 N, but neglected to include a detailed description of how the perturbation magnitude was controlled or measured (Ditroilo, Watsford, & De Vito, 2011; Ditroilo et al., 2013; Swanik et al., 2004; Watsford et al., 2010). However, others have calculated the perturbation magnitude as the product of the peak tangential shank segment acceleration and system mass (i.e., the summed mass of the shank and foot segment and the applied load), and these investigations have reported mean perturbation magnitudes between 30

and 139 N (J. Troy Blackburn et al., 2009, 2013; Waxman et al., 2015). This perturbation technique has been rationalized mechanically in that such a system will oscillate at its natural or resonant frequency, regardless of the magnitude of perturbation. (G J Wilson, Murphy, & Pryor, 1994) In support of this rationale, two studies found no relationship between K_{HAM} and perturbation magnitude, demonstrating that the portion of variance in K_{HAM} that can be attributed to perturbation magnitude is negligible (J. Troy Blackburn et al., 2013, 2011). However, a more recent study indicated that perturbation magnitude does in fact influence K_{HAM} (Waxman et al., 2015). Given these mixed reports and the relatively scant number of published findings in this area, it currently appears that future studies should attempt to place strict control over the application of the manual perturbation until this question can be further studied.

Instructions to Participants and the use of Surface Electromyography. Although this review has highlighted variation in hip and knee joint positioning between studies, evidence suggests that changes in hip and knee joint positioning does have an effect on both hamstring torque production and neuromuscular activation due to changes in hamstring muscle moment-arm lengths and length-tension relationships (Barr & Duncan, 1988; Smidt, 1973; Worrell et al., 2001). Thus, each individual study does require strict control over joint positioning in order to accurately obtain measures of K_{HAM} . In addition, the potential effect that reflexive neuromuscular responses and quadriceps co-contraction can have on K_{HAM} suggests that such factors need to be minimized to ensure the most accurate measures of K_{HAM} . Therefore, given the relative complexity of the assessment procedures, it seems imperative that participants be provided explicit instructions and are allowed adequate time to become familiarized to the task. In the current body of literature however, clear descriptions of the methods employed to provide participants with instructions and adequate familiarization to the task are rather limited. In a number of published papers, (D R Bell et al., 2012; David R. Bell et al., 2011; J. Troy Blackburn

et al., 2009, 2013, 2004; J.Troy Blackburn et al., 2004; Watsford et al., 2010) participants have been verbally instructed to contract their hamstring musculature to the level necessary to support the weight of their shank and foot, and the applied load, in the specified assessment position (knee flexion angle), and to try not to intervene on or voluntarily produce the oscillations following the perturbation. However, two other investigations (Kevin P. Granata et al., 2002; Swanik et al., 2004) have displayed surface electromyography data in real-time, and provided verbal cues, as a method to help participants focus on maintaining a constant level of hamstring contraction while minimizing any co-contraction of the quadriceps muscles.

Surface electromyography (sEMG) has also been used by a number of other studies, although not to provide participants with real-time feedback. Instead, sEMG has generally been used to evaluate the normalized neuromuscular activation of the hamstrings in response to the load applied, to visually inspect the data captured during each trial for any unwanted bursts of activity that may have occurred in response to the perturbation and ensure that the participants were not voluntarily generating the oscillations (characterized by a lack of decay in the oscillatory profile and a succession of bursts in the sEMG record), and to check for any unwanted cocontraction of the quadriceps (McNair et al., 1992). These general characteristics have since been adopted as criteria for defining an acceptable trial, with some investigators monitoring the sEMG recordings in real-time and having participants repeat trials deemed to be unacceptable (J. Troy Blackburn et al., 2004; J.Troy Blackburn et al., 2004; Ditroilo, Watsford, & De Vito, 2011; Ditroilo et al., 2013; Kevin P. Granata et al., 2002). However, Jennings and Seedhom (Jennings & Seedhom, 1998) opted to not include the use of sEMG in their investigation because it had become apparent in their pilot testing (unpublished data) that any co-contraction of the quadriceps would lead to undamped oscillations; an observation also reported by McNair et al (McNair et al., 1992). This observation has also been supported by published data demonstrating that quadriceps

muscle co-contraction is typically rather small during the assessment of $K_{\rm HAM}$, with mean sEMG values ranging between 1-8%MVIC (Kevin P. Granata et al., 2002). Therefore, although the use of sEMG may serve several valuable purposes, such as those previously discussed, it does add an additional level of complexity to the research design and does not appear to be absolutely necessary for the assessment of $K_{\rm HAM}$ alone.

Summary. The current section has presented the overall measurement of K_{HAM} and the experimental procedures involved, as well as potential issues concerning participant positioning, the stiffness-load relationship and the methods of assigning the applied load, perturbation magnitude, instructions given to participants, and the use of sEMG. In terms of participant positioning, evidence suggests that assessing K_{HAM} in a prone position is ecologically valid because it allows researchers to approximate the length-tension relationship of the hamstrings that occurs during functional activities; however, hip and knee joint positioning does have an effect on both neuromuscular activation and torque production, which thereby has an indirect influence on K_{HAM} . Tibial rotation also has the ability to influence K_{HAM} through altered neuromuscular activation of the hamstrings. Therefore, hip and knee joint positioning need to be strictly controlled when assessing K_{HAM} and caution should be taken when attempting to compare findings between studies that have assessed K_{HAM} using different hip and knee flexion angles. The same is true for the perturbation; although current evidence is limited, there is data to suggest that perturbation magnitude does have an effect on K_{HAM} . Thus, future studies should attempt to standardize the magnitude of the perturbation across both trials and participants.

In terms of the stiffness-load relationship and the methods of assigning the applied load, it must be recognized that the $K_{\rm HAM}$ value obtained under a certain applied load reflects the amount of stiffness that the surrounding joint structures display at a specific level of tension. Therefore, the choice of an assessment load should be justified by the particular research question

at hand, and that caution should be exercised when attempting to compare findings between studies using different loads. Ideally, studies interested in understanding the role of $K_{\rm HAM}$ during functional movement should attempt to use an applied load that elicits a similar neuromuscular response to that of the functional movement itself. However, given that some individuals may not be able to tolerate higher assessment loads, there may be a certain trade-off between the assessment load used and the quality of the $K_{\rm HAM}$ data collected. Lastly, it does not appear that the use of sEMG is necessary when assessing $K_{\rm HAM}$ as long as explicit instructions are provided and adequate time for familiarization is allowed. The investigator should however visually inspect the oscillatory profile recorded during each trial in order to ensure the overall quality of the data.

Hamstring Musculo-Articular Stiffness as it Relates to Dynamic Knee Stability

As discussed previously, dynamic knee stability is accomplished through a complementary relationship between passive and active restraint mechanisms (Johansson & Sjolander, 1993; Lew et al., 1993). It has also been discussed that the ACL provides more than 80% of total passive restraint to anterior tibial translation (Butler et al., 1980), and that the hamstring muscles function synergistically with the ACL (Baratta et al., 1988; Solomonow et al., 1987). Together, such findings suggest that the hamstring muscles may play a key role in stabilizing role during functional movement. In this regard, it was observed in an early investigation that ACL-deficient individuals, who were capable of preventing a pivot-shift in their ACL-deficient knee though increased neuromuscular activation of their hamstrings, returned to higher levels of functional activity following injury compared to those who could not (Walla et al., 1985). In addition, other researchers found that ACL-deficient individuals who completed a training program designed to improve the reaction time of the hamstrings, experienced fewer episodes of the knee giving way compared to those who did not undergo training (Ihara &

Nakayama, 1986). Anterior tibial translation occurs during episodes of the knee giving way, and it was later rationalized by McNair et al (McNair et al., 1992) that this anterior tibial translation is likely to stretch the hamstring muscles as well as the secondary ligamentous and capsular restraints. Given that stiffness is simply the ratio of change in force to change in muscle length, McNair et al (McNair et al., 1992) then hypothesized that higher levels of K_{HAM} might effectively resist such anterior tibial translation, thereby enhancing dynamic knee stability in conservatively managed ACL-deficient individuals. In testing this hypothesis, McNair et al (McNair et al., 1992) found that ACL-deficient individuals' knee functional ability was positively associated with $K_{\rm HAM}$ at multiple applied loads. It was also found that there were no bilateral differences in $K_{\rm HAM}$ between the ACL-deficient individuals' injured and uninjured limbs, which led McNair et al (McNair et al., 1992) to postulate that individuals with higher levels of K_{HAM} may have a greater likelihood of returning to higher levels of competition following ACL rupture. Although the relationship between K_{HAM} and knee stability is a significant addition to the literature in its own right, the secondary finding of no bilateral differences in K_{HAM} between injured and uninjured limbs has been questioned by others (Jennings & Seedhom, 1998; Swanik et al., 2004) due to previous reports of the ACL potentially playing a role in regulating stiffness (Johansson, Sjölander, & Sojka, 1991).

Jennings and Seedhom (Jennings & Seedhom, 1998) investigated K_{HAM} in a mixed cohort of ACL-injured individuals (some having previously undergone surgical reconstruction and others who had not) and a group of healthy (uninjured) controls and found that, although healthy individuals demonstrated no bilateral differences in K_{HAM} , ACL-injured individuals demonstrated greater K_{HAM} in their injured- compared to uninjured-limb. Additionally, no significant differences in K_{HAM} were observed between the ACL-injured individuals' healthy contralateral limb and that of the healthy controls (Jennings & Seedhom, 1998). The finding of greater K_{HAM} in

the ACL-injured limb versus healthy contralateral control limb appears to be supported by the work of Johansson et al. (Johansson et al., 1991), which suggested that the ACL plays a role in regulating muscle stiffness by potentially pre-programming intrinsic muscle stiffness via reflexmediated stiffness through the γ -muscle-spindle system, thereby regulating the stability of the joint. Hence, when the ACL becomes injured, it is plausible that K_{HAM} may become altered in that limb in order to provide some compensatory protection to the knee joint in the absence of the sensory contribution of the ACL to knee stabilization. In contrast, however, Swanik et al (Swanik et al., 2004) investigated differences in K_{HAM} between ACL-injured individuals and healthy controls, and found that ACL-injured individuals displayed significantly lower K_{HAM} than healthy individuals. It has been argued that the overall lack of agreement between studies may be partially explained by the role of rehabilitation programs in potentially altering K_{HAM} (Jennings & Seedhom, 1998). For example, the ACL-injured individuals studied by both McNair et al (McNair et al., 1992) and Swanik et al (Swanik et al., 2004) had undergone rehabilitation programs, which may have had an effect on K_{HAM} , whereas the ACL-injured individuals studied by Jennings and Seedhom (Jennings & Seedhom, 1998) had not. To muddy the waters further, a prospective investigation on the relationship between K_{HAM} and acute hamstring injury in Australian rules football reported that injured players displayed significantly greater K_{HAM} than uninjured players (Watsford et al., 2010). Additionally, it was reported that although bilateral averages for K_{HAM} differed between injured and uninjured players, K_{HAM} in the injured limb was not different from that of uninjured players; rather, K_{HAM} in the uninvolved limb of injured players was found to be significantly greater than that of the uninjured cohort (Watsford et al., 2010). This overall uncertainty regarding how K_{HAM} potentially contributes to knee stability and ACL loading, how it is potentially modified post-injury, and whether or not it can be modified via targeted training, has prompted other investigations to examine such topics in healthy (uninjured) male and female populations.

Blackburn, Norcross, and Padua (J. Troy Blackburn et al., 2011) first investigated the influence of K_{HAM} on anterior tibial translation in healthy males and females by first assessing $K_{\rm HAM}$ using an applied load equal to 10% of the participant's body mass (Figure 2.5A), and then eliciting anterior tibial translation by releasing a 20% body mass load, attached to posterior aspect of the proximal shank, which abruptly shifted the tibia anterior relative to the femur (Figure 2.5B). Based on the median anterior tibial translation value, these male and female participants were divided in to two groups (i.e. high versus low anterior tibial translation); and these groups were then compared on measures of K_{HAM}, anterior tibial translation, MVIC hamstring strength, and hamstring neuromuscular activation. Compared to the low anterior tibial translation group, the high anterior translation group was reported to display greater anterior tibial translation and lower K_{HAM} , while hamstring strength and hamstring neuromuscular activation (J. Troy Blackburn et al., 2011) were similar between groups. Additionally, after grouping all individuals together, K_{HAM} was reported to be significantly and negatively correlated with anterior tibial translation ($R^2 = 0.29$); however; no other significant correlations were found (J. Troy Blackburn et al., 2011). Based on such findings, it was proposed that higher levels of K_{HAM} , but not hamstring strength, may help enhance knee joint stability and reduce ACL loading, whereas lower levels of K_{HAM} may increase ACL injury risk by allowing greater anterior tibial translation (J. Troy Blackburn et al., 2011). This theory then led to a second study by the same research group which aimed to examine the influence of K_{HAM} on lower-extremity kinematics and kinetics in healthy males and females during a double-leg jump-landing task (J. Troy Blackburn et al., 2013). In this study, K_{HAM} was assessed using an applied load equal to 45% of the participant's MVIC hamstring torque while lower-extremity kinematics and kinetics were assessed during a

double-leg jump-landing task, which involved performing a jump-landing from a 0.3-meter tall box placed 50% of the participant's body height away from two force platforms (J. Troy Blackburn et al., 2013). After equally stratifying males and females into two groups (i.e. high K_{HAM} versus low K_{HAM}), it was reported that both groups displayed similar peak knee-flexion and knee-valgus (abduction) angles, but that individuals with higher K_{HAM} values displayed greater knee-flexion angles at the instants of peak internal knee-varus moment, peak internal knee-extension moment, and peak proximal tibia anterior shear force (J. Troy Blackburn et al., 2013). Further, individuals in the high K_{HAM} group displayed significantly smaller peak internal knee-varus moments, and a "statistical trend" (although not statistically significant) towards lesser proximal tibia anterior shear force (effect size = 0.63), compared to individuals in the low K_{HAM} group (J. Troy Blackburn et al., 2013).

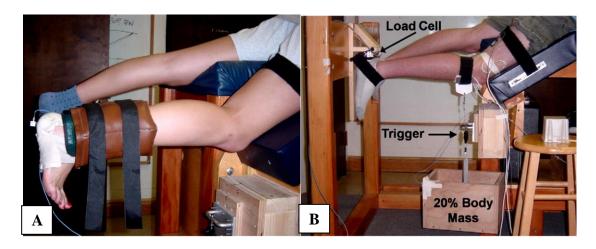


Figure 2.5. (A) Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment Using an Applied Load Equal to 10% of the Participant's Body Mass; (B) Experimental Apparatus Used to Elicit Anterior Tibial Translation (J. Troy Blackburn et al., 2011).

Although the primary focus of this dissertation is on biomechanical factors that directly influence ACL loading in the sagittal plane (i.e. proximal tibial anterior shear force, anterior tibial acceleration, and anterior tibial translation), the fact that noncontact ACL injuries likely involve

combined knee-joint loading in multiple planes (Shimokochi & Shultz, 2008) should not be ignored. In this regard, Hewett et al (Timothy E Hewett et al., 2005) prospectively screened female athletes performing double-leg drop jumps prior to their athletic seasons and demonstrated that athletes who went on to experience noncontact ACL injuries displayed knee valgus angles that were 8° greater, and external knee-valgus moments that were 2.5 times greater, than those who completed the season uninjured. In addition, the external knee-valgus moments obtained during this preseason screening were able to predict noncontact ACL injury with 73% sensitivity and 78% specificity (Timothy E Hewett et al., 2005). While this has been the only study to prospectively identify knee-valgus moments as a risk factor for noncontact ACL injury, retrospective video analyses support the theory that knee valgus may likely be involved at the time of injury (Koga et al., 2010; Tron Krosshaug et al., 2007; Olsen et al., 2004). Given that Blackburn et al (J. Troy Blackburn et al., 2013) found that individuals in the low K_{HAM} group displayed peak internal knee-varus moments (i.e. the musculoskeletal response to an external knee-valgus moment) that were 3.6 times greater than the high K_{HAM} group, such findings may suggest that higher levels of K_{HAM} also potentially enhance dynamic knee stability in the frontal plane.

In combination, the findings reported by Blackburn et al (J. Troy Blackburn et al., 2013, 2011) provide evidence to support the theory that greater K_{HAM} , but not hamstring strength, potentially enhances knee joint stability, and that lesser (or insufficient) K_{HAM} may result in increased knee joint loading, potentially increasing noncontact ACL injury risk. This theory is further supported by the findings of McNair et al (McNair et al., 1992), in which ACL-deficient individuals with higher K_{HAM} displayed greater knee function than those with lower K_{HAM} . Because individuals with higher K_{HAM} are reported to display greater knee function (McNair et al., 1992), less anterior tibial translation (J. Troy Blackburn et al., 2011) and proximal tibia

anterior shear force (J. Troy Blackburn et al., 2013), and smaller internal knee-varus moments (J. Troy Blackburn et al., 2011), than individuals with lower K_{HAM} , a more recent attempt has been made to examine the extent to which K_{HAM} can be modified via targeted training.

Blackburn and Norcross (J. Troy Blackburn & Norcross, 2014) aimed to determine whether K_{HAM} could be enhanced via isometric and isotonic training, and whether enhancing K_{HAM} would alter knee joint biomechanics in a manner indicative of reduced ACL loading. In this study, healthy male and female participants were randomly assigned to an isometric training group, isotonic training group, or a control group, and the effects of a 6-week of training on K_{HAM} , hamstring strength, hamstring neuromuscular activation, anterior tibial translation, and landing biomechanics, were then evaluated. There was no statistically significant group by time interaction observed; however, K_{HAM} significantly increased (15.7%) pre- to post-training in the isometric training group (J. Troy Blackburn & Norcross, 2014). Within the isometric training group, no changes in hamstring strength or neuromuscular activation were observed pre- to post-training, and K_{HAM} was not found to be correlated with either of these measures (J. Troy Blackburn & Norcross, 2014). In terms of ACL loading parameters, anterior tibial translation, proximal tibia anterior shear force, and internal knee-varus moment changed pre- to post-training in a manner consistent with reduced ACL loading; however none of these changes reached statistical significance (J. Troy Blackburn & Norcross, 2014).

Increases in stiffness have previously been reported in response to 10 weeks of either endurance, plyometric, or isometric training (Grosset, Piscione, Lambertz, & Pérot, 2009; K Kubo, Kanehisa, Ito, & Fukunaga, 2001); however, these studies have measured tendon stiffness specifically, which is different from the global measure of musculo-articular stiffness. Based on the first and only study (J. Troy Blackburn & Norcross, 2014) to evaluate the effect of targeted training on the enhancement of K_{HAM} , it appears that K_{HAM} may in fact be a modifiable

neuromechanical property. In addition, the finding that stiffness and strength are unrelated properties is supported by previous work on $K_{\rm HAM}$ (J. Troy Blackburn et al., 2011) and on tendon stiffness (Keitaro Kubo et al., 2009). Muscle strength quantitatively describes the ability of the muscle to produce force whereas stiffness quantitatively describes the ability to resist muscle lengthening. Therefore, Blackburn and Norcross (J. Troy Blackburn & Norcross, 2014) suggested that the lack of a relationship between strength and $K_{\rm HAM}$ likely indicates that changes in $K_{\rm HAM}$ can be attributed to enhanced neural efficiency and changes in material and/or architectural musculotendinous properties as opposed to improved strength. However, a separate study conducted by this same research group reported that $K_{\rm HAM}$ and strength were positively correlated with one another ($R^2 = 0.29$) and contended that the relationship between $K_{\rm HAM}$ and strength was intuitive, in that a muscle that is capable of producing greater force should also be able to provide greater resistance to lengthening (J. Troy Blackburn & Pamukoff, 2014).

Summary. Collectively, the rather equivocal results regarding whether or not unilateral or bilateral differences in $K_{\rm HAM}$ exist in ACL-injured populations, and how too much or too little $K_{\rm HAM}$ may be related to knee stability and hamstring injury, illustrates the complexity of understanding the functionality, or clinical relevance, of this measure. Numerous factors appear to be involved in the regulation of $K_{\rm HAM}$, such as hip and knee joint positioning and preparatory and reactive neuromuscular control strategies, among others (McNair & Marshall, 1994; G J Wilson et al., 1994, 1991). For example, Bach et al (T. M. Bach, Chapman, & Calvert, 1983) suggested that the neuromuscular control apparatus modifies stiffness, depending on the requirements of the task, in order to optimize the mechanical properties of muscle. Wilson et al (G J Wilson et al., 1994) and Rudolph et al (Rudolph, Axe, Buchanan, Scholz, & Snyder-Mackler, 2001) have proposed that lower stiffness may be advantageous during functional activities for more efficient absorption of joint loads and storage of elastic energy. Conversely,

Granata et al. (K. P. Granata et al., 2002) suggested that too little stiffness may permit excessive joint motion, resulting in greater loading of the passive joint restraints, thereby increasing injury risk. Although a positive relationship been K_{HAM} and knee joint function has been demonstrated in ACL-deficient individuals (McNair et al., 1992), a second study by McNair and Marshall (McNair & Marshall, 1994), studying drop-jump landings, revealed that ACL-injured individuals displayed greater preparatory hamstring muscle activity, lower ground reaction forces, and lower $K_{\rm HAM}$ compared to an uninjured cohort. This finding is supported by another study which reported that ACL-injured individuals demonstrated significantly greater preparatory activity in the lateral hamstring, less K_{HAM} , and were relatively functional (based on single leg hop maximal distance and Lysholm knee rating scale) when compared to uninjured controls (Swanik et al., 2004). Such findings support the work of Rudolph et al (Rudolph et al., 2001) and suggest that there is an important relationship or interaction among neuromuscular activation amplitude and timing, stiffness, and function, in ACL-injured individuals. Swanik et al (Swanik et al., 2004) suggest however, that additional research within ACL-injured populations is needed because it currently remains unclear whether reduced K_{HAM} is a genetic, predisposing factor for injury, or a compensatory adaptation benefiting the dynamic restraint mechanism.

The same appears to be true for healthy populations. Although healthy individuals with higher levels of K_{HAM} display less anterior tibial translation during controlled open-chain perturbations translation (J. Troy Blackburn et al., 2013, 2011), and knee biomechanics indicative of lesser ACL loading during double-leg jump landings (J. Troy Blackburn et al., 2013; J. Troy Blackburn & Norcross, 2014), the conclusions that can be drawn from such findings regarding the influence of K_{HAM} on ACL loading parameters are limited for several reasons. First, research demonstrates that noncontact ACL injuries most often occur in a closed-kinetic-chain when cutting or landing on a single leg (B P Boden et al., 2000; Barry P Boden et al., 2009; Koga et al.,

2010; Olsen et al., 2004). Thus, open-kinetic-chain perturbations and double-leg landing tasks may not adequately represent the situations in which noncontact ACL injuries commonly occur. Second, the reported relationships between K_{HAM} and ACL loading parameters have been established using two different methods of assigning the applied assessment load (i.e. 10% body mass vs. 45% MVIC torque). Because K_{HAM} is reported to be influenced by differences in the applied load and thus differences in neuromuscular activation, the comparisons that can be made between these studies are limited. Third, the reported relationships between K_{HAM} and ACL loading parameters have been established with males and females included in the same statistical analyses, without equal sex-stratification. This review has previously highlighted the fact that females perform functional landing tasks with a more posterior center of mass position (DiStefano et al., 2005; Yu et al., 2006), less hip and knee flexion (Schmitz et al., 2007), higher quadriceps and lower hamstring muscle activation (Malinzak et al., 2001), and greater posterior ground reaction forces and knee extensor moments (Schmitz et al., 2007), than similarly trained males. Additionally, females are reported to display greater knee valgus angles and external kneevalgus moments compared to similarly trained males (Barry P Boden et al., 2009; Tron Krosshaug et al., 2007; McLean, Huang, Su, & Van Den Bogert, 2004; McLean, Huang, & van den Bogert, 2008). As it will be discussed in the next section, females are also reported to display less K_{HAM} than males (J. Troy Blackburn et al., 2009). Because previous studies on relationships between K_{HAM} and ACL loading parameters have failed to control for the potential influence of these between-sex differences, it could be argued that the true extent to which K_{HAM} is associated with biomechanical factors that directly influence ACL loading (i.e. proximal tibia anterior shear force, anterior tibial translation, and anterior tibial acceleration) during functional landing tasks remains unknown. Finally, although isometric training has been demonstrated to increase K_{HAM} in the absence of increases in muscle strength (J. Troy Blackburn & Norcross, 2014), conflicting

reports regarding the relationship between K_{HAM} and strength have been presented, and the underlying adaptations which contribute to enhanced K_{HAM} have not yet been identified.

Intrinsic Factors that Contribute to Hamstring Musculo-Articular Stiffness

A number of studies have been conducted in an effort to better understand the underlying intrinsic factors that contribute to an individual's K_{HAM} . To date, such studies have investigated potential relationships between K_{HAM} and mechanical, geometric, and architectural properties of skeletal muscle (J. Troy Blackburn et al., 2011, 2004; J. Troy Blackburn & Pamukoff, 2014), between-sex differences in these properties (J. Troy Blackburn et al., 2009; J. Troy Blackburn & Pamukoff, 2014; J.Troy Blackburn et al., 2004), and the influence of female menstrual cycle hormones (D R Bell et al., 2012; David R. Bell et al., 2011; David R Bell et al., 2009; Eiling, Bryant, Petersen, Murphy, & Hohmann, 2007). Unfortunately however, the underlying factors that influence an individual's K_{HAM} are still relatively unclear. The findings of such studies are presented throughout the remainder of this section.

Mechanical Properties of Skeletal Muscle. Flexibility (or extensibility) of the hamstrings has been reported to contribute to noncontact ACL injury risk, in that individuals with greater flexibility experience injuries more often than individuals with less flexibility (B P Boden et al., 2000). In addition, females display greater flexibility than males (J.Troy Blackburn et al., 2004; Hutchinson & Ireland, 1995), and are also at an increased risk for noncontact ACL injury (El A Arendt et al., 1999; Dick et al., 2007). Flexibility is defined as the available range of motion at a given joint, and provides an indication of the muscle's ability to elongate without consideration of the associated force response (Gleim & McHugh, 1997). In contrast, stiffness is mechanically defined as the ratio of change in force to a change in length, or simply the ability to resist lengthening. Given these definitions, it has been theorized that stiffness and flexibility may

be related concepts in that the denominator of the stiffness equation, change in length, may be influenced by one's flexibility (J. Troy Blackburn et al., 2004). Additionally, because $K_{\rm HAM}$ is derived from both contractile and non-contractile components (i.e. series elastic, parallel elastic, and contractile components), reduced passive resistance to muscle lengthening (i.e. passive stiffness) may result in decreased K_{HAM} for a given level of neuromuscular activation, which suggests that passive stiffness may also be a contributory factor (J. Troy Blackburn et al., 2004). In examining such relationships, Blackburn et al (J. Troy Blackburn et al., 2004) demonstrated that passive hamstring stiffness accounted for 25% of the variance in $K_{\rm HAM}$, but that hamstring flexibility only accounted for an additional 2% of the variance in K_{HAM} beyond what could be explained by passive stiffness alone. From these findings, it was concluded that the identified relationship between passive stiffness and K_{HAM} emphasized the dependence of K_{HAM} on muscle cross-bridge formation versus the relatively smaller contribution from the parallel elastic tissue (J. Troy Blackburn et al., 2004). However, the above findings are limited due to the fact that the statistical analyses used to examine such relationships included both males and females and that these analyses were performed on absolute, rather than normalized, values. This is problematic because an earlier study using the same sample demonstrated that males displayed greater K_{HAM} and passive hamstring stiffness than females, while females displayed greater hamstring flexibility than males (J.Troy Blackburn et al., 2004). However, these between-sex differences were accounted for after normalizing K_{HAM} to the applied moment (i.e. the product of the total system mass and length of the shank segment), and normalizing passive stiffness and flexibility to the mass of the thigh segment (J. Troy Blackburn et al., 2004). Therefore, it seems plausible that the relationships reported may have been driven by between-sex differences as opposed to there being a true relationship between these properties, which reinforces the need for sex-stratified designs in future studies.

Skeletal Muscle Geometry and Architecture. As previously mentioned, K_{HAM} is most often assessed using an applied load equal to either 10% of the participant's body mass or 45% of the participant's MVIC torque. Because males generally display greater body mass and body height, it has been demonstrated that males experience a significantly greater applied moment [i.e. the product of the total system mass (applied load + shank and foot segment mass) and the length of the shank segment] when K_{HAM} is assessed using a 10% body mass load (J.Troy Blackburn et al., 2004) Despite the greater applied moment however, males and females have been reported to display no differences in neuromuscular activation (normalized sEMG amplitude) of the hamstrings during the assessment of K_{HAM} , which has been suggested to indicate that males and females are loaded similarly from a neural perspective (J.Troy Blackburn et al., 2004). Taken together, this suggests that males are able to produce a greater resistive (internal) moment than females for a given level of neuromuscular effort (J.Troy Blackburn et al., 2004). Males possess greater muscle mass and muscle cross-sectional area than females (Chow et al., 2000; Miller, MacDougall, Tarnopolsky, & Sale, 1993; Staron et al., 2000). Therefore, the greater K_{HAM} displayed by males may simply be a function of increased muscle mass and crosssectional area, thus allowing males to resist a greater load at a similar level of muscle activation, potentially protecting the ACL from deleterious loading at a reduced metabolic cost compared to similarly trained females (J.Troy Blackburn et al., 2004). The notion that between-sex differences in K_{HAM} may be related to differences in the material properties of muscle has led to investigations aimed to assess the influence of such properties on K_{HAM}, as well as between-sex differences therein.

Between-sex differences in muscle geometry and architecture have previously been identified for the soleus, gastrocnemius, and triceps surae musculature, and it has been suggested that these differences contribute to between-sex variability in musculo-articular stiffness (J. Troy

Blackburn et al., 2006; Chow et al., 2000; Keitaro Kubo, Kanehisa, & Fukunaga, 2003). However, only two studies have evaluated the influence of structural and material properties on K_{HAM} (J. Troy Blackburn et al., 2009; J. Troy Blackburn & Pamukoff, 2014). Collectively, these studies have investigated between-sex differences in hamstring muscle (biceps femoris) and fascicle length, cross-sectional area, stress, strain, elastic modulus, hamstring muscle strength, posterior thigh fat thickness, and hamstring (biceps femoris) tendon stiffness. These studies have also examined the associations between these factors and K_{HAM} . In general, elastic modulus, stress, and strain, and fascicle length have been shown to be similar between sex; however females are reported to have shorter resting muscle length and smaller muscle cross-sectional area, less hamstring strength and tendon stiffness, and greater posterior thigh fat thickness, compared to males (J. Troy Blackburn et al., 2009; J. Troy Blackburn & Pamukoff, 2014). In examining relationships between these factors, it has been reported that K_{HAM} is positively associated with cross-sectional area (J. Troy Blackburn et al., 2009; J. Troy Blackburn & Pamukoff, 2014), tendon stiffness, fascicle length, and strength, and negatively associated with posterior thigh fat thickness (J. Troy Blackburn & Pamukoff, 2014). However, after normalizing these variables to body mass, posterior thigh fat thickness and strength were the only factors significantly associated with K_{HAM} (J. Troy Blackburn & Pamukoff, 2014). Similar to the findings regarding the influence of active extensibility and passive stiffness on $K_{\rm HAM}$, these findings are based on statistical analyses that combine males and females, making it difficult to draw definitive conclusions. Further, although hamstring strength has been shown to be positively associated with K_{HAM} (J. Troy Blackburn & Pamukoff, 2014), other studies performed by the same lab group have reported no relationship between normalized strength and K_{HAM} (J. Troy Blackburn et al., 2011; J. Troy Blackburn & Norcross, 2014). Thus, there is a need for additional studies that incorporate sex-stratified designs.

Menstrual Cycle Hormones. There is a general consensus that the risk of experiencing a noncontact ACL injury is not equal across phases of the menstrual cycle, with the greatest risk occurring during the pre-ovulatory phase (i.e. from menses onset to ovulation) (Shultz et al., 2010). This is due to previous studies demonstrating an effect of circulating menstrual cycle hormones on soft tissue mechanics (Belanger et al., 2004; Heitz, Eisenman, Beck, & Walker, 1999; Karageanes, Blackburn, & Vangelos, 2000; Romani, Curl, Lovering, & McLaughlin, 2001). For example, estrogen and progesterone receptors have been identified on the ACL (Liu et al., 1996), and physiological levels of estrogen have been demonstrated to reduce collagen synthesis, thereby making the ACL more susceptible to injury (Liu, Al-Shaikh, Panossian, Finerman, & Lane, 1997). In support of this, anterior knee laxity (ligamentous laxity) has been reported to increase near ovulation and during the latter half of the menstrual cycle (the time at which estrogen concentrations are highest) (Deie, Sakamaki, Sumen, Urabe, & Ikuta, 2002; Heitz et al., 1999; Romani et al., 2001; Shultz, Gansneder, Sander, Kirk, & Perrin, 2006). However, others have reported that laxity does not change across the menstrual cycle (Belanger et al., 2004; Bruce D Beynnon et al., 2006; Eiling et al., 2007; Karageanes et al., 2000). Similarly, estrogen receptors have also been identified within skeletal muscle (Lemoine et al., 2003), which are believed to modulate muscle strength (Sarwar, Niclos, & Rutherford, 1996) and muscle metabolism (Hackney, 1999). However, other studies have reported that muscle strength does not change across the menstrual cycle (Abt et al., 2007; Fridén, Hirschberg, & Saartok, 2003; Hertel, Williams, Olmsted-Kramer, Leidy, & Putukian, 2006; K. Kubo et al., 2009). Other studies on the effects of menstrual cycle hormones on stiffness have also produced somewhat equivocal results.

To date, four studies have investigated the effects of menstrual cycle hormones on measures of lower-extremity stiffness. Eiling et al. (Eiling et al., 2007) studied the effects of estrogen (across the menstrual cycle) on lower-extremity hopping stiffness and reported

significantly lower stiffness at the time of ovulation (the time at which estrogen levels were highest). In contrast, Bell et al (David R Bell et al., 2009) evaluated K_{HAM} specifically and found no statistically significant differences between post-menses (within 3 days following a selfreported onset of menses) and post-ovulation (within 3 days after ovulation) phases of the menstrual cycle. Null results were also reported in a later study by Bell et al (David R. Bell et al., 2011) who found that lower-extremity hopping stiffness and K_{HAM} were not influenced by hormonal fluctuation across the menstrual cycle or by the use of oral contraceptives. More recently however, Bell et al (D R Bell et al., 2012) examined whether estrogen and free testosterone concentrations were associated with K_{HAM} in females during the follicular phase of the menstrual cycle (3-5 days after menses onset) and found K_{HAM} to be negatively associated with both free testosterone and estrogen. Although the collective findings of this work are mixed, available comparisons between studies are limited due to differences in the testing time points used within the menstrual cycle, the methods of identifying these time points, and the methods of obtaining hormone concentrations. Participants were required to have regular menstrual cycle histories for 3-6 months prior to participation in all studies; however, Eiling et al., 2007) actually monitored each individual's menstrual cycle for regularity, whereas others relied on self-report (D R Bell et al., 2012; David R. Bell et al., 2011; David R Bell et al., 2009). Eiling et al (Eiling et al., 2007) examined four phases of the menstrual cycle, which were estimated by averaging the lengths of previous menstrual cycles and then using a calendar-based counting method to determine the testing time points. Testing then took place within two days of the estimated menses onset, mid-follicular, and mid-luteal phases, while testing during the ovulation phase took place on the exact calculated day of ovulation. Bell et al (David R. Bell et al., 2011; David R Bell et al., 2009) examined two phases, with testing sessions occurring within 3-5 days of self-reported menses onset and again following a positive urine-based ovulation test. In

contrast, another study by Bell et al (D R Bell et al., 2012) examined only the follicular phase by testing within 3-5 days of self-reported menses onset. It has previously been demonstrated that calendar-based methods for determining menstrual cycle phases are inadequate when the accurate identification of ovulation is essential, and that urinary-based ovulation tests should be used to more accurately identify menstrual cycle events (Wideman, Montgomery, Levine, Beynnon, & Shultz, 2012). With regard to differences in methods for obtaining hormone concentrations, one of three studies actually took blood samples to determine hormone concentrations (D R Bell et al., 2012; David R. Bell et al., 2011; Eiling et al., 2007), whereas one study did not measure any hormone concentrations; instead, the investigators attempted to create individualized testing sessions around each subjects menstrual cycle through the use of ovulation kits and then extrapolated that information to an average female hormonal profile (David R Bell et al., 2009). Based on these limitations, and the equivocal results identified, additional studies appear warranted.

Summary. Given that higher levels of K_{HAM} have been associated with higher functional knee ability in ACL-deficient individuals (McNair et al., 1992) and characteristics of lesser ACL loading in healthy individuals (J. Troy Blackburn et al., 2013, 2011), and that K_{HAM} has been shown to be modifiable (J. Troy Blackburn & Norcross, 2014), several studies have been conducted in an attempt to gain a greater understanding of the intrinsic factors that potentially contribute to an individual's K_{HAM} . This effort has also been in part motivated by the fact that females have been reported to display less K_{HAM} than males (J Troy Blackburn et al., 2009; J.Troy Blackburn et al., 2004; Kevin P. Granata et al., 2002) and coincidentally experience an increased risk of noncontact ACL injury (El A Arendt et al., 1999; Dick et al., 2007). Although the findings presented in this section suggest that intrinsic factors, such as passive hamstring stiffness and hamstring flexibility (J. Troy Blackburn et al., 2004), hamstring muscle cross-sectional area (J.

Troy Blackburn et al., 2009; J. Troy Blackburn & Pamukoff, 2014), posterior thigh fat thickness and hamstring strength (J. Troy Blackburn & Pamukoff, 2014), and circulating menstrual cycle hormones (D R Bell et al., 2012), influence *K*_{HAM}, it has also been shown that a number of these factors differ between sex. It has been demonstrated that some of these differences are removed after differences in anthropometric characteristics are accounted for (J. Troy Blackburn et al., 2004; J. Troy Blackburn & Pamukoff, 2014; Kevin P. Granata et al., 2002); however, some of these aforementioned relationships have been determined without accounting for such characteristics by normalizing data prior to statistical analysis or employing sex-stratified research designs. Based on such discrepancies, it could be argued that men and women are simply too different to be included in the same analyses, and should therefore be examined separately in future work.

Conclusion

This review of literature has presented what is currently known about the potential mechanism(s) of noncontact ACL injury, the factors that contribute dynamic knee stability and ACL loading during functional athletic movement, and the potential role of hamstring musculo-articular stiffness (K_{HAM}) in influencing ACL loading. Although the precise mechanism of noncontact ACL injury remains unclear, it is well accepted that the ACL is most directly loaded via anterior tibial translation (Butler et al., 1980; K. L. Markolf et al., 1995). It is also well accepted that anterior tibial translation naturally occurs as the relatively extended knee ($<30^{\circ}$ flexion) transitions from non-weight bearing to weight bearing (Fleming et al., 2001; Torzilli et al., 1994), and that appropriate neuromuscular control strategies are necessary to stabilize the knee joint during weight acceptance to protect the passive joint structures from deleterious loading (Riemann & Lephart, 2002). In this regard, the quadriceps muscles function

antagonistically to the ACL, and aggressive quadriceps contraction on a relative extended knee has been demonstrated to increase anterior tibial translation and ACL loading via increased net proximal tibia anterior shear force due to the quadriceps line of pull on the anteriorly oriented patellar tendon (DeMorat et al., 2004). In contrast, the hamstring muscles function agonistically with the ACL, and adequate co-contraction of this muscle group is capable of reducing overall anterior tibial translation and ACL loading by reducing this net anterior shear force by inducing a posterior shear force on the proximal tibia (Draganich & Vahey, 1990; Li et al., 1999; MacWilliams et al., 1999; Pandy & Shelburne, 1997; Withrow et al., 2006, 2008). However, due to the inherent difficulties associated with measuring muscle forces and ACL loading *in-vivo*, the demonstrated effects of hamstring co-contraction on ACL loading have been limited to cadaver models and musculoskeletal modeling simulation studies, or *in-vivo* during isometric knee-extension exercises. Therefore, the true extent to which the hamstrings are able to effectively reduce ACL loading during functional athletic tasks remains unknown.

Because K_{HAM} is simply the ratio of change in force to change in muscle-tendon unit length, it is theorized that individuals with higher K_{HAM} may have an increased capacity to resist proximal tibia shear force and thus anterior tibial translation, thereby resulting in less ACL loading compared to individuals with lower K_{HAM} . Although a direct link between K_{HAM} and noncontact ACL injury risk has yet to be established, current evidence suggests that higher levels of K_{HAM} may protect the ACL from deleterious loading during the time at which such injuries are reported to occur (J. Troy Blackburn et al., 2013, 2011). However, current evidence regarding the influence of K_{HAM} on knee joint biomechanics is limited to studies of open-kinetic-chain perturbations (J. Troy Blackburn et al., 2011) and double-leg jump-landing tasks (J. Troy Blackburn et al., 2013). The evidence presented in this review indicates that noncontact ACL injuries are more likely to occur during single-leg cutting and landing maneuvers (B P Boden et

al., 2000; Barry P Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), and laboratory-based studies show that single-leg landing tasks elicit biomechanical characteristics associated with increased knee joint loading compared to double-leg landing tasks (Pappas et al., 2007; Wang, 2011; Yeow et al., 2010). Thus, open-kinetic-chain perturbations and double-leg jump-landings may not adequately represent the situations in which noncontact ACL injuries commonly occur.

There are also a number of methodological issues associated with previous studies on the K_{HAM} which limits the generalizability of the findings reported. For example, some studies have assessed K_{HAM} by standardizing the assessment load as a percentage of each individual's body mass whereas other have standardized the assessment load as a percentage of each individual's maximal isometric hamstring torque. Because K_{HAM} is influenced by neuromuscular activation levels (Ditroilo, Watsford, & De Vito, 2011; Jennings & Seedhom, 1998), the ability to make comparisons between studies that have used different methods of standardizing the applied load is limited. In addition, much of what is currently known about the influence of K_{HAM} on biomechanical factors indicative of ACL loading has been established with males and females included in the same statistical analyses and without equal sex-stratification. This review has presented findings to suggest that females display less K_{HAM} than males (J. Troy Blackburn et al., 2009), and that females display a more posteriorly-oriented center of mass position (DiStefano et al., 2005; Yu et al., 2006), less hip and knee flexion (Schmitz et al., 2007), higher quadriceps and lower hamstring muscle activation (Malinzak et al., 2001), and greater posterior ground reaction forces and knee extensor moments (Schmitz et al., 2007) than males during functional athletic tasks. This is problematic because the combination of peak posterior ground reaction force, knee extensor moment, knee flexion angle, quadriceps muscle activation, and sex, has been shown to account for 86.1% of the variance in proximal tibia anterior shear force during a vertical stopjump task (Sell et al., 2007). Therefore, the independent contribution of $K_{\rm HAM}$ on biomechanical

factors that directly influence ACL loading (i.e. proximal tibia anterior shear force, anterior tibial translation, and anterior tibial acceleration) during functional landing tasks remains unknown. Also unknown is whether the factors that are predictive of peak proximal tibial anterior shear force during double-leg stop-jump tasks are similar for single-leg stop-jump tasks. Addressing these methodological factors are imperative if we are to fully understand the independent contribution of K_{HAM} as a contributing factor to ACL loading, thus our approach to injury prevention.

CHAPTER III

METHODS

Participants

Eighty healthy, highly-active, college-aged individuals (40 men, 40 women), between 18 and 30 years of age, were recruited from the University of North Carolina at Greensboro (UNCG) to participate in this study. To be eligible for participation, individuals needed to engage in greater than the equivalent of 300 minutes of moderate-intensity physical activity per week (as assessed via the International Physical Activity Questionnaire, Appendix B) and regularly participate in activities that involved running, cutting, jumping, and landing (e.g. basketball, soccer, tennis, rugby, and volleyball). Individuals were excluded from participation if they: 1) had ever injured their anterior or posterior cruciate ligaments (ACL and PCL, respectively), their medial or lateral collateral ligaments (MCL and LCL, respectively), or their medial or lateral menisci, 2) had experienced a lower-extremity injury within a 6-month window prior to recruitment, 3) had ever undergone lower-extremity surgery, 4) had any known medical conditions affecting their connective tissue or vestibular system, 5) were currently pregnant or attempting to become pregnant, or 6) were allergic to adhesive. Prior to enrollment in this study, all participants read and signed an informed consent form approved by the University of North Carolina at Greensboro's Institutional Review Board for the Protection of Human Subjects (Appendix A). Each participant received \$10 compensation for their participation in this study

Procedures

All data were collected during a single testing session in the Applied Neuromechanics Research Laboratory on the campus of UNCG. For female participants, testing was constrained to the follicular phase of the menstrual cycle (days 1-8 following self-report of the onset of menstrual bleeding) in order to control for any potential effects of cycling hormones on knee laxity (Park, Stefanyshyn, Loitz-Ramage, Hart, & Ronsky, 2009; Shultz, Gansneder, et al., 2006; Shultz, Kirk, Johnson, Sander, & Perrin, 2004), stiffness (D R Bell et al., 2012; Eiling et al., 2007; Park, Stefanyshyn, Loitz-Ramage, et al., 2009), or knee joint biomechanics (Park, Stefanyshyn, Ramage, Hart, & Ronsky, 2009; Shultz et al., 2011, 2012). Upon arrival to the laboratory, participants were asked to provide their written informed consent and complete the following intake documents: 1) Physical Activity and Healthy History Questionnaire, 2) International Physical Activity Questionnaire (IPAQ), and 3) Marx Activity Rating Scale (Appendix B). Additionally, female participants were asked to complete a menstrual cycle history questionnaire (Appendix B). Once this paperwork was completed and inclusion/exclusion criteria were confirmed, participants changed into laboratory-provided compression shorts and a tightfitting athletic top. Anatomical and anthropometric characteristics (body mass, body height, and anterior knee joint laxity) were then obtained using reliable methods previously established by our laboratory (Shultz, Nguyen, et al., 2006). Next, participants were instrumented with wireless surface electromyography (sEMG) sensors (Delsys Trigno; Delsys Inc., Boston, MA). Following sEMG sensor placement, participants performed a 5-minute warm-up on a stationary cycle ergometer (Life Fitness, Schiller Park, IL) at a cadence of 70-80 RPM and a target rating of perceived exertion (RPE) of \geq 3-4 on a Borg CR-10 RPE scale (Borg, 1998). Once the warm-up had been completed, the remainder of the experimental protocol was executed in the following

order: 1) quadriceps and hamstring maximal voluntary isometric contraction (MVIC), 2) hamstring musculo-articular stiffness (K_{HAM}), and 3) stop-jump landing biomechanics.

Anterior Knee Laxity

Anterior knee laxity (AKL) was measured using a KT-2000TM Knee Arthrometer (MEDmetric® Corp; San Diego, CA), and defined as the amount of anterior tibial displacement relative to the femur when subjected to an anterior-directed force of 133 N. The rationale for measuring AKL was that greater AKL has previously been associated with greater anterior tibial translation during weight acceptance (Shultz, Shimokochi, et al., 2006), and that characteristics of the load-displacement curve (stiffness) of AKL have been shown to influence knee anterior shear forces during double-leg drop-jump landings (Schmitz et al., 2013). Therefore, AKL was collected in order to account for passive restraint characteristics that potentially influence stopjump landing biomechanics. Participants were positioned supine as per the manufacturer's guidelines, with the thigh supported by a bolster placed just proximal to the popliteal fossa, the knees flexed to 25°, and the ankle placed in the manufacturer provided foot cradle (Figure 3.1). A Velcro strap was then placed around the participant's thighs to minimize any rotation of the lower-extremity. Next, the KT-2000TM was attached to the leg in proper alignment with the medial and lateral joint lines of the knee. With the participant in a relaxed state, three anterior-to posterior-directed forces were applied to the anterior aspect of the proximal tibia in order to identify a stable neutral position, followed by the application of an anterior-directed force just over 133 N to measure the anterior tibial displacement in millimeters (mm). A bubble level fixed to the device was used to ensure that an anterior-directed pull is achieved. A total of 3 trials were recorded and subsequently averaged for use in statistical analyses. The measurement of AKL has been shown to have good-to-excellent reliability and precision (Shultz, Nguyen, et al., 2006), and

the primary investigator has previously established good test-retest reliability (ICC_{2,3} = 0.83) and precision (SEM = 0.25 mm) using the methods described.



Figure 3.1. Experimental Set-Up and Participant Positioning for the Anterior Knee Laxity (AKL) Assessment.

Quadriceps and Hamstring Muscle Maximal Voluntary Isometric Contraction

Maximal voluntary isometric contraction (MVIC) of the quadriceps and hamstring musculature was used to normalize sEMG data recorded during the assessment of stop-jump landing biomechanics. In addition, MVIC torque data were recorded so that hamstring MVIC torque data could be used to standardize the applied load (i.e. 30% MVIC torque) during the assessment of K_{HAM} (see Hamstring Musculo-Articular Stiffness Assessment methods below). Prior to sEMG sensor placement, attachment sites were shaved with a disposable hand razor and cleaned with isopropyl alcohol. Sensors were then attached to the medial and lateral quadriceps (i.e. vastus medialis and vastus lateralis, respectively) and hamstring (i.e. semitendinosus/semimembranosus and biceps femoris, respectively) muscle bellies of the left leg, and aligned parallel to the orientation of the muscle fibers. Following confirmation of correct

sEMG sensor placements via manual muscle testing, all sensors were then secured using doublesided adhesive and cohesive athletic tape to minimize movement artifact.

Quadriceps and hamstring MVICs were assessed using a Biodex System 3 dynamometer (Biodex Medical Systems, Shirley, NY). For the quadriceps assessment, participants were positioned supine, with the hip and knee fixed in 30° of flexion. Straps were then secured across the chest, hips, thigh, and distal shank, to ensure a constant body position (Figure 3.2A). Hamstring assessments were carried out in a similar fashion, with the only difference being that participants were positioned prone (Figure 3.2B). All participants performed three submaximal practice trials (25%, 50%, and 75% of maximal isometric effort), and one maximal practice trial (100% maximal isometric effort), prior to performing three maximal test trials from which data were recorded. Each MVIC trial was held for 5 seconds, and 1-minute rest intervals were provided between trials in order to minimize the likelihood of fatigue. Quadriceps trials were performed first and hamstring were performed second.

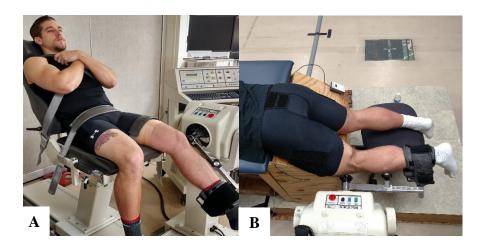


Figure 3.2. Participant Positioning During Maximal Voluntary Isometric Contraction Assessments for the Quadriceps (A) and Hamstrings (B).

Hamstring Musculo-Articular Stiffness

Hamstring musculo-articular stiffness ($K_{\rm HAM}$) was assessed via the free-oscillation technique, whereby the leg is modeled as a single-degree-of-freedom mass-spring system. The damping effect that the hamstring muscles impose on oscillatory flexion-extension at the knee joint is then quantified, following a perturbation (J. Troy Blackburn et al., 2011, 2004; McNair et al., 1992). Prior to the $K_{\rm HAM}$ assessment, a twin axis electrogoniometer (Biometrics Ltd, Ladysmith, VA) was attached to the knee joint in a neutral knee position, in accordance with the manufacturer's guidelines. The telescopic block of the electrogoniometer was placed in parallel to an imaginary line between the head of the fibula and the lateral malleolus, whereas the fixed-end block was placed in parallel to an imaginary line between the greater trochanter and lateral condyle of the femur (Figure 3.3A).



Figure 3.3. Participant Positioning and Instrumentation for the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. Electrogoniometer placement (A); Thermoplastic splint & ankle weights (B); Accelerometer (C); Monitor displaying real-time knee-flexion angle data (D).

Following electrogoniometer attachment, participants were positioned prone, with the trunk and thigh supported in 30° of hip flexion and the shank and foot segment free to move (Figure 3.3). A thermoplastic splint was then secured to the plantar aspect of the foot and

posterior shank in order to standardize ankle position (Figure 3.3B), and a load was attached to the distal shank, at the level of the malleoli, using cuff-style ankle weights (Figure 3.3B). With the thermoplastic splint and ankle weights secured, the participant's shank was then passively positioned so that the knee was in 30° of flexion; the participant was then required to maintain this position via isometric contraction of the hamstring muscles. During this time, real-time knee joint angle data were displayed on a monitor via the electrogoniometer, giving participants a visual target to maintain (Figure 3.3D). Within 5 seconds of the participant holding this position, a brief downward perturbation was then manually applied to the posterior aspect of the calcaneus, resulting in slight knee extension and subsequent damped oscillatory flexion-extension. This damped oscillatory motion was characterized as the tangential acceleration of the shank and foot segment, captured via a triaxial accelerometer (Sensor dimensions: 2.54x2.54x1.91 cm; NeuwGhent Technology, USA) attached to the thermoplastic splint (Figure 3.3C). Participants were verbally instructed not to interfere with or voluntarily produce the oscillations following the perturbation, and attempt to keep the hamstring muscles active only to the level necessary to support the mass of the shank and foot segment, and the applied load, in the testing position (J. Troy Blackburn et al., 2004). This assessment was performed under two different loading conditions. In the first condition, the applied load was assigned as 10% of the participant's body mass (K_{HAM} BM). In the second condition, the applied load was assigned as 30% of the participant's MVIC hamstring torque (K_{HAM} MVIC). In order to avoid a potential order effect, these conditions were assigned in a counterbalanced fashion. Under each loading condition $(K_{\text{HAM}}$ BM and K_{HAM} MVIC), participants performed 3 to 5 practice trials, followed by 5 test trials in which data were recorded and used for analysis. All test trials were separated by 30second rest intervals to reduce the likelihood of fatigue.

Stop-Jump Landing Biomechanics

Landing biomechanics were assessed during double-leg and single-leg stop-jump landing tasks. Prior to performing each of the tasks, all participants were outfitted with standardized footwear (Adidas, Uraha 2, Adidas North America, Portland, OR) in order to experimentally control for the effects of footwear on landing biomechanics. Next, participants were instrumented with six four-marker clusters of optical LED markers (Phase Space, San Leandro, CA) so that three-dimensional kinematic data could be obtained using an eight-camera IMPULSE motion tracking system (Phase Space, San Leandro, CA). Specifically, marker clusters were placed on the posterior thorax (spinous process of the C7 vertebrae) and sacrum (Figure 3.4B), and on the lateral thigh (mid-shaft), medial and lateral tibial flares, lateral shank (mid-shaft), and foot of the left leg (Figure 3.4A). The posterior thorax marker cluster was secured via a thin shoulder harness, whereas the sacral and tibial flare marker clusters were secured directly to the skin using double-sided adhesive tape. Lateral thigh and shank marker clusters were secured to the participant's compression shorts and a thin shank sleeve, respectively, using hook and loop material. Participants were then digitized using MotionMonitor software (Innovative Sports Training, Chicago, IL). Ankle and knee joint centers were determined as the midpoint between the medial and lateral malleoli and medial and lateral femoral epicondyles, respectively. Hip joint center was determined using the Bell method (A. L. Bell, Brand, & Pedersen, 1989).

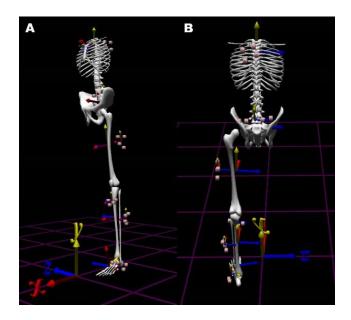


Figure 3.4. LED Marker-Cluster Placement for Three-Dimensional Motion Capture. Marker clusters were placed on the posterior thorax (spinous process of the C7 vertebrae) and sacrum (B), and on the lateral thigh (mid-shaft), medial and lateral tibial flares, lateral shank (mid-shaft), and foot of the left leg (A).

For the double-leg stop-jump task, participants began standing on a starting line, placed at a distance of 40% of their standing height behind the rear edge of two non-conducting force platforms (Type 4060-130; Bertec Corporation., Columbus, OH) (Figure 3.5). Participants were instructed to: 1) perform a double-leg broad jump towards the two force platforms, 2) land evenly with one foot on the center of each platform, 3) jump for maximum vertical height immediately following landing, and 4) land evenly once again with one foot on each platform (Figure 3.5A). The single-leg stop-jump task was performed in identical fashion to the double-leg stop-jump, but only on a single leg (Figure 3.5B). In an effort to prevent any experimenter bias, participants were not provided with any special instructions regarding their stop-jump biomechanics. After performing 3 to 5 practice trials, participants performed 5 successful test trials, during which data were recorded. Thirty-second rest intervals were provided between each test trial to minimize the likelihood of fatigue. A trial was considered successful if the participant: 1) jumped (double-leg)

or hopped (single-leg) from the starting line and landed on the center of the force platform(s), 2) jumped for maximum vertical height immediately after landing, and 3) landed back on the force platform(s) following the vertical jump. Unsuccessful trials were discarded and repeated.



Figure 3.5. Visual Depiction of the Double-Leg (A) and Single-Leg (B) Stop-Jump Landing Tasks.

Data Sampling and Reduction

Quadriceps and Hamstring Muscle Maximal Voluntary Isometric Contraction

Quadriceps and hamstring MVIC torque data were recorded as the mean of the peak torques obtained over the 3 MVIC trials for each muscle group, normalized to the participant's body mass, and reported in Newton-meters per kilogram of body mass (N·m·kg⁻¹). Surface electromyography (sEMG) data were sampled at 1000 Hz and recorded using MotionMonitor software (Innovative Sports Training Inc., Chicago, IL). The sEMG data from each MVIC trial were later rectified and filtered from 10 Hz to 350 Hz within MotionMonitor using a fourth-order, zero-lag, Butterworth filter, and processed using a centered root mean square (RMS) algorithm with a 100-millisecond time constant. These data were then be exported to MATLAB (MathWorks Inc., Natick, MA) for data reduction using a custom-written program. Specifically, the peak RMS sEMG amplitudes for each muscle during each 5-second trial were obtained and

subsequently averaged. The mean peak RMS sEMG amplitudes calculated for each muscle were then be used to normalize all sEMG data recorded during the stop-jump landing tasks.

Hamstring Musculo-Articular Stiffness

Accelerometer data were sampled at 1000 Hz and interfaced with MotionMonitor software for data collection. These data were then be low-pass filtered at 10 Hz using a fourth-order zero-lag Butterworth filter within MotionMonitor, and later exported to MATLAB for data reduction using a custom-written program. Specifically, the time interval between the first two oscillatory peaks (t_1 and t_2) of the accelerometer time-series was used to calculate the damped frequency of oscillation ($1/[t_2-t_1]$) for each trial (Figure 3.6). Hamstring musculo-articular stiffness (K_{HAM}) was then calculated using the equation: $K_{\text{HAM}} = 4\pi \, ^2 mf^2$, where m is the summed mass of the shank and foot segment and the applied load, and f is the damped frequency of oscillation. K_{HAM} values were normalized to body mass ($N \cdot m^{-1} \cdot kg^{-1}$), and the average of 5 trials for each condition (i.e. K_{HAM} _BM and K_{HAM} _MVIC) was then calculated for use in statistical analyses.

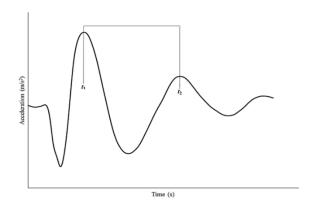


Figure 3.6. Example Accelerometer Time-Series Data Obtained During the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. t_1 and t_2 represent the time points at which the first two oscillatory peaks occur; these time points are then used to calculate the damped frequency of oscillation.

Stop-Jump Landing Biomechanics

Kinetic, kinematic, and sEMG hardware were interfaced with MotionMonitor software for data collection. Additionally, all data were time-synchronized via an analog syncing pulse that was manually triggered during each trial of the double- and single-leg stop-jump landing tasks. Kinetic and sEMG data were sampled at 1000 Hz, whereas kinematic data were sampled at 240 Hz and subsequently linearly interpolated to 1000 Hz within MotionMonitor. For kinematic data, a segmental reference system was defined for all body segments with the z-axis as the medial-lateral axis (flexion/extension), the y-axis as the distal-proximal longitudinal axis (internal/external rotation), and the z-axis as the anterior-posterior axis (abduction/adduction). Joint motions were then calculated within MotionMonitor using Euler angle definitions with a rotational sequence of Z Y' X" (Kadaba, Ranakrishnan, Wootten, Gainey, & Cochran, 1989). Regardless of joint, all flexions, internal rotations, and adductions were defined as positive values. Intersegmental kinetic data were calculated within MotionMonitor using inverse dynamics (Gagnon & Gagnon, 1992). All data were then exported to MATLAB to be filtered, normalized, and reduced using a custom-written program.

Within MATLAB, kinetic and kinematic data were low-pass filtered at 12 Hz using a fourth-order zero-lag Butterworth filter, whereas ground reaction force data were low-pass filtered at 60 Hz. Joint moments were then normalized to the product of each participant's body weight and body height (Nm·BW⁻¹·Ht⁻¹), and ground reaction forces were normalized to each participant's body weight (%BW). Surface EMG (sEMG) data were filtered from 10 Hz to 350 Hz using a fourth-order zero-lag Butterworth filter and then processed using a centered RMS algorithm with a 25-millisecond time constant. These sEMG data were then normalized to the mean peak RMS sEMG data obtained for each muscle during the quadriceps and hamstring MVIC assessments (%MVIC); composite averages were then calculated for the medial and lateral

quadriceps and medial and lateral hamstring muscles in order to represent neuromuscular activation for the quadriceps and hamstring muscle groups, respectively.

Following data filtering and normalization, all neuromuscular and biomechanical variables of interest were then extracted from each of the landing tasks. Specifically, neuromuscular variables of interest included hamstring and quadriceps muscle pre-activation (HAM_{PRE} and QUAD_{PRE}, respectively), which were defined as the normalized mean RMS sEMG amplitude obtained over a 150-millisecond time interval prior to initial ground contact (IC; instant at which vertical ground reaction force first exceeded 10 N). Kinematic variables of interest included trunk center of mass position (TrunkCOM_{IC}) and hip and knee flexion angles at IC (HF_{IC} and KF_{IC}, respectively), hip and knee flexion excursion angles (HF_{EXC} and KF_{EXC}), knee flexion angle at the instant of peak posterior ground reaction force (KF_{PKpGRF}), average hip and knee flexion velocities (HFV and KFV) across the landing phase [i.e. interval of time from IC to the instant at which the body's center of mass reached its lowest point (maximal descent)], peak anterior tibial translation (ATT), and peak anterior tibial acceleration (ATA). Joint excursions were calculated by subtracting the angle obtained at IC from the peak angle obtained during the landing phase of each jump. Anterior tibial translation (ATT) was calculated as the maximum anterior displacement (mm) of the proximal tibia (tibial flare marker-cluster) relative to the femur (lateral thigh marker-cluster). Anterior tibial acceleration (ATA) was obtained by calculating the second derivative of ATT (m/s⁻²). Kinetic variables of interest included peak posterior and vertical ground reaction forces (pGRF_{PEAK} and vGRF_{PEAK}), peak proximal tibia anterior shear force (PTASF), peak knee extensor moment (KEM_{PEAK}), and knee extensor moment at the instant of pGRF_{PEAK} (KEM_{PKpGRF}). The average of 5 trials were then calculated for each stop-jump landing task (single-leg and double-leg) and used for analysis.

Statistical Approach

All statistical analyses were performed in SPSS (version 23; IBM Corp, Armonk, NY), and R (version 3.3.2; R Foundation for Statistical Computing, Vienna, Austria) statistical software. An *a priori* alpha level of 0.05 (α = 0.05) was used to denote statistical significance. The statistical analyses used to test each of the research hypotheses are detailed below.

Hypotheses 1a and 1b

Hypothesis 1a stated: Compared to the double-leg stop-jump, the single-leg stop-jump will elicit a more upright landing posture (as evidenced by a more posteriorly-oriented trunk center of mass position and less hip and knee flexion at initial ground contact), slower hip and knee average angular velocities, smaller hip and knee flexion excursions, larger posterior and vertical ground reaction forces and knee extensor moments, greater preparatory muscle activation, and biomechanical factors indicative of greater ACL loading (greater peak proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation).

Hypothesis 1b stated: During each stop-jump landing task, females will display a more upright landing posture (as evidenced by a more posteriorly-oriented trunk center of mass position and less hip and knee flexion at initial ground contact), slower hip and knee average angular velocities, smaller hip and knee flexion excursions, larger posterior and vertical ground reaction forces and knee extensor moments, greater preparatory muscle activation, and biomechanical factors indicative of greater ACL loading (greater peak proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation), compared to males.

To test these research hypotheses, a total of four separate mixed-model multivariate analyses of variance (MANOVA) will be conducted. In each MAONVA model, landing type (single-leg landing/double-leg landing) will be the within-subjects factor and sex (male/female)

will be the between-subjects factor. The dependent variables for each model will then come from one of the following variable groups: 1) kinematics at initial contact (five variables), 2) kinematics and kinetics at landing (five variables), 3) preparatory muscle activation (two variables), and 4) biomechanical factors indicative of ACL loading (three variables). The kinematics at initial contact variables will include center of mass position and hip and knee flexion angles at initial ground contact (COM_{IC}, HF_{IC}, and KF_{IC}), and hip and knee flexion angular velocities at initial ground contact (HFV_{IC} and KFV_{IC}). The kinematics and kinetics at landing variables will include hip and knee flexion excursion angles (HF_{EXC} and KF_{EXC}), peak internal knee extensor moment (KEM_{PEAK}), and peak posterior and vertical ground reaction forces (pGRF_{PEAK} and vGRF_{PEAK}). The preparatory muscle activation variables will include quadriceps and hamstring preparatory muscle activation (QUAD_{PRE} and HAM_{PRE}). The biomechanical factors indicative of ACL loading variables will include peak proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation (PTASF, ATA, ATT). To answer hypothesis 1a, the main effect for landing type will be examined using Wilks' Lambda (Λ). If the main effect for landing type is statistically significant, dependent t-tests with a Bonferroni correction will be used to determine pairwise differences. To answer hypothesis 1b, the main effect for sex will be examined using Wilks' A. If the main effect for sex is statistically significant, independent t-tests with a Bonferroni correction will be used to determine pairwise differences within each landing task.

Hypotheses 2a and 2b

Hypothesis 2a is that after controlling for body positioning at initial ground contact (i.e. center of mass position and hip and knee flexion angles), higher K_{HAM} values will be predictive of biomechanical characteristics indicative of lower sagittal plane ACL loading during landing (i.e.

less proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation) within each sex. Hypothesis 2b is that the extent to which K_{HAM} predicts biomechanical factors indicative of sagittal plane ACL loading will be dependent on the method by which K_{HAM} is measured (i.e. 10% of body mass versus 45% maximal voluntary isometric torque). Specifically, it is hypothesized that K_{HAM} will be more predictive of biomechanical factors indicative of sagittal plane ACL loading when K_{HAM} is assessed using the 45% maximal voluntary isometric torque method compared to the 10% body mass method.

To test these research hypotheses, a total of three separate hierarchical multiple linear regression analyses will be conducted for each sex (i.e. separate models for males and females). For each model, the criterion variable will be one of the three biomechanical factors indicative of ACL loading (i.e. PTASF, ATT, or ATA). To statistically control for body positioning at initial ground contact, COM_{IC}, HF_{IC}, and KF_{IC} will be added as predictor variables in the first block using the ENTER method; and both K_{HAM} measures (i.e. K_{HAM} _BM and K_{HAM} _MVIC) will then be added as predictor variables in the second block using the ENTER method. The weaker predictor will then be removed after examining the strength of the standardized regression coefficients of both K_{HAM} measures in the same model, leaving only one measure of K_{HAM} in the final model. The statistical significance criterion will be set at $\alpha = 0.05$. Within each sex, the initial regression models that will be tested are as follows:

PTASF = CONSTANT + β 1(COMIC) + β 2(HFIC) + β 3(KFIC) + β 4(KHAM_BM) + β 5(KHAM_MVIC) ATT = CONSTANT + β 1(COMIC) + β 2(HFIC) + β 3(KFIC) + β 4(KHAM_BM) + β 5(KHAM_MVIC) ATA = CONSTANT + β 1(COMIC) + β 2(HFIC) + β 3(KFIC) + β 4(KHAM_BM) + β 5(KHAM_MVIC)

Hypotheses 3a and 3b

Hypothesis 3a is that the linear combination of peak posterior ground reaction force, knee extensor moment, knee flexion angle, preparatory quadriceps muscle activation, and sex, will be highly predictive of biomechanical factors indicative of sagittal plane ACL loading. This hypothesis is based on the previous work of Sell et al (Sell et al., 2007), who demonstrated greater preparatory quadriceps muscle activation (QUAD_{PRE}), peak posterior ground reaction force, external knee flexion moment, and knee flexion angle, and sex (being female), significantly predicted greater proximal tibia anterior shear force during a double-leg stop jump landing task. Hypothesis 3b is that K_{HAM} will be a significant independent predictor in the final regression model when added to the pool of possible predictors, with higher K_{HAM} being predictive of biomechanical characteristics indicative of lower sagittal plane ACL loading (i.e. less proximal tibia anterior shear force, anterior tibial acceleration, and anterior tibial translation).

To test these research hypotheses, two separate stepwise multiple linear regression analyses will be conducted. In the first regression model, the predictor variables will include quadriceps muscle pre-activation (QUAD_{PRE}), peak posterior ground reaction force (pGRF_{PEAK}), knee extension moment at peak posterior ground reaction force (KEM_{PKpGRF}), knee flexion angle at peak posterior ground reaction force (KF_{PKpGRF}), and sex (male/female), and proximal tibia anterior shear force (PTASF) will be included as the criterion variable. The second regression model will be identical to the first, with the only exception being that hamstring musculo-articular stiffness (K_{HAM}) will be included in the pool of potential predictors. In both models, the statistical significance criterion will be set at $\alpha = 0.05$.

Power Analysis

An a priori power analysis was performed using G*Power (G*Power, Version 3.1.9.2) in order to determine the sample size needed to test the hypothesis that, after controlling for body positioning at initial ground contact (i.e. COM_{IC} , HF_{IC} , and KF_{IC}), higher K_{HAM} values will be predictive of biomechanical characteristics indicative of lower sagittal plane ACL loading during landing. This power analysis indicated that, with 4 predictor variables (number of tested predictors = 1; total number of predictors = 4), 40 participants per sex (80 participants total) would be needed to achieve statistical power between 0.73 and 0.94 and to detect a medium $(R^2\Delta)$ = 0.15; f^2 = 0.18) to large effect size ($R^2\Delta = 0.25$; f^2 = 0.33) at a statistical significance criterion of $\alpha = 0.05$. The effect sizes used for this power analysis were conservatively estimated based on the theory that there would need to be at least a medium effect size for K_{HAM} to be considered a clinically meaningful factor for ACL loading. Secondary power analyses were also performed in order to determine the statistical power that would be achieved for the hypotheses associated with aims 1 and 3 given a total sample size of 80 participants. For the MANOVA models, the power analysis indicated that with two groups and a total sample size of 80 participants, statistical power could be expected to range between 0.61 and 0.99 given a medium to large effect size and a statistical significance criterion of $\alpha = 0.05$. Similarly, for the stepwise multiple linear regression model with the potential for 6 total predictors in aim 3, the power analysis indicated that statistical power could be expected to range between 0.69 and 0.98 given a medium to large effect size and a statistical significance criterion of $\alpha = 0.05$.

CHAPTER IV

MANUSCRIPT I

Title

The Effects of Task and Sex on Neuromuscular and Biomechanical Characteristics during Double-Leg and Single-Leg Stop-Vertical Jumps

Abstract

Background: Between-sex differences in neuromuscular and biomechanical characteristics may help explain females' increased risk for noncontact anterior cruciate ligament (ACL) injury. Although these injuries more commonly occur when landing on a single leg, many of these between-sex differences have been established using double-leg landing tasks as injury models.

Purpose: To compare the neuromuscular and biomechanical demands of a double-leg stop-jump (DLSJ) to that of a single-leg stop-jump (SLSJ) in males and females.

Study Design: Descriptive laboratory study.

Methods: Sixty-eight males and females (Males = 34, 21.5 ± 2.0 years, 1.8 ± 0.01 m, 81.4 ± 10.98 kg; Females = 32, 21.1 ± 2.0 years, 1.7 ± 0.01 m; 63.0 ± 9.1 kg) performed the DLSJ and SLSJ, during which neuromuscular and biomechanical data were recorded. Passive anterior knee laxity (AKL) was also measured. Mixed-model multivariate analyses of covariance were used to examine the effects of task and sex, and their interaction, on these characteristics after adjusting for the effects of AKL.

Results: Compared to the DLSJ, participants performed the SLSJ with a more posterior trunk center-of-mass position (P < .001) and smaller knee-flexion angles (P < .001) at initial ground contact, less knee-flexion excursion (P = .038), greater ground reaction forces (P < .001), knee-extension moments (P = .033), and proximal tibia anterior shear forces (P < .001), and less anterior tibial translation (P = .007). Additionally, compared to the DLSJ, females perform the SLSJ with a greater reduction in hip-flexion velocity (P < .001) and a smaller increase in hip-extension moment (P < .001) than males. With this, compared to the DLSJ, participants with greater amounts of AKL performed the SLSJ with a greater increase in anterior shear force than participants with lesser AKL (P < .001). Further, irrespective of task, females displayed smaller knee-flexion angles at initial contact (P = .047), less hip-flexion excursion (P = .006), slower hip-flexion velocities (P = .040), smaller hip-extension moments (P < .001), and greater anterior tibial translation (P = .006), compared to males.

Conclusion: Compared to the DLSJ, the SLSJ elicited characteristics associated with increased ligamentous loading and a landing posture that was more representative of what has been observed during injurious situations. While females displayed more "risky" biomechanics than males during both tasks, females displayed different biomechanical "strategies" at the hip compared to males during the SLSJ, which suggests that the demands placed on the body during the SLSJ were likely greater for our female participants.

Clinical Relevance: The SLSJ appears to place increased demands on lower-extremity; however, the way in which individuals respond to these increased demands differs for males and females. Thus, the use of sex-stratified research designs, and single-leg tasks as injury models, in future work appears warranted. Additionally, current injury prevention efforts should focus on incorporating more single-leg jumping and landing activities while promoting softer (more flexed) landing styles.

Key Terms: anterior cruciate ligament; biomechanics; electromyography; shear force;

Introduction

sex

Female athletes are at substantially greater risk for experiencing a noncontact anterior cruciate ligament (ACL) injury compared to their male counterparts (Agel, Arendt, & Bershadsky, 2005; Arendt, E. A., Agel, & Dick, 1999). Because a large proportion of these injuries occur when landing from a jump (Boden et al., 2009; Krosshaug et al., 2007; Olsen et al., 2004), several laboratory-based studies have attempted to identify the factors that likely contribute to this sex bias in injury risk by examining between-sex differences in neuromuscular and biomechanical characteristics during a variety of jump-landing tasks. Collectively, this body of work generally demonstrates that females display greater preparatory activation of the thigh musculature, a more upright body position (i.e. smaller trunk-, hip-, and knee-flexion angles) at initial ground contact (IC), lesser sagittal-plane joint excursions, slower angular velocities, greater ground reaction forces, greater knee-flexion and knee-abduction moments (external), and greater proximal-tibia-anterior-shear force, compared to males (Chappell, Creighton, Giuliani, Yu, & Garrett, 2007; Chappell et al., 2002; Decker, Torry, Wyland, Sterett, & Richard Steadman, 2003; Kernozek, Torry, H, Cowley, & Tanner, 2005; Pappas et al., 2007; Schmitz et al., 2007; Shultz, S. J., Nguyen, Leonard, & Schmitz, 2009; Weinhandl, Irmischer, & Sievert, 2015; Weinhandl, Joshi, & O'Connor, 2010a; Yu et al., 2006). As many of these female landing characteristics have been associated with increased ACL loading both in-vivo (Beynnon et al., 1995; Fleming, Renstrom, Beynnon, et al., 2001) and *in-vitro* (DeMorat et al., 2004; Fujiya et al., 2011; Li et al., 1999; Markolf et al., 1995; Torzilli et al., 1994; Withrow et al., 2006), current biomechanical theory suggests that females are at increased risk for injury because they land in a manner that

exposes the knee to greater amounts of ligamentous loading. As such, current injury prevention programs largely focus on modifying the "risky" landing mechanics displayed by females to be more in line with that of their male counterparts in an effort to reduce females' risk for injury.

Several injury prevention programs have been shown to effectively modify neuromuscular and biomechanical characteristics in a manner that is thought to result in reduced ACL loading (Chappell & Limpisvasti, 2008; Greska, Cortes, Van Lunen, & Oñate, 2011; Hewett, Stroupe, Nance, & Noyes, 1996; Padua & Distefano, 2009). Additionally, females who complete such programs are demonstrated to be at reduced risk for injury compared to their untrained female counterparts (Emery & Meeuwisse, 2010; LaBella et al., 2011; Walden, Atroshi, Magnusson, Wagner, & Hagglund, 2012). Despite this success however, noncontact ACL injuries continue to occur at a relatively high rate, and the associated sex bias remains (Prodromos et al., 2007), indicating that the most appropriate risk factors to target through current injury prevention efforts have not yet been identified. With this, not all studies of between-sex differences agree that females display neuromuscular and biomechanical differences compared to males that would potentially result in greater ACL loading and injury risk. For example, some studies have argued that females actually display no differences in preparatory neuromuscular activation (Fagenbaum & Darling, 2003), body positioning at IC (Kernozek et al., 2005), ground reaction forces (Decker et al., 2003), or knee-flexion moments (Decker et al., 2003), compared to males, whereas others have found females to display greater knee-flexion angles at IC (Fagenbaum & Darling, 2003), faster knee-flexion velocities (Decker et al., 2003), and greater sagittal-plane joint excursions (Decker et al., 2003; Kernozek et al., 2005; Shultz, S. J. et al., 2009). An important aspect of this prior work is that not all studies of between-sex differences have used the same jump-landing task as a model for injury. Specifically, three studies used a double-leg drop-landing (Decker et al., 2003; Kernozek et al., 2005; Weinhandl et al., 2015), three used a single-leg drop-landing

(Fagenbaum & Darling, 2003; Schmitz et al., 2007), two used a double-leg drop-vertical jump (Salci, Kentel, Heycan, Akin, & Korkusuz, 2004; Shultz, S. J. et al., 2009), three used a double-leg stop-vertical jump (Chappell et al., 2007; Chappell et al., 2002; Yu et al., 2006), and two used both double- and single-leg drop-landings within a single investigation (Pappas et al., 2007; Weinhandl, Joshi, & O'Connor, 2010b). This distinction between tasks is important because different tasks have been shown to affect neuromuscular and biomechanical outcome measures (Cruz et al., 2013), and thus may be hindering researchers' ability to clearly identify the most appropriate factors to be targeted through current injury prevention strategies.

The reason that different tasks have the ability affect neuromuscular and biomechanical outcomes is that different tasks alter the demands placed on the lower extremity. For example, Cruz et al (2013) compared biomechanical differences between a double-leg drop-landing, dropvertical jump, and forward-vertical jump, beginning atop a 30-cm box. The drop-landing involved dropping from the box and landing. The drop-vertical jump was similar to the drop-landing, but involved a subsequent maximal vertical jump upon landing; and the forward-vertical jump was similar to the drop-vertical jump, but involved an initial forward approach jump from a distance equal to 50% body height. It was found that the forward-vertical jump elicited greater trunk- and hip-flexion angles, greater ground-reaction and proximal-tibia-anterior-shear forces, and greater sagittal- and frontal-plane knee moments, compared to both the drop-landing and drop-vertical jump; and that both the forward-vertical jump and drop-vertical jump tasks elicited greater knee flexion compared to the drop-landing (Cruz et al., 2013). The authors then concluded that these differences were likely owed to the forward-vertical jump being a more biomechanically demanding task due to the increased energy requirements needed to both successfully absorb greater impact forces and complete the subsequent vertical jump upon landing (Cruz et al., 2013). While tasks that include both horizontal- and vertical-deceleration components, and a subsequent

movement upon landing, may elicit demands that are more in line with the demands of the highly-dynamic maneuvers associated with noncontact ACL injury, it is well known that this type of injury is far more likely to occur when landing on a single leg (Boden et al., 2000; Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004). To this end, there is emerging evidence that double-leg tasks elicit different neuromuscular and biomechanical characteristics than those observed when performing the same task on a single leg (Pappas et al., 2007; Taylor, Ford, Nguyen, & Shultz, 2016; Wang, L. I., 2011; Weinhandl et al., 2010b; Yeow et al., 2010). Hence, the sex-specific landing mechanics that potentially contribute to noncontact ACL injuries resulting from a single-leg landing maneuver may not be fully represented when using a double-leg task as a model for injury.

While studies of between-sex differences during functional athletic tasks are abundant in the literature, fewer studies have investigated differences between double-leg and single-leg tasks (Pappas et al., 2007; Taylor et al., 2016; Wang, L. I., 2011; Weinhandl et al., 2010b; Yeow et al., 2010), and we are only aware of two studies having investigated sex differences both within and between tasks (Pappas et al., 2007; Weinhandl et al., 2010b). When comparing double- and single-leg drop-landings, it has been shown that landing on a single leg elicits smaller hip- and knee-flexion angles at IC (Pappas et al., 2007; Weinhandl et al., 2010b; Yeow et al., 2010), smaller peak knee-flexion angles (Pappas et al., 2007) and hip- and knee-flexion excursions (Weinhandl et al., 2010b; Yeow et al., 2010), greater knee-flexion and knee-abduction moments, greater ground-reaction forces (Weinhandl et al., 2010b; Yeow et al., 2010), and greater quadriceps and hamstring neuromuscular activation (Pappas et al., 2007), suggesting that landing on a single-leg is more "risky" in terms of ACL injury due a decreased base of support and an increased demand on the lower-extremity musculature. This is also true of double- and single-leg stop-vertical jumps (DJSJ and SLSJ, respectively), with the SLSJ being shown to elicit smaller

initial and peak hip- and knee-flexion angles and slower angular velocities, greater external kneeflexion and valgus moments (Taylor et al., 2016; Wang, L. I., 2011), and greater ground-reaction and proximal-tibia-anterior-shear forces (Wang, L. I., 2011). Given that (a) double-leg tasks that include horizontal- and vertical-deceleration components, and a subsequent movement upon landing, are shown to be more demanding than double-leg landings, (b) that demand increases even further when performing the same task on a single-leg, and (c) that noncontact ACL injuries are more likely to result when landing on a single leg, it seems that researchers interested in identifying sex-specific landing mechanics would want to use an injury model which includes all of these components so that the demands of the task are more representative of situations in which noncontact ACL injuries occur. However, the only previous studies to compare sex differences both within and between tasks have used double- and single-leg drop-landings as injury models (Pappas et al., 2007; Weinhandl et al., 2010b), and we are currently unaware of any studies that have examined such differences using tasks that include a horizontal deceleration component, such as the DLSJ and SLSJ. Identifying between-sex differences during DLSJ and SLSJ tasks, and whether males and females display different neuromuscular and biomechanical strategies when transitioning from the DLSJ to the SLSJ, may help further elucidate sex-specific landing mechanics that potentially place females at increased risk for noncontact ACL injury.

The purpose of this study was to compare the neuromuscular and biomechanical demands of the DLSJ and SLSJ in males and females. It was hypothesized that the SLSJ would elicit a landing style considered to be more "risky" in terms of ACL loading and noncontact injury (i.e. greater preparatory neuromuscular activation, a more upright body position at IC, lesser sagittal-plane joint excursions and slower angular velocities, greater ground reaction forces and resultant joint moments, and greater proximal tibia anterior shear force, anterior tibial translation, and

anterior tibial acceleration) compared to the DLSJ, and that these characteristics would be more pronounced in females.

Methods

Participants

Thirty-four males (age = 21.47 ± 2.02 years, height = 1.82 ± 0.06 m; mass = 81.35 ± 10.97 kg) and 32 females (age = 21.09 ± 1.96 years, height = 1.66 ± 0.08 m; mass = 63.02 ± 9.11 kg) volunteered to participate in this study. All participants were considered physically active – defined as engaging in greater than the equivalent of 300 minutes of moderate-intensity physical activity per week (assessed via the International Physical Activity Questionnaire-Short Form) – at the time of enrollment, and regularly participated in activities that involved running, cutting, jumping and landing. In addition, participants were without any history of knee ligamentous or meniscal injury, lower-extremity surgery, lower-extremity injury in the 6 months prior to recruitment, or any known medical conditions affecting their connective tissue or vestibular system. This study was approved by the university's Institutional Review Board for the Protection of Human Subjects prior to recruitment, and written informed consent was obtained from each participant prior to testing. Each participant received \$10 compensation for their participation in this study.

Procedures

All data were collected during a single testing session in a controlled biomechanics laboratory setting. To control for the potential effects of menstrual cycle hormones on knee-joint biomechanics (Park, Stefanyshyn, Ramage, Hart, & Ronsky, 2009; Shultz, S. J. et al., 2012; Shultz, S. J. et al., 2011), female testing was constrained to the follicular phase of their menstrual

cycle (day 1-8 following self-reported menstrual bleeding onset). Upon arrival, participants were outfitted with compression shorts and a tight-fitting athletic top. Barefoot measures of body height and mass were then obtained, and the remainder of the testing session was carried out in the following order: (1) anterior knee-joint laxity assessment, (2) surface electromyography instrumentation, (3) five-minute warm-up, (4) maximal voluntary isometric contraction testing, and (5) stop-vertical jump landing biomechanics. The warm-up was performed on a stationary cycle ergometer (Life Fitness, Schiller Park, IL) at a cadence of 70-80 RPM and a target rating of perceived exertion of \geq 3-4 on a Borg CR-10 RPE scale (Borg, 1998). All data were obtained from the left lower extremity, which corresponded with the dominant limb (defined a-priori as the stance limb when participants were asked which limb they would use to kick a ball for maximum horizontal distance) in 58 of our 66 participants (88%).

Anterior Knee Laxity. Anterior knee laxity (AKL) was measured using a KT-2000TM Knee Arthrometer (MEDmetric® Corp; San Diego, CA, USA), and defined as the amount of anterior tibial displacement, relative to the femur, when subjected to an anterior-directed force of 133 N. Our rationale for measuring AKL was that greater AKL has previously been associated with greater anterior tibial translation during weight acceptance (Shultz, S. J., Shimokochi, et al., 2006), and characteristics of the AKL load-displacement curve (stiffness) have been shown to influence proximal-tibia-anterior-shear forces during double-leg drop-vertical jumps (Schmitz et al., 2013), and greater knee joint stiffness and extensor loading during landing (Shultz, S. J., Schmitz, Nguyen, & Levine, 2010a). Therefore, because females typically have more AKL than males, AKL was collected for use as a covariate in our statistical analyses to account for a passive restraint characteristic that potentially influences landing biomechanics.

Participants were positioned as per the manufacturer's guidelines, with the thigh supported by a bolster placed just proximal to the popliteal fossa, the knees flexed to 25°, the

ankle placed in the manufacturer-provided foot cradle, and a strap secured around the thighs to minimize lower-extremity rotation (Figure 4.1). The KT-2000TM was then attached to the leg in alignment with the medial and lateral joint lines of the knee. With the participant in a relaxed state, three anterior- to posterior-directed forces were applied to the anterior aspect of the proximal tibia in order to identify a stable neutral joint position. Next, an anterior-directed force of 133 N was applied to the proximal tibia, and anterior tibial displacement was measured to the nearest half-millimeter. A bubble level fixed to the device was used to ensure that an anterior-directed pull was achieved. A total of 3 trials were recorded and subsequently averaged for use in statistical analyses. The measurement of AKL has been shown to have good-to-excellent reliability and precision (Shultz, S. J., Nguyen, et al., 2006), and the primary investigator (JPW) has previously established good test-retest reliability (ICC2,3 = 0.83) and precision (SEM = 0.25 mm) using the methods described.



Figure 4.1. Experimental Set-Up and Participant Positioning for the Anterior Knee Laxity (AKL) Assessment.

Surface Electromyography Instrumentation. A wireless surface electromyography (sEMG) system (Delsys Trigno; Delsys Inc., Boston, MA, USA) was used to assess preparatory neuromuscular activation (150 milliseconds prior to initial ground contact) of the quadriceps and

hamstring muscles during the stop-vertical jump tasks. Prior to sensor placement, attachment sites were shaved and cleaned using a disposable hand-razor and isopropyl alcohol, respectively. Sensors were then placed over the muscle bellies of medial and lateral quadriceps (vastus medialis and lateralis, respectively) and hamstring muscles (semitendinosus/semimembranosus and biceps femoris long-head, respectively) using double-sided adhesive tape. Once sensor placements were confirmed via manual muscle testing, cohesive athletic tape was wrapped around the participant's thigh to ensure sensor placement and minimize movement artifact.

Maximal Voluntary Isometric Contraction Testing. Maximal voluntary isometric contraction (MVIC) testing was performed for the quadriceps and hamstring muscle groups using a Biodex System 3 isokinetic dynamometer (Biodex Medical Systems, Shirley, NY, USA). Quadriceps MVIC testing was performed with participants positioned supine and the hip and knee fixed in 30° of flexion (Figure 4.2A). Hamstring MVIC testing was performed with participants positioned prone and the hip and knee fixed in 30° of flexion (Figure 4.2B). For each test, straps were secured across the torso, hips, thigh, and distal shank to ensure a constant body position. Prior to completing 3 maximal effort test trials, participants performed 4 practice trials at 25%, 50%, 75%, and 100% of self-perceived maximal effort. Practice and test trials were held for 5 seconds, separated by 60-second rest-intervals, and verbal encouragement was provided by the investigators to ensure consistency across trials. The sEMG data recorded during each test trial were then used for normalization purposes (see Data Sampling and Reduction).

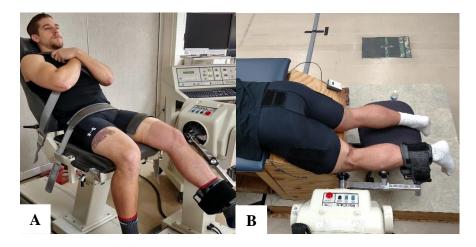


Figure 4.2. Participant Positioning During Maximal Voluntary Isometric Contraction Assessments for the Quadriceps (A) and Hamstrings (B).

Stop-Vertical Jump Tasks. Preparatory neuromuscular activation and sagittal-plane landing biomechanics were assessed during double-leg and single-leg stop-vertical jump tasks (DLSJ and SLSJ, respectively) using an 8-camera IMPULSE motion tracking system (Phase Space, San Leandro, CA) and two non-conducting force platforms (Type 4060-130; Bertec Corporation., Columbus, OH, USA). All participants were standardized athletic shoes (adidas, Uraha 2, adidas North America, Portland, OR, USA) in order to experimentally control for the effects footwear on landing biomechanics. Participants were then instrumented with optical LED marker clusters (4 markers per cluster; Phase Space, San Leandro, CA, USA) secured to the foot, shank, thigh, pelvis, and trunk (Figure 4.3). Once instrumented, participants were digitized using MotionMonitor software (Innovative Sports Training, Chicago, IL). Ankle and knee joint centers were determined as the midpoint between the medial and lateral malleoli and medial and lateral femoral epicondyles, respectively. Hip joint centers were determined using the Bell method (Bell, A. L., Brand, & Pedersen, 1989).

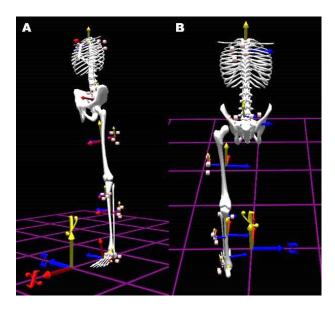


Figure 4.3. LED Marker-Cluster Placement for Three-Dimensional Motion Capture. Marker clusters were placed on the posterior thorax (spinous process of the C7 vertebrae) and sacrum (B), and on the lateral thigh (mid-shaft), medial and lateral tibial flares, lateral shank (mid-shaft), and foot of the left leg (A).

For the DLSJ and SLSJ tasks, a line was placed at a distance equal to 40% of the participant's body height behind the rear edge of the force platforms. This line was used as a starting position from which participants initiated each DLSJ and SLSJ trial. For the DLSJ, participants were verbally instructed to perform a double-leg broad jump towards the force platforms, land evenly with one foot on each platform, jump for maximum vertical height upon landing, and then land evenly once again with one foot on each platform (Figure 4.4A). The single-leg stop-jump was performed in identical fashion to the double-leg stop-jump, but using only the left leg (Figure 4.4B). In an effort to prevent any experimenter bias, the investigators did not provide participants with any special instructions or feedback regarding their landing biomechanics. For each task, participants performed 3 to 5 practice trials prior to performing 5 successful test trials during which data were recorded. Thirty-second rest intervals were provided between trials to minimize the likelihood of fatigue. Trials were considered to be successful if the

participant: (1) jumped (double-leg) or hopped (single-leg) from the starting line and landed on the force platform(s), (2) jumped for maximum vertical height upon initial landing, and (3) landed back on the force platform(s) following the vertical jump. Unsuccessful trials were discarded and repeated.

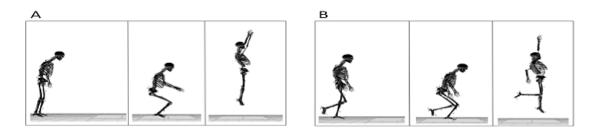


Figure 4.4. Visual Depiction of the Double-Leg (A) and Single-Leg (B) Stop-Jump Landing Tasks.

Data Sampling and Reduction

All kinetic, sEMG, and kinematic hardware were integrated and time-synchronized with MotionMonitor software (Innovative Sports Training Inc., Chicago, IL) for data collection. Kinetic and sEMG data were sampled at 1000 Hz, whereas kinematic data were sampled at 240 Hz and subsequently linearly interpolated to 1000 Hz. Quadriceps and hamstring sEMG data obtained during MVIC testing and stop-vertical jump landing tasks were band-pass filtered from 10 Hz to 350 Hz using a fourth-order zero-lag Butterworth filter, and subsequently processed using centered root mean square (RMS) algorithms with 25- and 100-millisecond time constants, respectively. Kinetic and kinematic data were low-pass filtered at 12 Hz using a fourth-order zero-lag Butterworth filter, whereas peak ground reaction force data were low-pass filtered at 60 Hz. A segmental reference system was defined for all body segments, with the z-axis as the medial-lateral axis (flexion-extension), the y-axis as the longitudinal axis (internal-external rotation), and the x-axis as the anterior-posterior axis (abduction-adduction). Joint motions were

then calculated using Euler angle definitions with a rotational sequence of Z Y' X" (Kadaba et al., 1989). Trunk center-of-mass (CoM) position was defined as the anterior(+)-posterior(-) displacement (cm) of the trunk's CoM relative to the center of pressure. Anterior tibial translation was defined as the anterior displacement (mm) of the proximal tibia marker cluster relative to the lateral thigh marker cluster (Figure 3). Anterior tibial acceleration was derived by calculating the second derivative of anterior tibial translation. Joint moments and proximal-tibia-anterior-shear force were calculated using inverse dynamics (Gagnon & Gagnon, 1992). All data were later exported from MotionMonitor to be reduced in MATLAB (Mathworks, Inc., Natick, MA, USA) using a custom-written program.

Within MATLAB, the RMS sEMG data recorded from each muscle during each stopjump trial were normalized to the mean peak RMS sEMG amplitude recorded from each
respective muscle during MVIC testing. Composite averages were then calculated for the medial
and lateral quadriceps, and medial and lateral hamstrings, and used to represent normalized
neuromuscular activation (%MVIC) of the quadriceps and hamstring muscle groups, respectively.
All joint moment data were normalized to the product of each participant's body weight and body
height (BW⁻¹·Ht⁻¹), and all force data were normalized to body weight (BW). Following data
filtering and normalization, all neuromuscular and biomechanical variables of interest were then
extracted. Specifically, neuromuscular variables of interest included hamstring and quadriceps
muscle pre-activation (HAM_{PRE} and QUAD_{PRE}, respectively), which were defined as the
normalized mean RMS sEMG amplitude obtained over a 150-millisecond time interval prior to
initial ground contact (IC; instant at which vertical ground reaction force first exceeded 10 N).
Biomechanical variables of interest included: trunk CoM position and hip- and knee-flexion
angles at IC (TrunkCoM_{IC}, HF_{IC}, and KF_{IC}, respectively); trunk CoM, and hip- and knee-flexion
excursions (TrunkCoM_{EXC}, HF_{EXC}, and KF_{EXC}, respectively); peak posterior and vertical ground

reaction forces (vGRF_{PK} and pGRF_{PK}, respectively), hip- and knee-extension moments (HEM_{PK} and KEM_{PK}), proximal tibia anterior shear force (PTASF_{PK}), anterior tibial acceleration (ATA_{PK}), and anterior tibial translation (ATT_{PK}), throughout the landing phase; and average hip- and knee-flexion angular velocities throughout the landing phase (HFV and KFV, respectively). Excursions were calculated by subtracting IC values from peak values. The landing phase was defined as the interval of time from IC to maximal descent (i.e. the point in time at which the CoM reached its lowest vertical position during landing). Means of all 5 DLSJ and SLSJ trials were then calculated for use in statistical analyses.

Statistical Analyses

Four separate mixed-model analyses of covariance (MANCOVAs) were conducted using SPSS, version 23 (IBM Corp., Armonk, NY, USA), with an *a-priori* alpha level set at 0.05 (α = 0.05) to denote statistical significance. In each MANCOVA model, sex was used as the between-subjects factor (two levels: male or female), task was used as the within-subjects factor (two levels: DLSJ or SLSJ), and anterior knee laxity (AKL) was used as the covariate. The dependent variables in each MANCOVA model came from one of the following variable groups: 1) kinematics (TrunkCoM_{IC}, HF_{IC}, KF_{IC}, TrunkCoM_{EXC}, HF_{EXC}, KF_{EXC}, HFV, and KFV); 2) kinetics (vGRF_{PK}, pGRF_{PK}, HEM_{PK}, and KEM_{PK}); 3) neuromuscular (QUAD_{PRE} and HAM_{PRE}); and 4) biomechanical characteristics of ACL loading (ATT_{PK}, ATA_{PK}, and PTASF_{PK}). Our decision to include AKL as a covariate was based on the fact that AKL is different in males and females, and has previously been shown to influence knee-joint biomechanics (Schmitz et al., 2013; Shultz, S. J., Shimokochi, et al., 2006).

Results

Males and females were not statistically different from one another in terms of age (t_{67} = 0.78, P = .430) or AKL (M: 7.4 ± 2.6, F: 7.8 ± 2.4 mm; t_{67} = -0.68, P = .502); however, males were significantly taller (t_{67} = 8.85, P < .001) and heavier (t_{67} = 7.52, P < .001) than females.

When examining the results of the MANCOVA analyses, the three-way sex-by-laxity-bytask interactions were not statistically significant for kinematic (Wilks' $\Lambda = .86$, $F_{(8, 54)} = 1.10$, multivariate $\eta_p^2 = .14$, P = .376, $1-\beta = .46$), kinetic (Wilks' $\Lambda = .96$, $F_{(3, 61)} = 0.76$, multivariate $\eta_p^2 = .14$, P = .376, P = .46), kinetic (Wilks' P = .96), kinetic (Wilks' P = .96), P = .96, P = .96, P = .96, multivariate P = .96, P = .= .04, P = .524, $1-\beta = .20$) neuromuscular (Wilks' $\Lambda = .98$, $F_{(2, 61)} = 0.57$, multivariate $\eta_p^2 = .02$, P= .570, 1- β = .14), or ACL loading-characteristic (Wilks' Λ = .90, $F_{(3, 60)}$ = 2.12, multivariate η_p^2 = .10, P = .107, $1-\beta = .52$) variable groups. Additionally, the two-way sex-by-laxity interactions were not statistically significant for kinematic (Wilks' $\Lambda = .85$, $F_{(8,54)} = 1.15$, multivariate $\eta_p^2 =$.15, P = .345, $1-\beta = .48$), kinetic (Wilks' $\Lambda = .93$, $F_{(3,61)} = 1.58$, multivariate $\eta_p^2 = .07$, P = .204, 1-β = .40) neuromuscular (Wilks' Λ = .99, $F_{(2, 61)}$ = 0.31, multivariate η_p^2 = .01, P = .737, 1-β = .10), or ACL loading-characteristic (Wilks' $\Lambda = .98$, $F_{(3, 60)} = 0.47$, multivariate $\eta_p^2 = .02$, P =.705, $1-\beta = .14$) variable groups. Together, these findings indicate that the effect of laxity (i.e. AKL) on between-task differences that are dependent on sex, and the effect of AKL on betweensex differences, were not different across sex. Thus, our use of AKL as a covariate was justified. The remainder of this section focuses only on the main effects of sex and task, and the sex-bytask interactions. All data are presented as adjusted means (i.e. the original mean adjusted for the covariate) ± standard error, unless otherwise stated; these values were adjusted based on a fixed AKL value of 7.66 mm. Results from the MANCOVA analyses for the main effects of sex and task are presented in Table 4.1. Results from the MANCOVA analyses for the interaction effects are presented in Table 4.2. All unadjusted means and standard deviations are located in the appendix (Appendix C1).

There were significant sex-by-task interactions for kinematic (Wilks' $\Lambda = .76$, $F_{(8,55)} =$ 2.15, multivariate $\eta_p^2 = .24$, P = .046, $1-\beta = .80$) and kinetic (Wilks' $\Lambda = .84$, $F_{(4,59)} = 2.92$, multivariate $\eta_p^2 = .17$, P = .029, $1-\beta = .75$) variables groups. Follow-up univariate analyses revealed that, after adjusting for AKL, the effect of task on HFV ($F_{(1,62)} = 5.60$, $\eta_p^2 = .08$, P =.021, $1-\beta = .64$) and HEM_{PK} ($F_{(1,62)} = 7.62$, $\eta_p^2 = .11$, P = .008, $1-\beta = .78$) was different for males and females. Irrespective of task, females landed with slower HVF ($F_{(1,62)} = 4.40$, $\eta_p^2 = .07$, P =.040, 1- β = .54; Table 4.1) and lesser HEM_{PK} ($F_{(1,62)}$ = 18.47, η_p^2 = .23, P < .001, 1- β = .99; Table 4.1) compared to males; however, the interaction was such that females performed the SLSJ with a 36.5% reduction in HFV and a 16.7% increase in HEM_{PK} whereas males performed the SLSJ with less than a 1% change in HFV and a 33.3% increase in HEM_{PK} (Figure 4.5A, B). There was also a statistically significant sex-by-task interaction for biomechanical characteristics of ACL loading (Wilks' $\Lambda = .87$, $F_{(3, 61)} = 2.94$, multivariate $\eta_p^2 = .13$, P = .040, $1-\beta = .67$), however, this interaction was not significant at the univariate level. Further, there was a statistically significant laxity-by-task interaction for biomechanical characteristics of ACL loading (Wilks' Λ = .74, $F_{(3)}$ $_{61)}$ = 7.15, multivariate η_p^2 = .26, P < .001, 1- β = .98). Follow-up univariate analyses revealed that, after adjusting for AKL, the effect of AKL on PTASF_{PK} was task dependent ($F_{(1, 63)} = 11.52$, $\eta_p^2 = .16$, P = .001, $1-\beta = .92$). Irrespective of sex, the SLSJ elicited significantly greater PTASF_{PK} compared to the DLSJ ($F_{(1, 63)} = 34.24$, $\eta_p^2 = .35$, P < .001, $1-\beta = 1.00$; Table 4.1); however; the interaction was such that individuals with higher magnitudes of AKL displayed greater increases in PTASF_{PK} when performing the SLSJ compared to individuals with lower magnitudes of AKL (Table 4.2; Figure 4.5C). No other statistically significant interactions were observed.

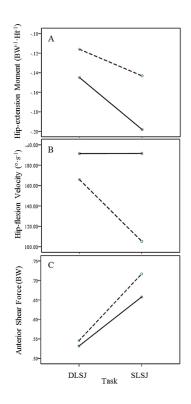


Figure 4.5. Profile Plots Depicting the Sex-by-Task Interactions for Peak Hip-Extension Moment (A) and Average Hip-Flexion Angular Velocity (B), and the Laxity-by-Task Interaction for Peak Proximal Tibia Anterior Shear Force (C). Male and female participants are represented by the solid and dashed lines, respectively.

When examining differences between tasks, the MANCOVA analyses revealed significant main effects for kinematic (Wilks' Λ = .44, $F_{(8,55)}$ = 8.89, multivariate η_p^2 = .56, P < .001, 1- β = 1.00), kinetic (Wilks' Λ = .41, $F_{(4,59)}$ = 20.90, multivariate η_p^2 = .59, P < .001, 1- β = 1.00), and ACL loading-characteristic (Wilks' Λ = .51, $F_{(3,61)}$ = 19.83, multivariate η_p^2 = .49, P < .001, 1- β = 1.00) variable groups. Irrespective of sex, and after adjusting for AKL, males and females performed the SLSJ with their TrunkCoM_{IC} positioned more posteriorly ($F_{(1,62)}$ = 16.55, η_p^2 = .21, P < .001, 1- β = .98; Table 4.1); with lesser KF_{IC} ($F_{(1,62)}$ = 24.23, η_p^2 = .28, P < .001, 1- β = 1.00; Table 4.1) and KF_{EXC} throughout landing ($F_{(1,62)}$ = 4.50, η_p^2 = .07, P = .038, 1- β = .55; Table 4.1); and with greater vGRF_{PK} ($F_{(1,62)}$ = 63.87, η_p^2 = .51, P < .001, 1- β = 1.00; Table 4.1),

pGRF_{PK} ($F_{(1, 62)} = 21.13$, $\eta_p^2 = .25$, P < .001, $1-\beta = 1.00$; Table 4.1), and KEM_{PK} ($F_{(1, 62)} = 4.77$, $\eta_p^2 = .07$, P = .033, $1-\beta = .58$; Table 4.1), and lesser ATT_{PK} ($F_{(1, 63)} = 7.83$, $\eta_p^2 = .11$, P = .007, $1-\beta = .79$; Table 4.1). No other statistically significant between-task differences were observed.

When examining between-sex differences, the MANCOVA analyses revealed significant main effects for kinematic (Wilks' Λ = .68, $F_{(8,55)}$ = 3.29, multivariate η_p^2 = .32, P = .004, 1- β = .95) and ACL loading-characteristic (Wilks' Λ = .84, $F_{(3,61)}$ = 3.83, multivariate η_p^2 = .16, P = .014, 1- β = .79) variable groups. Irrespective of task, and after adjusting for AKL, females landed with lesser KF_{IC} ($F_{(1,62)}$ = 4.12, η_p^2 = .06, P = .047, 1- β = .52; Table 4.1), lesser HF_{EXC} throughout landing ($F_{(1,62)}$ = 8.00, η_p^2 = .11, P = .006, 1- β = .80, Table 4.1), and greater ATT_{PK} ($F_{(1,63)}$ = 8.25, η_p^2 = .12, P = .006, 1- β = .81; Table 4.1), compared to males. There was also a statistically significant main effect of sex for neuromuscular activation (Wilks' Λ = .90, $F_{(2,62)}$ = 3.29, multivariate η_p^2 = .10, P = .044, 1- β = .60); however, the univariate analyses for preparatory activation of the quadriceps ($F_{(1,63)}$ = 3.13, η_p^2 = .05, P = .082, 1- β = .41) and hamstring ($F_{(1,63)}$ = 1.64, η_p^2 = .03, P = .205, 1- β = .24) muscles were not significant.

Table 4.1. The Effects of Sex and Task on All Variables of Interest. Data are presented as the adjusted mean \pm standard error [95% confidence interval].

	Sex Main Effect		Task Main Effects	
	Male (n = 34)	Female (n = 32)	DLSJ	SLSJ
Kinematics				
TrunkCOM _{IC} (cm)*	-11.6 ± 0.7 [-13.1, -10.2]	-11.3 ± 0.7 [-12.8, -9.8]	-10.3 ± 0.6 [-11.4, -9.2]	-12.7 ± 0.6 [-13.8, -11.5]
TrunkCOM _{EXC} (cm)	$11.8 \pm 0.8 [10.2, 13.4]$	11.7 ± 0.8 [10.1, 13.4]	$12.1 \pm 0.7 [10.8, 13.4]$	$11.4 \pm 0.7 [10.1, 12.7]$
HF _{IC} (°)	26.0 ± 5.0 [15.9, 36.0]	$27.5 \pm 5.1 [17.3, 37.7]$	$34.8 \pm 3.9 [26.9, 42.6]$	$18.7 \pm 3.8 [11.2, 26.2]$
$HF_{EXC}(^{\circ})$ †	$56.4 \pm 3.6 [49.3, 63.6]$	$41.9 \pm 3.7 [34.6, 49.2]$	$54.4 \pm 2.9 [48.7, 60.2]$	$44.0 \pm 2.8 [38.3, 49.6]$
HFV (°·s⁻¹)†	191.5 ± 18.7 [154.2, 228.8]	135.6 ± 18.9 [97.7, 173.5]	$178.7 \pm 15.7 [147.3, 210.0]$	148.4 ± 13.7 [121.1, 175.8]
KF _{IC} (°)†	7.9 ± 1.2 [5.4,10.3]	4.3 ± 1.3 [1.8, 6.8]	11.1 ± 0.9 [9.2, 12.9]	1.1 ± 1.0 [-0.8, 3.0]
KF _{EXC} (°)	60.8 ± 1.8 [57.3, 64.3]	$57.6 \pm 1.8 [54.1, 61.2]$	$66.3 \pm 1.4 [63.4, 69.1]$	$52.1 \pm 1.2 [49.7, 54.6]$
KFV (°·s⁻¹)	231.1 ± 5.2 [220.8, 241.4]	$243.5 \pm 5.2 [233.0, 253.9]$	265.5 ± 4.8 [255.9, 275.1]	209.1 ± 3.9 [201.2, 216.9]
Kinetics				
$vGRF_{Pk}(BW)^*$	2.3 ± 0.1 [2.2, 2.5]	2.2 ± 0.1 [2.0, 2.3]	1.6 ± 0.1 [1.5, 1.7]	2.8 ± 0.1 [2.7, 3.0]
$pGRF_{Pk}(BW)^*$	-0.6 ± 0.0 [-0.7, -0.6]	-0.6 ± 0.0 [-0.6, -0.5]	-0.5 ± 0.00 [-0.5, -0.4]	-0.7 ± 0.0 [-0.8, -0.7]
$HEM_{Pk}(BW^{\text{-}1}\!\cdot\!Ht^{\text{-}1})\boldsymbol{\dagger}$	-0.17 ± 0.01 [-0.19, -0.16]	-0.13 ± 0.01 [-0.14, -0.12]	-0.13 ± 0.00 [-0.14, -0.12]	-0.17 ± 0.01 [-0.18, -0.16]
$KEM_{Pk} (BW^{-1} \cdot Ht^{-1})^*$	-0.08 ± 0.00 [-0.09, -0.07]	-0.08 ± 0.00 [-0.09, -0.07]	-0.08 ± 0.00 [-0.08, -0.07]	-0.08 ± 0.00 [-0.09, -0.07]
Neuromuscular				
QUAD _{PRE} (%MVIC)	$30.3 \pm 3.5 [23.1, 37.4]$	21.4 ± 3.5 [14.4, 28.5]	23.6 ± 2.2 [19.1, 28.1]	$28.1 \pm 3.1 [22.0, 34.2]$
HAMPRE (%MVIC)	8.8 ± 1.1 [6.68, 10.9]	10.7 ± 1.1 [8.6, 12.8]	$6.5 \pm 0.5 $ [5.5, 7.4]	$13.0 \pm 1.3 [10.5, 15.6]$
ACL Loading Charact	eristics			
$ATT_{Pk} (mm)$ † *	$21.5 \pm 5.3 [10.9, 32.1]$	43.4 ± 5.5 [32.5, 54.4]	41.2 ± 4.2 [32.9, 49.5]	$23.8 \pm 5.1 [13.6, 33.9]$
$ATA_{Pk}(m \cdot s^{-2})$	$17.0 \pm 1.1 [14.9, 19.1]$	19.8 ± 1.1 [17.6, 22.0]	$19.6 \pm 1.0 [17.6, 21.7]$	17.2 ± 0.7 [15.7, 18.6]
PTASF _{Pk} (BW)*	$0.60 \pm 0.03 \; [0.54, 0.65]$	$0.63 \pm 0.03 \; [0.58, 0.68]$	$0.54 \pm 0.02 [0.50, 0.57]$	$0.69 \pm 0.02 [0.64, 0.73]$

^{*}Indicates a significant task main effect (P < .05). † Indicates a significant sex main effect (P < .05).

Table 4.2. Sex-by-Task and Laxity-by Task Interaction Effects for All Variables of Interest. Data are presented as the adjusted mean \pm standard error [95% confidence interval].

	Males (n = 34)		Females (n = 32)	
	DLSJ	SLSJ	DLSJ	SLSJ
Kinematics				
TrunkCOM _{IC} (cm)	-10.6 ± 0.8 [-12.2, -9.0]	-12.7 ± 0.8 [-14.3, -11.1]	-10.0 ± 0.8 [-11.6, -8.4]	-12.6 ± 0.8 [-14.2, -11.0]
$TrunkCOM_{EXC}$ (cm)	$12.2 \pm 0.9 [10.3, 14.1]$	11.4 ± 0.9 [9.6, 13.3]	$12.1 \pm 0.9 [10.2, 13.9]$	11.4 ± 0.9 [9.5, 13.2]
HF _{IC} (°)	34.7 ± 5.5 [23.7, 45.7]	$17.3 \pm 5.3 [6.7, 27.8]$	34.8 ± 5.6 [23.6, 46.0]	$20.1 \pm 5.4 [9.4, 30.8]$
HF _{EXC} (°)	$60.3 \pm 4.1 [52.1, 68.4]$	$52.6 \pm 4.0 [44.6, 60.6]$	$48.6 \pm 4.1 \ [40.3, 56.8]$	$35.3 \pm 4.1 [27.2, 43.4]$
HFV (°·s⁻¹)‡	$191.5 \pm 22.1 [147.4, 235.5]$	191.5 ± 19.3 [153.1, 230.0]	165.9 ± 22.4 [121.1, 210.7]	105.3 ± 19.6 [66.2, 144.4]
KF _{IC} (°)	$12.6 \pm 1.3 [10.0, 15.2]$	$3.1 \pm 1.4 \ [0.4, 5.8]$	9.5 ± 1.3 [6.9, 12.2]	-1.0 ± 1.4 [-3.7, 1.8]
KF _{EXC} (°)	$68.8 \pm 2.0 [64.8, 72.8]$	$52.7 \pm 1.7 [49.3, 56.2]$	63.8 ± 2.0 [59.7, 67.9]	51.5 ± 1.8 [48.0, 55.0]
KFV (°⋅s ⁻¹)	261.5 ± 6.8 [247.9, 275.0]	200.8 ± 5.5 [189.7, 211.8]	269.6 ± 6.9 [255.8, 283.3]	217.4 ± 5.6 [206.2, 228.6]
Kinetics				
$vGRF_{Pk}$ (BW)	$1.7 \pm 0.1 [1.5, 1.8]$	3.0 ± 0.1 [2.8, 3.1]	$1.6 \pm 0.1 [1.4, 1.7]$	2.7 ± 0.1 [2.6, 2.9]
pGRF _{Pk} (BW)	-0.5 ± 0.00 [-0.5, -0.4]	-0.8 ± 0.0 [-0.8, -0.7]	-0.4 ± 0.0 [-0.5, -0.4]	-0.7 ± 0.0 [-0.8, -0.7]
$HEM_{Pk} (BW^{-1} \cdot Ht^{-1})$ ‡	-0.15 ± 0.01 [-0.16, -0.13]	-0.20 ± 0.01 [-0.22, -0.18]	-0.12 ± 0.01 [-0.13, -0.10]	-0.14 ± 0.01 [-0.16, -0.13]
$KEM_{Pk} (BW^{-1} \cdot Ht^{-1})$	-0.08 ± 0.00 [-0.08, -0.07]	-0.08 ± 0.01 [-0.09, -0.07]	-0.08 ± 0.00 [-0.08, -0.07]	-0.08 ± 0.01 [-0.10, -0.07]
Neuromuscular				
QUAD _{PRE} (%MVIC)	$27.2 \pm 3.2 [20.9, 33.5]$	$33.3 \pm 4.3 [24.7, 42.0]$	$19.9 \pm 3.2 [13.6, 26.3]$	$22.9 \pm 4.3 [14.2, 31.6]$
HAMPRE (%MVIC)	$6.0 \pm 0.7 [4.7, 7.3]$	$11.6 \pm 1.8 [8.0, 15.2]$	7.0 ± 0.7 [5.6, 8.3]	$14.5 \pm 1.8 [10.9, 18.0]$
ACL Loading Character	ristics			
ATT_{Pk} (mm)	25.5 ± 5.8 [13.9, 37.2]	$17.5 \pm 7.1 [3.4, 31.6]$	$56.8 \pm 6.0 [44.9, 68.8]$	30.0 ± 7.3 [15.5, 44.6]
$ATA_{Pk} (m \cdot s^{-2})$	$17.4 \pm 1.4 [14.6, 20.3]$	$16.6 \pm 1.0 [14.5, 18.6]$	21.8 ± 1.5 [18.9, 24.8]	$17.8 \pm 1.1 [15.6, 19.9]$
PTASF _{Pk} (BW)‡‡	$0.53 \pm 0.02 \; [0.48, 0.58]$	$0.66 \pm 0.03 \; [0.59, 0.72]$	$0.55 \pm 0.03 \; [0.50, 0.60]$	$0.72 \pm 0.03 \; [0.65, 0.78]$

[‡] Indicates a significant sex-by-task interaction (P < .05). ‡‡ Indicates a significant AKL-by-task interaction (P < .05).

Discussion

The purpose of this study was to compare neuromuscular and biomechanical demands of DLSJ to those of a SLSJ, and then determine whether the SLSJ accentuated characteristics indicative of greater ACL loading in females as compared to males. The findings of this study revealed that, after adjusting for AKL, males and females performed the SLSJ with significantly smaller knee-flexion angles and a more posteriorly-oriented trunk CoM position at IC, less knee-flexion excursion, greater vertical and posterior ground reaction forces, knee-extension moments, and proximal tibia anterior shear forces, and less anterior tibial translation, compared to the DLSJ (Tables 2 & 3). Moreover, the increase in anterior shear force observed during the SLSJ was found to be accentuated in participants with higher magnitudes of AKL. In addition, while females performed both stop-jump tasks with smaller knee-flexion angles at IC, less hip-flexion excursion, slower hip-flexion velocities, smaller hip-extension moments, and greater amounts of anterior tibial translation, compared to males, females performed the SLSJ with a greater reduction in hip-flexion velocity, and a smaller increase in hip-extension moment, compared to males.

To the best of our knowledge, only two previous studies have examined differences between double-leg and single-leg jump-landing tasks using the stop-vertical jump as a model for injury. Compared to the DLSJ, Wang (Wang, 2011) reported that elite male volleyball players performed the SLSJ with smaller hip- and knee-flexion angles at IC, less hip- and knee-flexion excursions and slower angular velocities throughout landing, greater posterior and vertical ground-reaction forces and knee-extension moments, and greater proximal tibia anterior shear force. Similarly, Taylor et al (Taylor et al., 2016) reported that recreationally-active females performed the SLSJ with smaller peak knee-flexion angles, and greater hip- and knee-extension moments, compared to the DLSJ. Thus, our findings that participants in the current study

performed the SLSJ with smaller knee-flexion angles and a more posterior trunk CoM position at IC, less knee-flexion excursion, greater ground-reaction and anterior-shear forces, and greater knee-extension moments, compared to the DLSJ (Tables 1 & 2), are in general agreement with prior work. These findings are also in agreement with previous comparisons between double- and single-leg drop-landings (Pappas et al., 2007; Weinhandl et al., 2010b; Yeow et al., 2010). Furthermore, although we did not observe any significant between-task differences in neuromuscular activation, initial hip angles, hip excursions and moments, or hip and knee velocities, the direction-of-change in these characteristics when performing the SLSJ (i.e. increased activation and moments, and reduced initial contact angles, excursions, and velocities) was consistent with what has been observed in previous comparisons between double- and single-leg jump-landing tasks (Pappas et al., 2007; Wang, L. I., 2011; Weinhandl et al., 2010b; Yeow et al., 2010).

We hypothesized that the SLSJ would elicit a landing style considered to be more "risky" in terms of ACL loading and noncontact injury compared to the DLSJ, and that these characteristics would be more pronounced in females. Landing in a more upright position is thought to contribute to the noncontact ACL injury mechanism because several retrospective video analyses have observed athletes to be leaning backward, with their landing leg positioned anterior to the body and their knee relatively extended (< 30° flexion), at the estimated time of injury (Boden et al., 2000; Boden et al., 2009; Sheehan, Sipprell, & Boden, 2012). In addition, trunk CoM position has been shown to retrospectively discriminate between athletes who sustained noncontact ACL injury and those who did not with 80% accuracy, with injured athletes displaying a more posterior trunk CoM position compared to their uninjured counterparts (Sheehan et al., 2012). Based on this evidence, we believe that the landing style displayed by participants when performing the SLSJ (i.e. landing with the trunk positioned more posterior and

smaller knee-flexion angles at IC) supports our hypothesis that the SLSJ would elicit more "risky" landing mechanics compared to the DLSJ. This hypothesis is further supported by previous studies demonstrating that landing in a more upright position elicits higher groundreaction forces, sagittal-plane hip and knee moments, and proximal tibia anterior shear forces, compared to landing in a more flexed position (Blackburn & Padua, 2009; Kulas, A. S., Hortobagyi, & Devita, 2010). This is also generally consistent with the between-task differences observed in the current study, and research suggesting that higher forces and moments can results in higher magnitudes of ACL loading when the knee is relatively extended (due to tibio-femoral joint geometry, e.g. the slope of the tibial plateau) and the quadriceps line of pull acting through the anteriorly-directed patellar tendon, among others (DeMorat et al., 2004; Li et al., 1999; Withrow et al., 2006). These findings, in combination with the fact that noncontact ACL injuries occur more frequently when landing on a single leg (Boden et al., 2000; Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), collectively suggest that the SLSJ may be a better task to use in future studies aimed at identifying factors that potentially contribute to ACL injury risk since it appears to provoke a landing posture that is more representative of what is observed during actual noncontact ACL injury situations, and that it subjects the knee joint to moments and forces considered to be indicative of increased ligamentous loading. Likewise, injury prevention programs should be encouraged to incorporate more single-leg jumping and landing activities into training and promote more flexed landing styles in future work.

While our findings of between-task differences support the notion that single-leg tasks elicit biomechanical characteristics associated with increased risk for injury compared to double-leg tasks, we were also interested in determining whether the characteristics elicited by the SLSJ would be more pronounced in females compared to males; which would be detected by a significant sex-by-task interaction. In this regard, we found significant interactions for hip-flexion

velocity and peak hip-extension moment, suggesting that females and males used different "strategies" at the hip when adjusting to the increased demands of the SLSJ. This may be clinically important in terms of noncontact ACL injury risk since the proximal and distal segments of the body's kinetic chain can dramatically affect knee-joint biomechanics (Griffin et al., 2006). Compared to the DLSJ, female participants performed the SLSJ with a greater reduction in hip-flexion velocity (F: 36.5% vs. M: < 1%; Table 4.2) and a smaller increase in peak hip-extension moment (F: 16.7% vs. M: 33.3%; Table 4.2) than males. We are unaware of any previous work that has provided evidence to support the idea that the effect of task (i.e. single- vs double-leg) on sagittal-plane hip velocities and moments is dependent on sex; however, unilateral landings have been shown to elicit reduced hip-flexion velocities (Wang, L. I., 2011), and increased hip-extension moments (Weinhandl et al., 2010a), compared bilateral landings during stop-vertical jump and drop-landing tasks, respectively. That said, landing with a reduced hip-flexion velocity and an increased hip-extension moment, in the presence of a more upright body position at IC and less knee-flexion excursion, has been characterized as a more "stiff" landing style that places greater reliance on the lower-extremity musculature and passive tissue structures to absorb the external forces on the body and protect the knee joint from deleterious loading (Devita & Skelly, 1992). Thus, our finding that females displayed a greater reduction in hip-flexion velocity compared to males when performing the SLSJ suggests that landing on a single leg was likely more challenging for females compared to males, which may in part help explain why females are at increased risk for injury. It is unclear as to why females were not observed to display a greater increase in hip-extension moment compared to males when performing the SLSJ; however, we suspect that between-sex differences in body positioning at IC likely contributed to this finding. As such, we suggest that future studies place greater focus on the how body positioning at IC potentially mediates the relationship between hip- and knee-joint

biomechanics when landing on a single leg. Additionally, given that the effect of task on hip biomechanics was different for males and females, using sex-stratified research designs in future studies appears warranted.

Because of the inherent difficulties associated with directly measuring the loads experienced by the ACL during dynamic athletic maneuvers, a number of studies have used proximal tibia anterior shear force as a biomechanical indicator of ACL loading since it has been demonstrated that this force, and its concomitant anterior tibial translation, most directly loads the ACL in the sagittal plane (Markolf et al., 1995; Markolf et al., 1990). In agreement with prior work (Wang, 2011), our findings revealed that the SLSJ elicited greater anterior shear force than the DLSJ. However, a rather unique finding of this study was the increase in shear force elicited by the SLSJ was dependent on the magnitude of an individual's knee laxity. Specifically, participants with higher versus lower magnitudes of anterior knee laxity (AKL) experienced a greater increase in anterior shear force when performing the SLSJ as compared to the DLSJ (Figure 4.5C). As mentioned previously, our rationale for including AKL as a covariate was based on previous reports that individuals with greater AKL values display greater amounts of anterior tibial translation during weight acceptance (Shultz, S. J., Shimokochi, et al., 2006), and greater knee-joint stiffness and extensor loading during double-leg drop-vertical jumps (Schmitz et al., 2013; Shultz, S. J. et al., 2010a). Additionally, characteristics of the AKL loaddisplacement curve (stiffness) have been shown to influence anterior shear force during dropvertical jumps (Schmitz et al., 2013). Because dynamic knee stability is derived from both passive (ligaments) and active (skeletal muscle) restraint mechanisms, individuals with higher amounts of AKL, and thus reduced passive restraint capabilities, may have altered their landing mechanics in order to compensate for the increased demands of the SLSJ in a way that ultimately elicited increased anterior shear force. Although not observed in this study, AKL is typically

greater in females compared to males (Rozzi, Lephart, Gear, & Fu, 1999). This observed sex difference, coupled with prospective injury risk studies identifying AKL as an independent predictor of ACL injury risk (Vacek et al., 2016), these effects of AKL on landing strategies during single-leg tasks should be considered in future injury risk screening and prevention studies.

With regard to between-sex differences, the characteristics displayed by males and females during both landing tasks in the current investigation generally support what has been reported in the literature. Specifically, we found that females performed both stop-jump tasks with smaller knee-flexion angles at IC, less hip-flexion excursion throughout landing, slower hipflexion angular velocities, smaller hip-extension moments, and greater amounts of anterior tibial translation, compared to males. Although this may be the first study to identify such differences during a SLSJ task, females have previously been reported to display smaller initial knee-flexion angles (Chappell et al., 2007; Decker et al., 2003; Yu et al., 2006), smaller sagittal-plane excursions (Schmitz et al., 2007; Weinhandl et al., 2010b), and slower angular velocities (Schmitz et al., 2007; Yu et al., 2006), than males during the DLSJ and during single- and doubleleg drop-landings. Thus, our findings are consistent with prior work, and add to the current body of literature by demonstrating that such differences are also present when performing a stopvertical jump on a single leg. It should be noted, however, that other studies using double-leg drop-landings have reported that females display similar (Kernozek et al., 2005) or greater (Fagenbaum & Darling, 2003) knee-flexion angles at IC, greater sagittal-plane angular excursions (Decker et al., 2003; Kernozek et al., 2005; Shultz, S. J. et al., 2009) and angular velocities (Decker et al., 2003), and similar hip-extension moments (Decker et al., 2003), compared to males. We believe that these discrepancies may likely be due to differences between tasks used as an injury model, and we encourage readers to take caution when attempting to compare the

findings of this study to previous work using tasks other than the stop-vertical jump as a model for injury. With this, females have also been reported to display greater preparatory neuromuscular activation of the quadriceps and hamstrings (Chappell et al., 2007; Shultz, S. J. et al., 2009), greater knee-extension moments (Chappell et al., 2002; Kernozek et al., 2005; Pappas et al., 2007; Schmitz et al., 2007; Shultz, S. J. et al., 2009; Yu et al., 2006), greater posterior and vertical ground-reaction forces (Kernozek et al., 2005; Pappas et al., 2007; Salci et al., 2004; Schmitz et al., 2007; Weinhandl et al., 2015; Yu et al., 2006), and greater proximal-tibia-anteriorshear force (Chappell et al., 2002; Yu et al., 2006), compared to males. Although not statistically significant, our data tend to support these findings (Tables 1 & 2). Aside from the task used potentially affecting our findings, the absence of such between-sex differences in the current study may be attributed to the use of a relative starting/approach distance (i.e. 40% body height) as opposed to the more common practice of having all participants perform a task from a fixed drop height or starting/approach distance. This is supported by the work of Huston et al (Huston, Vibert, Ashton-Miller, & Wojtys, 2001), who found similar drop-jump landing mechanics between males and females at lower drop heights, but that between-sex differences became more apparent as drop height increased. It could therefore be hypothesized that some of the differences reported by previous studies could be due greater demands being placed on females in order to perform a given task.

Limitations

The current study was conducted using a sample of physically-active male and female participants who regularly participated in activities that involved running, cutting, jumping and landing. While every effort was made to recruit a sample that was representative of an athletic population, the participants included in this study may have exhibited different neuromuscular and biomechanical characteristics from what might have been observed had we included highly-

competitive athletes. Additionally, the stop-vertical jump tasks used in this study were performed in a controlled laboratory setting, from a stationary starting position, and participants knew exactly what to expect prior to task execution. While starting each jump from a stationary position improved experimental control in this study, it reduces the external validity because such movements typically occur as part of a dynamic activity (e.g. a running approach with an abrupt deceleration and a jump landing, followed by a subsequent movement). With this, because our participants had knowledge of the specific tasks they were being asked to perform prior to initiating the movement, this may have allowed participants to preplan their movement patterns, and may not reflect the movement patterns that would be observed during competition, where participants are often required to react to unanticipated events. Thus, the tasks performed in this study may not be entirely representative of the more dynamic and unanticipated environment in which noncontact ACL injuries occur. We also acknowledge that, although there is general consensus that the mechanism(s) of noncontact ACL injury is multi-planar, this study only examined the effects of task and sex on sagittal-plane biomechanical characteristics, which may not fully represent the multi-directional nature of such injuries. Thus, future studies including athletic populations, and more dynamic and unanticipated tasks, are warranted.

Conclusion

The findings of this study add support to the theory that the task used as a model for noncontact ACL injury has the ability to affect biomechanical outcome measures, and that performing the a given landing task on a single leg elicits different biomechanical responses compared to performing the same task with both legs. These findings suggest that objective landing assessments should occur during conditions that maximize the external validity in order to more adequately assess how individuals move under conditions that are more representative of

the those commonly observed at the time of injury. Given that the SLSJ elicited characteristics thought to be associated with increased risk for noncontact ACL injury, and that noncontact ACL injuries are more likely to occur when landing on a single leg, it seems intuitive that researchers would want to use an injury model that best represents the situations in which these injuries occur. This is not to say that the SLSJ is the answer, but only to encourage researchers to carefully consider selecting a task that would best help answer the research question at hand. In addition, our finding that the SLSJ accentuated characteristics thought to be associated with ACL loading and injury risk to a greater extent in females compared to males, suggests that the demands of jumping and landing on a single leg are different for males and females. Furthermore, our finding that greater amounts of laxity were associated with greater increases in anterior shear force during the SLSJ when compared with the DLSJ, indicates that laxity my play a greater role in knee joint loading as the demands of the task increase. These sex-specific and laxity-specific results suggest that the risk factors may not be the same for men and women, and that prevention programs may need to be tailored to these individual and sex differences.

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CHAPTER V

MANUSCRIPT II

Title

The Influence of Hamstring Stiffness on Sagittal Plane ACL Loading Characteristics during a Single-Leg Stop-Jump Landing

Abstract

Background: Higher hamstring stiffness (K_{HAM}) has been shown to be predictive of lesser anterior tibial translation (ATT) and proximal-tibia-anterior-shear force (PTASF) during controlled perturbations and double-leg jump landings. However, these relationships have not been examined during more functional single-leg stop-jump (SLSJ) landing.

Purpose: To determine, within each sex, the extent to which K_{HAM} predicts ACL loading characteristics (i.e. PTASF, ATT, and anterior tibial acceleration (ATA)) during a SLSJ, after controlling for initial body positioning (i.e. trunk center-of-mass position and hip- and kneeflexion angles at initial ground contact).

Study Design: Cross-Sectional.

Methods: K_{HAM} was assessed in 69 males (n=36) and females (n=33) under two loading conditions (K_{HAM} _BM = 10% body mass load; K_{HAM} _MVIC = 30% maximal voluntary isometric contraction load). Landing biomechanics were assessed during a SLSJ. Separate, sex-specific hierarchical multiple linear regression analyses examined the extent to which K_{HAM} _BM and K_{HAM} _MVIC predicted ACL loading characteristics.

Results: Initial body positioning predicted 27.7% (P =.023) of the variance in ATA in females, and K_{HAM} _BM and K_{HAM} _MVIC explained an additional 0.3% (P=.720; overall R^2 =.280, P=.049) and 3.2% (P=.263; overall R^2 =.309, P=.030) of the variance in ATA, respectively. Initial body positioning predicted 29.5% (P =.010) and 31.9% (P=.010) of the variance in PTASF for males and females, respectively, with K_{HAM} _BM and K_{HAM} _MVIC explaining an additional 7.4% (P=.065; overall R^2 =.369, P=.005) and 6.3% (P=.092; overall R^2 =.357, P=.007) of the variance in males, and 1.8% (P=.393; overall R^2 =.337, P=.018) and 4.1% (P=.842; overall R^2 =.320, P=.025) of the variance in females. The change in R^2 was not statistically significant for any model.

Conclusion: K_{HAM} was not a predictor of ACL loading characteristics in either sex. These findings conflict with previous work and suggest that K_{HAM} may not be as effective during a single-leg leg landing, potentially due to a more erect landing style.

Clinical Relevance: K_{HAM} is modifiable, offering great potential for ACL injury prevention and rehabilitation; however, the ability of the hamstrings to adequately resist ACL loading characteristics when landing on a single-leg deserves further investigation. Clinicians are encouraged to equally focus on improving K_{HAM} and teaching safer landing positions.

Key Terms: shear force; stiffness; hamstrings; landing biomechanics; free-oscillation; anterior cruciate ligament

Introduction

Noncontact anterior cruciate ligament (ACL) injuries commonly occur as the relatively extended knee (< 30° flexion) initially transitions from non-weight bearing to weight bearing following ground contact during cutting and jump-landing maneuvers (Koga et al., 2010; Krosshaug et al., 2007; Olsen et al., 2004). Although the precise mechanism of injury is likely multi-planar, it is well accepted that the ACL is most directly loaded via sagittal plane biomechanics, such as impact-induced anterior tibial acceleration (ATA), proximal tibia anterior shear force (PTASF), and anterior tibial translation (ATT) (Butler et al., 1980; Markolf et al., 1995; McLean, S. G. et al., 2011). It is also well accepted that there is an anterior shift of the tibia relative to the femur (i.e. ATT) that naturally occurs as the knee initially transitions from nonweight bearing to weight bearing (Fleming, Renstrom, Beynnon, et al., 2001; Torzilli et al., 1994). Mechanistically, this is attributed to resultant ground reaction force-induced compressive loading, acting through the posterior-inferior slope of the tibial plateau, and the compulsory quadriceps-induced knee-extension moment needed to stabilize the knee joint and control the body's deceleration upon landing (Fleming, Renstrom, Beynnon, et al., 2001; Torzilli et al., 1994). At more extended knee angles ($< 30^{\circ}$ flexion), this quadriceps contraction acts through the anteriorly-oriented patellar tendon to create additional PTASF and ATT, further loading the ACL (DeMorat et al., 2004; Li et al., 1999). Thus, identifying modifiable factors able to effectively resist these biomechanical characteristics indicative of ACL loading (i.e. PTASF, ATA, and ATT) may help pave the way for evidence-based targeted intervention strategies aimed to reduce ACL loading and injury risk.

The hamstrings function antagonistically to the quadriceps due to their attachments on the posterior tibia and fibula, and several studies demonstrate that adequate co-contraction of the hamstring muscles can effectively reduce biomechanical characteristics of ACL loading, and

ACL loading itself, at knee flexion angles greater than 10-15° (Baratta et al., 1988; Beynnon et al., 1995; Draganich & Vahey, 1990; Imran & O'Connor, 1998; Li et al., 1999; MacWilliams et al., 1999; Pandy & Shelburne, 1997; Solomonow et al., 1987; Withrow et al., 2008). However, due to the inherent difficulties associated with measuring hamstring muscle forces and ACL loading in-vivo, these demonstrated effects are currently limited to open-kinetic-chain isometric knee-extension exercises, musculoskeletal modeling simulations, and cadaver models. As such, others have focused on measures of neuromuscular activation amplitude and timing, via surface electromyography, to further elucidate the role of the quadriceps and hamstring muscles during functional athletic movements such as landing from a jump. These studies show that preparatory activation of the quadriceps and hamstring muscles occurs in anticipation of initial ground contact in order to increase overall joint stiffness and enhance functional knee stability (Bryant et al., 2008; McNair & Marshall, 1994; Swanik et al., 2004). Given that noncontact ACL injuries occur within the first 50 milliseconds of initial ground contact (Koga et al., 2010; Krosshaug et al., 2007), any imbalance or delay in preparatory muscle activation may lead to improper limb positioning and higher ACL loading, increasing injury risk. In this regard, a property of the hamstring muscles that may play a critical role in helping resist biomechanical characteristics of ACL loading immediately upon landing is musculo-articular stiffness. Hamstring musculoarticular stiffness (K_{HAM}) is a modifiable neuromechanical property that quantifies the resistance of the hamstring musculo-articular unit to lengthening in response to an applied load (i.e. Δ Force $/\Delta$ Length) (Blackburn & Norcross, 2014). Thus, it is theorized that, for a given load, relatively stiffer hamstrings will permit a smaller change in length compared to more compliant hamstrings, thereby limiting tibiofemoral joint motion and the biomechanical characteristics that contribute to ACL loading.

There is currently a small, but growing body of literature to support the theory that K_{HAM} may play a critical role in ACL loading by helping control tibiofemoral motion. Specifically, ACL-deficient individuals with higher K_{HAM} display greater functional knee stability than more compliant individuals (McNair et al., 1992), which suggests that K_{HAM} may help supplement the stability roles of the native ACL. Additionally, greater K_{HAM} in healthy individuals is associated with lesser ATT (Blackburn et al., 2011) and PTASF (Blackburn et al., 2013) during controlled perturbations and double-leg landing tasks, respectively, which suggests that individuals with greater K_{HAM} may experience lesser ACL loading. Further, healthy females display less K_{HAM} (Blackburn et al., 2009; Blackburn et al., 2004), perform dynamic landing tasks with greater PTASF (Chappell et al., 2002; Sell et al., 2007; Yu et al., 2006), and are at substantially greater risk of experiencing noncontact ACL injury (Arendt, E. & Dick, 1995), compared to their male counterparts. But while these studies lend support to the theory that that higher levels of K_{HAM} may help protect the ACL from deleterious loading, actual evidence linking greater K_{HAM} to reduced ACL loading is limited to studies of non-weight bearing perturbations (Blackburn et al., 2011) and double-leg jump landings (Blackburn et al., 2013).

Noncontact ACL injuries are more likely to occur when cutting or landing on a single leg, and large between-limb asymmetries in weight-distribution have been observed during injuries resulting from double-leg landings (Boden et al., 2009; Hewett et al., 2009; Olsen et al., 2004). In addition, laboratory-based studies demonstrate that single-leg landings elicit greater peak ground reaction forces and knee-extension moments (internal), a more upright landing with lesser hip-and knee-flexion angles and angular velocities at initial ground contact, and greater PTASF, compared to double-leg landings (Pappas et al., 2007; Wang, L. I., 2011; Yeow et al., 2010). Thus, open-kinetic-chain perturbations and double-leg jump-landings may not adequately

represent the situations (thus the protective restraint capabilities of the hamstrings) in which noncontact ACL injuries commonly occur.

The current body of literature is also limited by the statistical analyses employed in previous work, as well as methodological differences in the way that $K_{\rm HAM}$ has been assessed. For example, the noted relationship between $K_{\rm HAM}$ and ACL-loading characteristics has been established with males and females included in the same statistical analyses and without equal sex-stratification (Blackburn et al., 2013; Blackburn et al., 2011). Because $K_{\rm HAM}$ is highly correlated with sex (Blackburn et al., 2009; Blackburn et al., 2004), this makes it difficult to tease out the unique contribution of $K_{\rm HAM}$ versus other sex-dependent factors. Similarly, between-sex differences in trunk center of mass position (relative to center-of-pressure) and hip- and kneeflexion angles during landing tasks have been reported (DiStefano et al., 2005; Malinzak et al., 2001; Schmitz et al., 2007; Yu et al., 2006), and such variables are shown to influence both ground reaction forces and PTASF (Sell et al., 2007; Yu et al., 2006). Despite these potential confounds, previous investigations on the relationship between $K_{\rm HAM}$ ACL-loading characteristics have not exercised statistical control over such variables, which may have influenced their findings.

Further complicating matters are methodological differences in the way that K_{HAM} has previously been assessed. Specifically, the relationship between K_{HAM} and ATT was established when K_{HAM} was assessed using a standardized load equal to 10% body mass (Blackburn et al., 2011), whereas the relationship between K_{HAM} and PTASF was established when K_{HAM} was assessed using a load equal to 45% maximal voluntary isometric contraction (Blackburn et al., 2013). Given that K_{HAM} is influenced by neuromuscular activation levels (Ditroilo et al., 2011; Jennings & Seedhom, 1998), it is likely that standardizing the applied load as a percentage of total body mass may not be as precise as when standardizing the load as a percentage of muscular

capability (i.e. maximal voluntary isometric contraction). Understanding the impact of these methodological differences will help inform preferred metrics in future studies, and aid in the interpretation of findings between studies using different standardized loading assignments.

Because K_{HAM} is modifiable through training (Blackburn & Norcross, 2014), understanding the extent to which K_{HAM} is uniquely associated with biomechanical characteristics of ACL loading in males and females during functional athletic tasks may pave the way for more targeted injury prevention strategies in future work. Thus, the purpose of this study was twofold:

1) to determine, within each sex, the extent to which K_{HAM} predicts biomechanical characteristics of ACL loading during a functional single-leg landing task, and 2) to determine whether this relationship is influenced by the method used to standardize the K_{HAM} assessment load. It was hypothesized that: 1) after controlling for trunk center-of-mass position and hip- and knee-flexion angles at initial ground contact, higher K_{HAM} would be predictive of less PTASF, ATA, and ATT, and 2) K_{HAM} would be more predictive of biomechanical factors indicative of ACL loading when assessed using a load assigned as a percentage maximal voluntary isometric contraction compared to using a load assigned as a percentage body mass.

Methods

Participants

An a priori power analysis was performed using G*Power, version 3.1.9.2 (Faul, Erdfelder, Buchner, & Lang, 2009), in order to determine the sample size needed to test our primary research hypothesis. Based on a total of 4 predictor variables, an anticipated medium $(R^2\Delta=0.15;\,f^2=0.18)$ to large $(R^2\Delta=0.25;\,f^2=0.33)$ effect size, and a statistical significant criterion of $\alpha=0.05$, this analysis indicated that 40 participants per sex (N = 80) would result in statistical power between .73 (medium effect) and .94 (large effect). The effect sizes used for this

power analysis were conservatively estimated based on the theory that there would need to be at least a medium effect size for $K_{\rm HAM}$ to be considered a clinically meaningful factor for ACL loading. As such, 40 males (21.6 \pm 2.1 years; 1.8 \pm 0.1 m; 81.8 \pm 13.5 kg) and 40 females (21.2 \pm 1.8 years; 1.7 \pm 0.1 m; 65.1 \pm 12.7 kg) were recruited for participation.

At the time of recruitment, all participants were physically active, defined as regularly engaging in greater than the equivalent of 300 minutes per week of moderate-intensity physical activity (assessed via the International Physical Activity Questionnaire-Short Form; Appendix B), and regularly participated in activities that involved, running, cutting, jumping and landing. Additionally, all participants were without any history of the following: 1) knee ligamentous or meniscal injury, 2) lower-extremity surgery, 3) lower-extremity injury in the 6 months prior to recruitment, and 4) known medical conditions affecting their connective tissue or vestibular system. This study was approved by the university's Institutional Review Board for the Protection of Human Subjects prior to recruitment, and written informed consent was obtained from each participant prior to testing.

Procedures

Participants visited the laboratory for a single testing session during which all data were collected. In order to control for any potential effects of cycling female hormones on stiffness (Bell, D. R. et al., 2012) or lower-extremity biomechanics (Shultz, S. J. et al., 2012; Shultz, S. J. et al., 2011), all female testing was constrained to the follicular phase of the menstrual cycle (i.e. days 1-8 following self-reported onset of menstrual bleeding). Upon arrival to the laboratory, participants changed into laboratory-issued compression shorts and a tight-fitting athletic top. After obtaining barefoot measures of body height and mass, participants then completed a 5-minute warm-up on a stationary cycle ergometer (Life Fitness, Schiller Park, IL) at a cadence of

70-80 RPM and were asked to maintain a target rating of perceived exertion (RPE) of \geq 3-4 on a Borg CR-10 RPE scale (Borg, 1998). Hamstring musculo-articular stiffness (K_{HAM}) and single-leg landing biomechanics were assessed following the warm-up. Because K_{HAM} does not differ between limbs in healthy individuals (Jennings & Seedhom, 1998), all measurements were obtained from the left lower-extremity, which corresponded with the dominant limb (defined a-priori as the stance limb when participants were asked which limb they would use to kick a ball for maximum horizontal distance) in most participants (68 of 80; 85%).

Hamstring Musculo-Articular Stiffness. Hamstring musculo-articular stiffness (K_{HAM}) was assessed via the free-oscillation technique, whereby the leg is modeled as a single-degree-offreedom mass-spring system, and the damping effect that the hamstring muscles impose on oscillatory flexion-extension at the knee joint is then quantified following a perturbation (Blackburn et al., 2011; Blackburn et al., 2004; McNair et al., 1992). Prior to assessing K_{HAM}, a twin-axis electrogoniometer (Biometrics Ltd, Ladysmith, VA) was attached to the participant's knee joint in order to obtain knee flexion angle data in real-time (Figure 5.1.A). Additionally, a thermoplastic splint was secured to the plantar aspect of the participant's foot and posterior shank in order to standardize ankle position. Participants were then positioned prone on a padded table with the trunk and thigh supported in 30° of flexion and lower-leg and foot segment free to move. Next, a standardized load was secured to the distal shank (at the level of the malleoli) using cuffstyle ankle weights (Figure 5.1.B). The participant's lower-leg was then passively positioned so that the knee was in approximately 30° of flexion, and the participant was instructed to maintain this position via isometric hamstring contraction; during this time, real-time knee joint angle data were displayed on a monitor, giving participants a visual target to maintain (Figure 5.1.D). Within 5 seconds of the participant holding this position, a brief downward perturbation was manually applied to the posterior aspect of the calcaneus, resulting in slight knee extension and subsequent

damped oscillatory flexion-extension. This damped oscillatory motion was then characterized as the tangential acceleration of the shank and foot segment, and captured via a triaxial accelerometer (Sensor dimensions: 2.54 x 2.54 x 1.91 cm; NeuwGhent Technology, USA) attached to the thermoplastic splint (Figure 5.1.C). Participants were verbally instructed not to interfere with or voluntarily produce the oscillations following the perturbation, and to attempt to keep the hamstring muscles active only to the level necessary to support the mass of the shank and foot segment, and the applied load, in the testing position (Blackburn et al., 2004; Waxman, Schmitz, & Shultz, 2015). This assessment was performed under two different loading conditions. In the first condition, the standardized applied load was assigned as 10% of the participant's body mass (K_{HAM} BM). In the second condition, the applied load was assigned as 30% of the participant's peak isometric hamstring torque production, which was obtained via maximal voluntary isometric contraction testing (K_{HAM}_MVIC; see below). It should be noted that although we attempted to use a 45% MVIC load to allow for better comparison with previous work, pilot testing (unpublished data) revealed that some participants (females in particular) were simply unable to stabilize the applied load in the required assessment position. As such, we decided to reduce the load to 30% MVIC specifically because the hamstrings have been shown to be activated ~30% MVIC during the stance phase of gait activities (Ciccotti, Kerlan, Perry, & Pink, 1994). To avoid a potential order effect, these conditions were assigned in a counterbalanced fashion. Under each loading condition, participants performed 3 to 5 practice trials, followed by 5 test trials in which data were recorded. All test trials were separated by 30second rest intervals to minimize any likelihood of fatigue.

Maximal voluntary isometric contraction (MVIC) testing for the hamstring musculature was performed using a Biodex System 3 dynamometer (Biodex Medical Systems, Shirley, NY). Participants were positioned prone with their hip and knee fixed in 30° of flexion, and a strap

secured just proximal to the medial and lateral malleoli to ensure a constant body position (Figure 5.2). This position was chosen in order to replicate the length-tension relationship of the hamstrings during the K_{HAM} assessments. Each participant performed four familiarization trials (25%, 50%, 75%, and 100% self-perceived MVIC), followed by three maximal effort test trials from which data were recorded. Each MVIC trial was held for 5 seconds, and 1-minute rest intervals were provided between trials to minimize any likelihood of fatigue. The peak isometric torque value obtained across the three test trials was then used to calculate a standardized load for each participant when K_{HAM} was assessed using a standardized load equal to 30% peak isometric torque (i.e. K_{HAM} _MVIC).

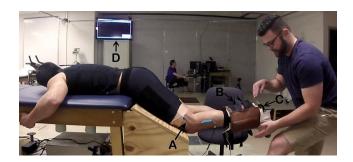


Figure 5.1. Participant Positioning and Instrumentation for the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. Electrogoniometer placement (A); Thermoplastic splint & ankle weights (B); Accelerometer (C); Monitor displaying real-time knee-flexion angle data (D).



Figure 5.2. Participant Positioning During Hamstring Maximal Voluntary Isometric Contraction (MVIC) Testing.

Single-Leg Landing Biomechanics. Single-leg landing biomechanics were assessed during a single-leg stop-jump landing task (SLSJ). To experimentally control for any potential effects of footwear on landing biomechanics, all participants were standardized footwear (Adidas Uraha 2, Adidas North America, Portland, OR). Participants were then instrumented with fourmarker clusters of optical LED markers so that three-dimensional kinematic data could be obtained using an eight-camera IMPULSE motion tracking system (Phase Space, San Leandro, CA). Specifically, marker clusters were placed on the posterior thorax (over the C7 spinous process) and sacrum, and on the lateral thigh (mid-shaft), medial and lateral tibial flares, lateral shank (mid-shaft), and foot of the left leg. The posterior thorax marker cluster was secured via a thin shoulder harness, whereas the sacral and tibial flare marker clusters were secured directly to the skin using double-sided adhesive tape. Lateral thigh and shank marker clusters were secured to the participant's compression shorts and a thin shank sleeve, respectively, using hook and loop material. Participants were then digitized using MotionMonitor software (Innovative Sports Training, Chicago, IL). Ankle and knee joint centers were determined as the midpoint between the medial and lateral malleoli and medial and lateral femoral epicondyles, respectively. Hip joint center were determined using the Bell method (A. L. Bell, Brand, & Pedersen, 1989).

For the SLSJ, participants began on a starting line placed at a distance of 40% of their body height behind the rear edge of a non-conducting force platform (Type 4060-130; Bertec Corporation., Columbus, OH) (Figure 5.3). Standing on their left leg, participants were instructed to: 1) perform a single-leg hop towards the force platform, 2) land on the platform using only their left leg, 3) jump for maximum vertical height immediately following landing, and 4) land on the platform again using only their left leg (Figure 5.3). In an effort to prevent any experimenter bias, participants were not provided with any special instructions regarding their stop-jump biomechanics. After performing 3 to 5 practice trials, participants completed 5 successful test

trials, during which data were recorded. Thirty-second rest intervals were provided between each test trial to minimize any likelihood of fatigue. A trial was considered successful if the participant: 1) hopped from the starting line and landed on the force platform, 2) jumped for maximum vertical height immediately after landing, and 3) landed back on the force platform following the vertical jump. Unsuccessful trials were discarded and repeated.



Figure 5.3. Visual Depiction of the Single-Leg Stop-Jump Landing (SLSJ) Task.

Data Sampling and Reduction

All kinetic and kinematic hardware were integrated and time-synchronized with MotionMonitor software (Innovative Sports Training Inc., Chicago, IL) for data collection. Accelerometer data recorded during the $K_{\rm HAM}$ assessments were sampled at 1000 Hz within MotionMonitor. These data were then low-pass filtered at 10 Hz, using a fourth-order zero-lag Butterworth filter, and subsequently exported from MotionMonitor to Matlab (Mathworks, Inc., Natick, MA) for data reduction. Within Matlab, the time-interval between the first two oscillatory peaks of the accelerometer time series was identified and used to calculate the damped frequency of oscillation for each trial (Figure 5.4). Hamstring musculo-articular stiffness ($K_{\rm HAM}$) was then calculated using the equation: $K_{\rm HAM} = 4\pi^2 m f^2$, where m is the summed mass of the lower-leg and foot segment and the applied load, and f is the damped frequency of oscillation. $K_{\rm HAM}$ values

were normalized to body mass $(N \cdot m^{-1} \cdot kg^{-1})$, and the average of 5 trials for each condition (i.e. K_{HAM} _BM and K_{HAM} _MVIC) was then calculated for use in statistical analyses.

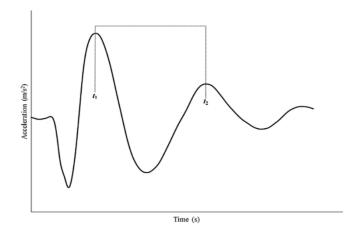


Figure 5.4. Example Accelerometer Time-Series Data Obtained During the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. t_1 and t_2 represent the time points at which the first two oscillatory peaks occur; these time points are then used to calculate the damped frequency of oscillation.

Kinetic and kinematic data recorded during the SLSJ were sampled at 1000 Hz and 240 Hz, respectively. Kinematic data were subsequently linearly interpolated to 1000 Hz. All data were then low-pass filtered at 12 Hz using a fourth-order zero-lag Butterworth filter. For kinematic data, a segmental reference system was defined for all body segments with the z-axis as the medial-lateral axis (flexion/extension), the y-axis as the longitudinal axis (internal/external rotation), and the z-axis as the anterior-posterior axis (abduction/adduction). Joint motions were then calculated using Euler angle definitions with a rotational sequence of Z Y' X" (Kadaba et al., 1989). Trunk center-of-mass (CoM) position was defined as the anterior-posterior displacement (cm) of the trunk's CoM relative to the center-of-pressure (CoP). Proximal tibia anterior-posterior shear force (PTASF) was calculated using inverse dynamics (Gagnon & Gagnon, 1992).

the proximal tibia (tibial flare marker cluster) relative to the femur (lateral thigh marker cluster). The second derivative of the anterior-posterior tibial translation data was then calculated to assess anterior-posterior tibial acceleration. All data were exported to Matlab in order to extract our variables of interest at the instant of initial ground contact (IC; time at which the vertical ground reaction force first exceeded 10 N) and throughout the landing phase (i.e. from IC to maximum vertical CoM displacement). These variables of interest included trunk CoM position and hip- and knee-flexion angles at IC (TCoM_{IC}, HF_{IC}, and KF_{IC}, respectively), and peak anterior tibial translation (ATT), anterior tibial acceleration (ATA), and PTASF, throughout the landing phase. Averages were calculated across 5 trials for use in statistical analyses.

Statistical Analysis

All statistical analyses were performed using SPSS (Version 23; IBM Corp, Armonk, NY) with an *a-priori* statistical significance criterion of $\alpha \le 0.05$. A confirmatory analysis of between-sex differences was performed across all variables of interest using separate one-way analysis of variance (ANOVA) models. Separate hierarchical multiple linear regression analyses for each sex were then conducted to determine the extent to which K_{HAM} was predictive of biomechanical factors indicative of ACL loading, after controlling for differences in body positioning at initial ground contact (i.e. TCoM_{IC}, HF_{IC}, and KF_{IC}). Of secondary interest was to determine whether K_{HAM} MVIC predicted biomechanical factors indicative of ACL loading to a greater extent than K_{HAM} MVIC measures raised concerns of multicollinearity (men: r = .43, p = .009; women: r = .69, p < .001); thus, we felt it would be inappropriate to include both measures in a single model and simply explore their effects. Instead, we empirically compared the proportions of variability in the criterion explained by each K_{HAM} measure, after controlling for

differences in body positioning at initial ground contact. To do this, a single biomechanical factor indicative of ACL loading (i.e. ATT, ATA, or PTASF) was used as the criterion variable in each model. Body positioning variables (i.e. $TCoM_{IC}$, HF_{IC} , and KF_{IC}) were collectively entered into the first block of each model as control predictor variables, and a single K_{HAM} measure (K_{HAM} _BM or K_{HAM} _MVIC) was then entered into the second block of each model as an additional predictor. These regression analyses were run separately for men and women, resulting in a total of six models per sex. In instances where K_{HAM} _BM and K_{HAM} _MVIC each resulted in a statistically significant R^2 change for a single criterion variable, the variation around the difference in R^2 (i.e. K_{HAM} _MVIC – K_{HAM} _BM) was then empirically determined by conducting a post-hoc bootstrap analysis with 5000 iterations.

Results

Four male participants and six female participants were excluded from statistical analyses after preliminary data inspection revealed unstable kinematic data at the hip. One additional female was excluded from analyses due to being unable to stabilize the $K_{\rm HAM}$ _MVIC assessment load. Thus, only data from 36 male (21.5 \pm 2.0 years, 1.8 \pm 0.1 m, 81.4 \pm 11.0 kg) and 33 female (21.1 \pm 2.0 years, 1.7 \pm 0.1 m, 63.0 \pm 9.1 kg) participants were analyzed.

Between-Sex Differences (ANOVA)

Means and standard deviations (M \pm SD) for all variables of interest are presented in Table 1. One-way ANOVA results revealed that, compared to males, females were significantly shorter, had less body mass, displayed greater K_{HAM} BM, and performed the SLSJ with less KF_{IC} (p < .05), supporting our rationale for using sex-specific regression models (Table 5.1).

Table 5.1. Means and Standard Deviations for all Variables of Interest.

	Males	Females			
	$(Mean \pm SD)$	$(Mean \pm SD)$	F(1, 68)	Cohen's d	P-Value
Age (years)	21.47 ± 2.02	21.09 ± 1.96	0.631	0.191	.430
Body Height (m)	1.82 ± 0.06	1.66 ± 0.08	78.349	2.263	< .001*
Body Mass (kg)	81.35 ± 10.97	63.02 ± 9.11	56.486	1.818	< .001*
K_{HAM} BM (N·m·kg ⁻¹)	13.43 ± 2.59	15.12 ± 2.27	8.305	-0.694	.005*
K_{HAM} _MVIC (N·m·kg ⁻¹)	13.52 ± 3.06	12.17 ± 2.97	3.423	0.448	.069
TCOM _{IC} (cm)	-23.40 ± 7.42	-21.14 ± 8.31	1.434	-0.287	.235
$\mathrm{HF}_{\mathrm{IC}}$ (°)	16.73 ± 19.78	18.89 ± 37.72	0.091	-0.072	.764
KF_{IC} (°)	2.87 ± 6.88	-0.90 ± 8.13	4.356	0.501	.041*
ATT_{Pk} (mm)	17.28 ± 46.50	31.90 ± 33.20	2.221	-0.362	.141
$ATA_{Pk} (m \cdot s^{-2})$	16.12 ± 5.94	17.60 ± 6.57	0.969	-0.236	.328
PTASF _{Pk} (BW)	0.67 ± 0.19	0.71 ± 0.19	0.660	-0.211	.420

Note. K_{HAM} _BM = hamstring musculo-articular stiffness assessed using a load equal to 10% body mass (BM); K_{HAM} _MVIC = hamstring musculo-articular stiffness assessed using a load equal to 30% maximal voluntary contraction (MVIC); $TCOM_{IC}$ = anterior(+)/posterior(-) trunk center of mass position at initial ground contact (IC); HF_{IC} = hip flexion angle at IC; KF_{IC} = knee flexion angle at IC; ATT_{Pk} = peak anterior tibial translation; ATA_{Pk} = peak anterior tibial acceleration; $PTASF_{Pk}$ = peak proximal tibia anterior shear force. *Denotes a statistically significant difference between sexes ($p \le .05$).

Hierarchical Multiple Linear Regression

Model summaries for the hierarchical multiple linear regression analyses are presented in Table 5.2. When examining the extent to which $K_{\rm HAM}$ predicted biomechanical factors indicative of ACL loading, after controlling for body positioning at IC, only the full models predicting ATA (females only) and PTASF (males and females) were found to be statistically significant (p < .05; Table 5.2). Parameter estimates, separated by sex, for the full (final) regression models predicting ATA and PTASF are presented in Table 3 and Table 4, respectively.

When predicting ATA, the full regression models were found to be statistically significant for female participants only. Specifically, these analyses revealed that the linear combination of TCoM_{IC}, HF_{IC}, KF_{IC}, and K_{HAM} BM; and the linear combination of TCoM_{IC}, HF_{IC}, KF_{IC}, and K_{HAM} MVIC, explained 28% (Overall $R^2 = .280$, F(4, 32) = 2.73, p = .049) and 30.9% (Overall $R^2 = .309$, F(4, 32) = 3.13, p = .030) of the variance in ATA, respectively. Once

differences in body positioning at IC were controlled for (R^2 = .277, p = .023), K_{HAM} _BM and K_{HAM} _MVIC were only able to explain an additional 0.3% ($R^2\Delta$ = .003, p = .720) and 3.2% ($R^2\Delta$ = .032, p = .263) of the variance in ATA, respectively; these changes in R^2 were not statistically significant (Table 5.2). In addition, regardless of whether the final model included K_{HAM} _BM or K_{HAM} _MVIC, only the parameter estimates for HF_{IC} were statistically significant (0.071, p = .014 and 0.068, p = .021, respectively). In each case, these parameter estimates indicated that, after holding all other predictors constant, greater hip-flexion angles at IC predicted greater peak anterior tibial acceleration in females (Table 5.3).

When predicting PTASF, the full regression models were found to be statistically significant for both male and female participants. For the male analyses, the linear combination of $TCoM_{IC}$, HF_{IC} , KF_{IC} , and K_{HAM} BM explained 36.9% of the variance in PTASF (Overall R^2 = .369, F(4, 35) = 4.53, p = .005), whereas linear combination of TCoM_{IC}, HF_{IC}, KF_{IC}, and K_{HAM} MVIC explained 35.7% of the variance in PTASF (Overall $R^2 = .357$, F(4, 35) = 4.31, p =.007). Once differences in body positioning at IC were controlled for $(R^2 = .295, p = .010)$, K_{HAM} BM and K_{HAM} MVIC explained and additional 7.4% (R² Δ = .074, p = .065) and 6.3% (R² Δ = .063, p = .092) of the variance in PTASF, respectively; these changes in R^2 were not statistically significant. For the female analyses, the linear combination of TCoM_{IC}, HF_{IC}, KF_{IC}, and K_{HAM} BM explained 33.7% of the variance in PTASF (Overall $R^2 = .337$, F(4, 32) = 3.56, p= .018), whereas linear combination of TCoM_{IC}, HF_{IC}, KF_{IC}, and K_{HAM}_MVIC explained 32.0% of the variance in PTASF (Overall $R^2 = .320$, F(4, 32) = 3.29, p = .025). Once differences in body positioning at IC were controlled for $(R^2 = .319, p = .010)$, K_{HAM} BM and K_{HAM} MVIC explained and additional 1.8% ($R^2\Delta = .018$, p = .393) and 4.1% ($R^2\Delta = .041$, p = .842) of the variance in PTASF, respectively; these changes in R^2 were not statistically significant. In addition, regardless of whether K_{HAM} BM or K_{HAM} MVIC was used to predict PTASF, only the parameter estimates

for KF_{IC} were statistically significant (range: 0.011 to 0.017, p-value range: < .001 to .005). In each case, these parameter estimates indicate that, after holding all other predictors constant, greater knee-flexion angles at IC predicted greater peak proximal tibia anterior shear force in both males and females (Table 5.4).

Table 5.2. Summary of Findings from Each of the Hierarchical Regression Models Examined.

Criterion	Predictors	Sex	Final R ²	Final Regression Equation		
ATT	TCOM _{IC} , HF _{IC} , KF _{IC} ,	Male	0.067	$\begin{split} ATT &= 68.35 + 1.15(TCOM_{IC}) + 0.13(HF_{IC}) - \\ 1.31(KF_{IC}) - 1.68(K_{HAM}_BM) \end{split}$		
	K _{HAM} _BM	Female	0.150	$\begin{split} ATT &= 51.12 + 0.26 (TCOM_{IC}) - 0.04 (HF_{IC}) - \\ 1.54 (KF_{IC}*) - 0.94 (\textit{K}_{HAM}_BM) \end{split}$		
ATT	TCOM _{IC} , HF _{IC} , KF _{IC} .	Male	0.065	$ATT = 61.96 + 1.15(TCOM_{IC}) + 0.06(HF_{IC}) - 1.25(KF_{IC}) - 1.12(K_{HAM}_MVIC)$		
	K _{HAM} _MVIC	Female	0.154	$\begin{split} ATT &= 23.11 + 0.21(TCOM_{IC}) - 0.03(HF_{IC}) - \\ 1.56(KF_{IC}^*) + 1.02(K_{HAM}_MVIC) \end{split}$		
ATA	TCOM _{IC} ,	Male	0.051	$ATA = 20.90* - 0.06(TCOM_{IC}) - 0.02(HF_{IC}) + 0.01(KF_{IC}) - 0.45(K_{HAM_}BM)$		
	HF _{IC} , KF _{IC} , K _{HAM} _BM	Female	0.280*	$\begin{split} ATA &= 23.13^* + 0.20(TCOM_{IC}) + 0.07(HF_{IC}^*) + \\ 0.13(KF_{IC}) &- 0.17(K_{HAM}_BM) \end{split}$		
ATA	TCOM _{IC} ,	Male	0.034	$ATA = 10.89 - 0.09(TCOM_{IC}) - 0.03(HF_{IC}) + 0.05(KF_{IC}) + 0.26(K_{HAM}_MVIC)$		
	HF _{IC} , KF _{IC} , <i>K</i> _{HAM} _MVIC	Female	0.309*	$ATA = 25.42* + 0.19(TCOM_{IC}) + 0.07(HF_{IC}*) + 0.13(KF_{IC}) - 0.40(K_{HAM}_MVIC)$		
PTASF	TCOM _{IC} ,	Male	0.369*	$\begin{split} PTASF &= 0.33 - 0.002 (TCOM_{IC}) - 0.002 (HF_{IC}) + \\ &0.02 (KF_{IC}^*) + 0.02 (K_{HAM}_BM) \end{split}$		
	HF_{IC} , KF_{IC} , $K_{HAM}BM$	Female	0.337*	$\begin{split} PTASF &= 0.90* \text{ - } 0.001(TCOM_{IC}) - 0.002(HF_{IC}) + \\ 0.01(KF_{IC}*) &- 0.01(\textit{K}_{HAM}_BM) \end{split}$		
PTASF	i comp,		0.357* $ PTASF = 0.38* - 0.002(TCOM_{IC}) - 0.001(HIO) - 0.02(K_{IC}*) + 0.02(K_{HAM}MVIC) $			
$ ext{HF}_{ ext{IC}}, ext{KF}_{ ext{IC},} \ ext{K}_{ ext{HAM}} ext{MVIC}$		Female	0.320*	$\begin{split} PTASF &= 0.73* - 0.002(TCOM_{IC}) - 0.002(HF_{IC}) + \\ 0.01(KF_{IC}*) &- 0.002(\textit{K}_{HAM}_MVIC) \end{split}$		

^{*}Denotes a statistically significant R^2 value or regression coefficient (p < .05)

Table 5.3. Parameter Estimates, Separated by Sex, for the Full Regression Models Predicting Anterior Tibial Acceleration (ATA).

					Correlations		
Predictor Variable	Parameter Estimate	Standard Error	t-Value	P-Value	Zero- Order	Part	Partial
MALE							
(Constant)	20.902	6.773	3.086	.004*			
$TCOM_{IC}$ (cm)	061	.143	426	.673	075	076	074
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$	016	.057	275	.785	081	049	048
KF_{IC} (°)	.008	.158	.051	.960	.025	.009	.009
K_{HAM} BM	445	.423	-1.052	.301	210	186	184
(Constant) TCOM _{IC} (cm)	10.894 090	6.315 .145	1.725 619	.094 .540	075	111	109
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$	031	.055	566	.576	081	101	100
KF_{IC} (°)	.053	.157	.336	.739	.025	.060	.059
K_{HAM} MVIC	.258	.350	.739	.466	.118	.132	.130
FEMALE							
(Constant)	23.129	8.206	2.818	.009*			
$TCOM_{IC}$ (cm)	.200	.129	1.547	.133	.277	.281	.248
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$.074	.028	2.628	.014*	.438	.445	.421
KF_{IC} (°)	.126	.130	.974	.338	.151	.181	.156
K_{HAM} BM	171	.473	362	.720	019	068	058
(Constant) TCOM _{IC} (cm)	25.421 .193	5.296 .125	4.800 1.551	.000* .132	.277	.281	.244
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$.068	.028	2.443	.021*	.438	.419	.384
KF_{IC} (°)	.128	.127	1.007	.323	.151	.187	.158
K_{HAM} MVIC	403	.353	-1.142	.263	250	211	179

^{*}Denotes a statistically significant parameter estimate $(p \le .05)$

Table 5.4. Parameter Estimates, Separated by Sex, for the Full Regression Models Predicting Proximal Tibia Anterior Shear Force (PTASF).

					Correlations		
Predictor Variable	Parameter Estimate	Standard Error	t-Value	<i>P</i> -Value	Zero- Order	Part	Partial
MALE							
(Constant)	.329	.177	1.861	.072			
$TCOM_{IC}$ (cm)	002	.004	439	.663	029	079	063
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$	002	.001	-1.167	.252	.026	205	166
$\mathrm{KF}_{\mathrm{IC}}\left(^{\circ}\right)$.017	.004	4.092	*000	.534	.592	.584
K_{HAM} BM	.021	.011	1.911	.065	.163	.325	.273
(Constant) TCOM _{IC} (cm)	.381 002	.165 .004	2.308 484	.028* .632	029	087	070
HF _{IC} (°)	001	.001	589	.560	.026	105	085
KF _{IC} (°)	.016	.004	3.934	*000	.534	.577	.566
K _{HAM} _MVIC	.016	.009	1.740	.092	.180	.298	.250
FEMALE							
(Constant)	.896	.232	3.858	.001*			
$TCOM_{IC}$ (cm)	001	.004	343	.734	082	065	053
$\mathrm{HF}_{\mathrm{IC}}\left(^{\circ}\right)$	002	.001	-1.918	.065	308	341	295
KF_{IC} (°)	.011	.004	3.086	.005*	.475	.504	.475
$K_{ m HAM}$ _BM	012	.013	868	.393	112	162	134
(Constant) TCOM _{IC} (cm)	.734 002	.155 .004	4.738 495	.000* .625	082	093	077
- ()	002	.004	493 -1.866	.073	082	093	077 291
HF _{IC} (°)							
KF _{IC} (°) K _{HAM} _MVIC	.011 002	.004 .010	3.014 201	.005* .842	.475 .037	.495 038	.470 031

^{*}Denotes a statistically significant parameter estimate ($p \le .05$)

Discussion

Previous laboratory-based studies have demonstrated that individuals with higher $K_{\rm HAM}$ display biomechanical characteristics indicative of lesser sagittal-plane ACL loading compared to individuals with lower $K_{\rm HAM}$ (Blackburn et al., 2013; Blackburn et al., 2011). It has also been demonstrated that females display significantly less $K_{\rm HAM}$ than similarly trained males, regardless

of whether the assessment load is assigned as a percentage of body mass or as a percentage of MVIC (Blackburn et al., 2009; Blackburn & Pamukoff, 2014; Blackburn et al., 2004; Granata et al., 2002), and perform dynamic landing tasks with characteristics indicative of greater sagittal-plane ACL loading (Chappell et al., 2002; Yu et al., 2006). These findings, coupled with the fact that females are at substantially greater risk of experiencing noncontact ACL injury compared to their male counterparts (Arendt, E. & Dick, 1995), have led to the notion that insufficient K_{HAM} may have important implications for ACL loading and injury risk. The present study was designed to expand on previous research by: 1) examining, within each sex, the extent to which K_{HAM} uniquely contributes to biomechanical characteristics of ACL loading during a functional single-leg landing (i.e. SLSJ), after controlling for differences in body positioning at IC; and 2) determining whether this relationship is influenced by the method used to standardize the K_{HAM} assessment load.

Contrary to our research hypotheses, K_{HAM} was not a unique predictor of ACL-loading characteristics in either sex, regardless of whether K_{HAM} was assessed using a standardized load equal to 10%BM or 30%MVIC. These findings also contradict previous reports of higher levels of K_{HAM} being associated with lesser ATT (Blackburn et al., 2011) and PTASF (Blackburn et al., 2013). However, the overall lack of agreement between our findings and previous work was not entirely unexpected given notable differences in the experimental and statistical methodologies employed, as well as the type of task used to assess biomechanical characteristics of ACL loading.

With few exceptions, K_{HAM} has been routinely assessed in a prone position, with the trunk and thigh supported in 30° of hip flexion, the knee initially flexed 30°, and a standardized load attached to the distal shank (Figure 5.1). However, the aforementioned relationship between K_{HAM} and ATT was established using an assessment load equal to 10%BM (Blackburn et al., 2011)

whereas the relationship between K_{HAM} and PTASF was established using an assessment load equal to 45%MVIC (Blackburn et al., 2013). This is problematic because K_{HAM} has been shown to be influenced by neuromuscular activation levels, with higher levels of neuromuscular effort being associated with higher K_{HAM} (Ditroilo et al., 2011; Jennings & Seedhom, 1998). As such, it could be argued that an individual with a high percentage of body fat, and thus a relatively low percentage of lean muscle mass, would require a greater neuromuscular effort to stabilize a 10%BM load than a body-mass matched individual with a low percentage of body fat. This would likely result in the high body fat individual displaying greater K_{HAM} than the low body fat individual, and potentially confound the relationship between K_{HAM} and ACL loading characteristics.

Alternatively, standardizing the applied load as a percentage of MVIC assigns the load relative to each individual's available lean muscle mass and maximal isometric strength capabilities, irrespective of body size. This would explain why females, who are well known to possess less lean body mass per unit of body weight than males, were found to display greater normalized K_{HAM} values than males when assessed using a 10%BM load (i.e. K_{HAM} _BM), but similar values to males when assessed using a 30%MVIC load (K_{HAM} _MVIC; Table 5.1). That said, the magnitudes of the K_{HAM} values obtained in the current study are consistent with prior work (Blackburn et al., 2009; Blackburn et al., 2013; Blackburn et al., 2011; Blackburn & Pamukoff, 2014; Blackburn et al., 2004). Additionally, our finding that the between-sex difference in K_{HAM} was eliminated when assessed using a 30%MVIC load is in agreement with a prior study using a 45%MVIC load (Blackburn & Pamukoff, 2014). However, our finding that females displayed greater K_{HAM} than males when assessed using a 10%BM load is contrary to previous reports of females displaying less K_{HAM} than males (Blackburn et al., 2009; Blackburn et al., 2004). While this finding deserves further investigation, we recommend that future studies

interested in examining the functional role that K_{HAM} potentially plays in ACL loading characteristics assess K_{HAM} using a load assigned as a percentage of MVIC in order to better ensure that individuals are loaded similarly from a neuromuscular perspective.

This is the first study to our knowledge to employ sex-specific statistical models when examining the relationship between measures of K_{HAM} and ACL loading characteristics. Our rationale for the use of sex-specific models was that between-sex differences in both $K_{\rm HAM}$ (Blackburn et al., 2009; Blackburn & Pamukoff, 2014; Blackburn et al., 2004; Granata et al., 2002) and landing biomechanics (Chappell et al., 2002; Pappas et al., 2007; Schmitz et al., 2007; Yu et al., 2006) have routinely been reported in the literature, yet studies rarely account for such differences in their statistical designs. When these between-sex differences are left unaccounted for, it becomes difficult to tease out whether correlated measures are being driven by a true relationship, or if they are simply being driven by a between-sex difference. For example, Blackburn et al (2011) established a relationship between K_{HAM} and ATT by separating males and females in high- and low-ATT groups based on the median ATT value, which resulted in a larger proportion of males in the low-ATT group, and a larger proportion of females in the high-ATT group. Given that this lab group has previously demonstrated that males display greater $K_{\rm HAM}$ than females using similar assessment methods (Blackburn et al., 2009; Blackburn et al., 2004), one could speculate that the finding of higher K_{HAM} being associated with lesser ATT may have been the result of between-sex differences in these factors as opposed to their being a true relationship. Similarly, the relationship between K_{HAM} and PTASF was established by roughly stratifying males and females into high- and low- $K_{\rm HAM}$ groups (Blackburn et al., 2013). Although it was noted that the high-K_{HAM} group displayed significantly greater knee-flexion angles at the instant of peak PTASF, these authors neglected to account for this difference in knee flexion before attributing the lower magnitudes of PTASF to higher K_{HAM} values (Blackburn et al., 2013). PTASF has been shown to increase as the knee becomes more extended due to an increase in the patellar tendon-tibial shaft angle (Li et al., 1999). Thus, the previous relationship between $K_{\rm HAM}$ and PTASF may have also been driven by between-sex differences in knee-flexion angles as opposed to differences in $K_{\rm HAM}$. After stratifying our analyses by sex, and controlling for differences in body positioning at IC, we observed no statistically significant relationship between either measure of $K_{\rm HAM}$ and any biomechanical characteristic of ACL loading (i.e. ATT, ATA, or PTASF). Sill, given the critical role of the hamstrings in controlling tibiofemoral motions and forces, there may be other methodological explanations for our lack of statistically significant findings.

When comparing the results of this study to prior research, it is important to consider the type of task used to examine the influence of $K_{\rm HAM}$ on ACL-loading characteristics. Previous studies have used a controlled non-weight bearing perturbation (Blackburn et al., 2011) and a double-leg jump-landing task (Blackburn et al., 2013) in order to examine the influence of $K_{\rm HAM}$ on ATT and PTASF, respectively, whereas we used a single-leg stop-vertical jump (SLSJ). We chose the SLSJ as a model for injury risk because observational video analyses have reported that those, at the estimated time of injury, injured athletes tend to land in a rear- or flat-footed position, with their knee relatively extended, their hip flexed, and their CoM positioned far posterior to their BoS (Boden et al., 2009; Sheehan et al., 2012). Thus, we felt that the SLSJ would elicit body positioning at IC that was more similar to what would be observed during an actual ACL injury situation than what would be elicited by the tasks used previously. This distinction is important because differences the model used to assess such relationships has the ability to influence initial body positioning, which in turn influences resultant landing forces and quadriceps-generated extensor moments, ultimately influencing ACL loading characteristics. Specifically, landing on a single leg has been shown to elicit a more upright landing posture at IC

compared to landing on both legs (Pappas et al., 2007; Wang, L. I., 2011; Yeow et al., 2010). In this regard, our data indicate that a majority of our participants landed with their trunk CoM positioned ~22 cm posterior to their BoS and their hip and knee flexed ~20° and ~10° at IC, respectively (Table 5.1), whereas the data presented by Blackburn et al (2013) indicate that a majority of their participants landed with approximately 20-30° knee flexion (trunk and hip data not reported). This is problematic because landing in a more upright position has been associated with higher ground reaction forces and knee-extensor moments (Blackburn & Padua, 2009; Sell et al., 2007; Wang, L. I., 2011; Yu et al., 2006), and higher PTASF (Kulas, A., Zalewski, Hortobagyi, & DeVita, 2008; Wang, L. I., 2011; Yu et al., 2006). With this, trunk CoM position (relative to the BoS) has been shown to discriminate between athletes who went on to sustain a noncontact ACL injury and those who did not with 80% accuracy, with injured athletes displaying a more posteriorly-oriented CoM position at the time of injury (Sheehan et al., 2012). Thus, landing with a more posteriorly-oriented trunk CoM, and smaller hip- and knee-flexion angles, is often considered to be "more risky" in terms of noncontact ACL injury mechanics. Although trunk CoM position data were not reported by Blackburn et al (2013), it is a reasonable assumption that the trunk CoM positions elicited by the SLSJ in the current study are greater than what would be expected during a double-leg landing. Collectively, these findings indicate that trunk CoM position and hip and knee angles at initial contact deserve consideration when attempting to better understand modifiable factors that potentially contribute to ACL loading and injury risk, and also draw attention to the model used to assess "at risk" landing biomechanics.

The body's positioning at IC is thought to play a critical role in the noncontact ACL injury mechanism(s) because the effect of both externally- (e.g. ground reaction) and internally-applied (e.g. muscle) forces on joint loads are influenced by both joint position and the orientation of each joint relative to one another. Landing with the hip and knee relatively

extended, and the trunk CoM positioned more posteriorly at IC has been suggested to elicit vigorous contraction of the trunk flexors and quadriceps muscles in an effort to "pull" the CoM anteriorly towards the BoS maintain balance and stability as the body decelerates (Sheehan et al., 2012). Given that the quadriceps line of pull through the patellar tendon is directed anteriorly at more extended knee angles, this vigorous quadriceps contraction would create additional compressive and anterior shear loading (i.e. PTASF), and thereby likely increase the load experienced by the ACL (DeMorat et al., 2004; Li et al., 1999). For the hamstrings however, the relative positioning of the hip and knee joint adversely affect the hamstrings line of pull on the posterior tibia and fibula, and thereby influence the hamstrings ability to effectively produce a posterior shear force component that would be of sufficient magnitude to reduce ACL loading (Herzog & Read, 1993). Although adequate co-contraction of the hamstring muscles have been reported to effectively reduce the net anterior shear force by creating a posterior shear force component, the hamstrings have only been shown to effectively accomplish this when the knee is flexed beyond 10-15° (Draganich & Vahey, 1990; Li et al., 1999; MacWilliams et al., 1999; Pandy & Shelburne, 1997; Withrow et al., 2008). Because the knee flexion angles observed in the current study were much smaller than those reported by Blackburn et al (2011) and Blackburn et al (2013), our lack of a statistically significant relationship between K_{HAM} and ACL-loading characteristics may simply be due to the fact that the hamstrings are not well-positioned to provide a protective effect at the knee joint when landing in such an extended position. Given that the hamstrings cross both the hip and knee, it may be that the relatively extended position of the hip influenced that length-tension relationship of the hamstrings, rendering them somewhat ineffective at resisting anterior loading. While additional studies are needed to better understand the functional role and capability of the hamstrings in resisting ACL loading during dynamic activity, our current findings suggest that equal attention should be placed on improving

hamstring strength/stiffness and teaching safer landing positions that have a greater potential to engage the protective role of the hamstrings when designing ACL prevention programs to best protect the knee from excessive ACL loading.

Limitations

We acknowledge that the current study is not without certain limitations. First, we recruited physically active males and females, who regularly performed activities that involved running, cutting, jumping and landing, in order to achieve a sample that was somewhat representative of the population in which noncontact ACL injuries commonly occur (i.e. athletes). Although our sample did contain a number of varsity collegiate athletes, it also contained a number of non-athlete individuals who simply participated in high volumes of physical activity at the time of recruitment, and may not be representative of those at risk for noncontact ACL injury. Thus, the results of this study are most generalizable to healthy, highly-active, college-aged males and females who regularly participate in multidirectional activities, and caution should be taken when attempting to generalize these results to other populations. Determining whether $K_{\rm HAM}$ influences ACL loading characteristics in athletic populations would help better elucidate whether this measure could be used as a screening tool of identifying athletes who may be at risk for injury. Second, the biomechanical data in this investigation were obtained from a single-leg stop-jump task (SLSJ) performed in a controlled laboratory setting, which may elicit different biomechanics from what would be observed during an actual practice or competition where most movements are unanticipated. It is also well accepted that the ACL loading is not limited to sagittal-plane biomechanics. Therefore, incorporating more challenging tasks, and examining the influence of K_{HAM} on frontal- and transverse-plane ACL loading characteristics (e.g. knee abduction/internal rotation angles and moments), are areas of future investigations. Third, due to the *in-vivo* nature of this study, we were unable to measure anterior cruciate ligament (ACL)

loading directly, and are therefore unable to know for certain whether or not $K_{\rm HAM}$ contributes to the loads experienced by the ACL. Finally, although the PTASF values obtained in the current study are in agreement with previously reported values (Chappell et al., 2002; Wang, L. I., 2011; Yu et al., 2006), the mean ATT values that we observed are much larger than what was reported during the controlled perturbation task (10.6 ± 11.0 mm) used by Blackburn et al (2011). Research has illustrated the difficulties associated with determining actual tibiofemoral motion during dynamic tasks, via traditional motion capture techniques, due to movement artifact induced by the large amount of soft tissue surrounding the knee joint (Leardini, Chiari, Della Croce, & Cappozzo, 2005). That said, it is likely that the highly dynamic nature of the single-leg landing task used in the current study resulted in substantially greater soft-tissue artifact than the tightly-controlled perturbation task used by Blackburn et al (Blackburn et al., 2011). Hence, the greater amount of measurement error induced by soft-tissue artifact in the current study may help explain the higher magnitude of ATT that we observed. Moreover, the associated increase in variability (standard deviation) may have reduced our likelihood of observing a relationship between measures of K_{HAM} and ATT. Additionally, because we obtained ATA by computing the second derivative of ATT, it is likely that the measurement error in ATT due to soft-tissue artifact was magnified even further for ATA. As such, future studies are encouraged to use PTASF as their primary biomechanical indicator of sagittal-plane ACL loading when using traditional motion capture techniques.

Conclusion

The findings of this study demonstrate that, after controlling for sagittal-plane body positioning at IC, K_{HAM} was not found to be predictive of ACL-loading characteristics during the

SLSJ in either sex, regardless of K_{HAM} assessment load used (%BM or %MVIC). Weighing all of the evidence, these findings suggests that K_{HAM} may not be as effective (or relevant) at resisting biomechanical characteristics of ACL loading during a single-leg landing where the individual's mass is positioned posteriorly and the hip and knee are closer to extension. While additional studies are needed to better understand the functional role of the hamstrings during dynamic single-leg movements, current prevention strategies are encouraged to focus on improving hamstring stiffness and promoting safer (more flexed) landing positions. Additionally, because between-sex differences in K_{HAM} were eliminated when load was assigned as a percentage of MVIC, we advise that future studies use this assessment method to ensure that neuromuscular demand is relatively consistent between individuals.

Acknowledgements

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CHAPTER VI

MANUSCRIPT III

Title

Predictors of Proximal Tibia Anterior Shear Force During a Vertical Stop-Jump: An Experiment Revisited

Abstract

Background: Proximal tibia anterior shear force (PTASF) is a biomechanical indicator of anterior cruciate ligament (ACL) loading that can be estimated in-vivo through inverse dynamics. While neuromuscular and biomechanical predictors of PTASF have been identified during a double-leg stop-jump landing, noncontact ACL injuries are more likely to occur when landing on a single-leg.

Purpose: To examine the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics collectively predicted PTASF during a single-leg stop-jump (SLSJ).

Study Design: Cross-sectional

Methods: Hamstring stiffness (K_{HAM}), anterior knee laxity (AKL), and SLSJ landing biomechanics, were assessed in 74 healthy physically-active individuals (male=37, female=37; 21.3±2.0 years, 1.8±0.1 m, 73.7±15.9 kg). Between-sex differences were evaluated via independent samples t-tests. A forward stepwise multiple linear regression analysis was then used to examine the extent to which these characteristics predicted PTASF.

Results: Independent t-tests revealed no statistically significant between-sex differences in $K_{\rm HAM}$ (P=.063), AKL (P=.974), or neuromuscular- or biomechanical-related characteristics evaluated during the SLSJ (P-value range=.079 to .978). Multiple linear regression revealed that the linear combination of preparatory neuromuscular activation of the lateral quadriceps, and knee-flexion angle and knee-extension moment at the instant of peak posterior ground reaction force, significantly predicted 78.4% (P<.001) of the variance in PTASF during the SLSJ. The parameter estimates indicated that greater knee-flexion angle (P<.001) and knee-extensor moment (P<.001), and lesser activation of the lateral quadriceps (P=.044), would predict greater magnitudes of PTASF.

Conclusion: Greater knee angles and moments, and lesser quadriceps activation, were shown to be predictive of greater PTASF when landing on a single leg, which would theoretically increase the forces experienced by the ACL. These findings are in general support of previous studies investigating predictors of PTASF during double-leg jump landing tasks, indicating that these characteristics are predictive of PTASF across a variety of landing tasks.

Clinical Relevance: While additional work is needed to better understand the relationship between PTASF and ACL loading in-vivo, the results of this study provide evidence to support the inclusion of preparatory quadriceps activation, knee angle and moment at the instant of peak posterior ground reaction force, and PTASF, as potential predictor variables in future studies aimed at prospectively identifying risk factors for noncontact ACL injury, or when examining adaptations elicited by current injury prevention efforts.

Key Terms: anterior cruciate ligament; biomechanics; sagittal plane; neuromuscular; shear force; landing

Introduction

Anterior cruciate ligament (ACL) injuries are estimated to affect more than 100,000 individuals annually in the United States alone, with the majority of these injuries occurring in young athletes between 15 and 25 years of age (Griffin et al., 2000). Aside from a high financial burden due to surgical reconstruction and rehabilitation costs (Brophy et al., 2009; Mather et al., 2013), these injuries are often accompanied by a number of undesirable consequences, including long-term disability and the early development of knee osteoarthritis, an increased risk of reinjury, and a reduced likelihood of returning to pre-injuries levels of sport or recreational activity (Ardern et al., 2015; Lohmander et al., 2004; Wright et al., 2007). Because of such consequences, the development of strategies aimed toward preventing the initial knee trauma continues to be a major research focus. In this regard, non-contact ACL injuries offer the greatest potential for injury prevention. Approximately two-thirds of all sport-related ACL injuries are noncontact in nature – in that they occur in the absence of physical contact with another player or object (Boden et al., 2000). Additionally, the incidence of noncontact ACL injury is considerably higher for female athletes compared to males (Arendt, E. & Dick, 1995; Beynnon, Vacek, et al., 2014). As such, there has been an ongoing effort to identify the neuromuscular and biomechanical characteristics that contribute to high knee-joint loads and ACL strain during sport-specific movement so that these factors can then be targeted via evidence-based injury-prevention strategies.

Noncontact ACL injuries most commonly occur as the relatively extended knee (< 30° flexion) initially transitions from non-weight bearing to weight bearing (i.e. initial ground contact) during athletic maneuvers (Boden et al., 2000; Koga et al., 2010; Krosshaug et al., 2007; Olsen et al., 2004). These maneuvers often involve a sharp deceleration, with or without a change of direction, such as when quickly cutting to evade an opponent or when landing from a jump on

a single leg. Although there is a general consensus that the mechanism(s) of injury is likely multiplanar (Shultz, S. J. et al., 2015), it is well accepted that the ACL is most directly loaded (strained) via proximal-tibia-anterior-shear force (PTASF) (Butler et al., 1980; Markolf et al., 1995). To this end, several measurable *in-vivo* neuromuscular and biomechanical characteristics have been shown to be correlated with PTASF, including neuromuscular activation of the quadriceps (Sell et al., 2007; Shultz, S. J. et al., 2009), sagittal-plane trunk and knee angles (Gheidi, Sadeghi, Moghadam, Tabatabaei, & Kernozek, 2014; Sell et al., 2007), sagittal-plane knee moments and angular velocities (Gheidi et al., 2014; Sell et al., 2007; Yu et al., 2006), and resultant ground reaction forces (Sell et al., 2007; Yu et al., 2006). In addition, the linear combination of preparatory neuromuscular activation of the vastus lateralis, peak posterior ground reaction force, knee-flexion angle and moment (external), and sex, has been demonstrated to predict a substantial proportion of the variance (86.1%) in peak PTASF during a deceleration task, with greater neuromuscular activation, ground reaction forces, knee angles, knee moments, and being female, predicting greater PTASF (Sell et al., 2007). Given that these characteristics were able to predict such a large proportion of the variance in PTASF, and that PTASF is a biomechanical indicator of ACL loading (Markolf et al., 1995), such characteristics have been studied by more recent investigations aimed at (1) prospectively identifying athletes who may potentially be at risk for future ACL injury, or (2) evaluating the effectiveness of current intervention strategies aimed at reducing knee loads and ACL strain (Chappell et al., 2002; Herman et al., 2009; Myers & Hawkins, 2010; Yu et al., 2006).

The aforementioned PTASF prediction model identified by Sell et al. (2007) is a noteworthy contribution to ACL literature; however, the findings of similar studies suggest that the characteristics that are predictive of PTASF, and the predictive ability (i.e. total proportion of variance explained) of such characteristics, may be dependent on the deceleration task used as a

model for injury, and the pool of possible predictor variables from which the prediction model is developed (Gheidi et al., 2014; Shultz, S. J. et al., 2009). Specifically, while Sell et al (2007) used a double-leg vertical stop-jump task to examine predictors of PTASF, Shultz et al. (2009) used a double-leg drop-vertical jump task and demonstrated that the linear combination of sex, hip- and knee-flexion excursion, peak knee-extension moment (internal), quadriceps and hamstring peak torque, and neuromuscular activation of the quadriceps and hamstrings pre- and post-landing, predicted 56.5% of the variance in PTASF. The model reported by Shultz et al (2009) indicated that, irrespective of sex, lesser hip-flexion excursions, greater knee-flexion excursions and kneeextension moments, and greater post-landing quadriceps activation, predicted greater PTASF. In another study, Gheidi et al (2014) used a single-leg drop landing task and reported that peak knee-extension moment (internal) and peak knee-flexion angle collectively predicted 30.6% of the variance in PTASF, with greater knee moments and lesser knee angles predicting greater PTASF. The distinction between tasks in these studies is important because both task and landing type (i.e. double- vs single-leg) have been shown to differentially affect neuromuscular and biomechanical outcome measures (Cruz et al., 2013; Pappas et al., 2007; Taylor et al., 2016; Wang, L. I., 2011). Given that noncontact ACL injuries are more likely to occur during singleversus double-leg jump landings (Boden et al., 2000; Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), and that they include both horizontal and vertical deceleration components, identifying neuromuscular and biomechanical predictors of PTASF during tasks that involve a single-leg jump landing with both horizontal and vertical components may be more representative of the factors that potentially contribute to noncontact ACL injury risk. Furthermore, recent studies indicate that other measurable *in-vivo* characteristics, such as hamstring musculo-articular stiffness and anterior knee laxity, have the ability to affect the resultant PTASF that an individual displays during dynamic landing tasks (Blackburn et al., 2013; Shultz, S. J., Schmitz, Nguyen, &

Levine, 2010b). Thus, understanding whether these additional measures are significant predictors of PTASF could help better inform future screening and injury prevention efforts.

The purpose of this study was to examine the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics (i.e. preparatory neuromuscular activation of the medial and lateral quadriceps and hamstrings, peak posterior ground reaction force, knee-flexion angle and knee-extension moment at the instant of peak posterior ground reaction force, hamstring stiffness, and anterior knee laxity) are able to collectively predict PTASF during a single-leg vertical stop-jump task. Based on the previous prediction model identified by Sell et al (2007), we hypothesized that the linear combination of sex, preparatory quadriceps activation, posterior ground reaction force, knee-flexion angle, and knee-extension moment, would be able to significantly predict PTASF during the single-leg vertical stop-jump. We also hypothesized that hamstring musculo-articular stiffness and anterior knee laxity would predict an additional proportion of the variance in PTASF in the final prediction model.

Methods

Participants

Eighty healthy men (n = 40) and women (n = 40) volunteered to participate in this study. At the time of testing, all participants were physically active, in that they regularly engaged in greater than the equivalent of 300 minutes of moderate-intensity physical activity per week (assessed via the International Physical Activity Questionnaire, Appendix B), and participated in activities that involved running, cutting, jumping and landing (assessed via the Marx Activity Rating Scale, Appendix B). Exclusion criteria for this study included any history of: (1) knee ligamentous or meniscal injury, (2) lower-extremity surgery, (3) lower-extremity injury within 6 months of testing, (4) medical conditions that could affect the connective tissue, and (5)

vestibular system disorder diagnoses. This study was approved by the university's Institutional Review Board for the Protection of Human Subjects prior to recruitment, and written informed consent was obtained from each participant prior to testing. Each participant received \$10 compensation for their participation in this study.

Procedures

All data were collected during a single testing session. In order to control for any potential effects of menstrual cycle hormones on knee-joint biomechanics (Park, Stefanyshyn, Ramage, et al., 2009; Shultz, S. J. et al., 2012; Shultz, S. J. et al., 2011), hamstring stiffness (Bell, D. R. et al., 2012), and anterior knee laxity (Park, Stefanyshyn, Loitz-Ramage, Hart, & Ronsky, 2009; Shultz, S. J., Carcia, & Perrin, 2004; Shultz, S. J., Gansneder, Sander, Kirk, & Perrin, 2006; Shultz, Sandra J., Kirk, Johnson, Sander, & Perrin, 2004), all female participants underwent testing during the follicular phase of their menstrual cycle (i.e. days 1-8 following selfreported onset of menstrual bleeding). Upon arrival to the laboratory, participants were outfitted with compression shorts and a tight-fitting athletic top. After barefoot measures of body height and mass were obtained, the remainder of the testing session was then performed in the following order: (1) anterior knee-joint laxity assessment, (2) five-minute warm-up, (3) quadriceps and hamstring maximal voluntary isometric contraction testing, (4) hamstring musculo-articular stiffness assessment, and (5) stop-jump landing biomechanics. The warm-up was completed on a stationary cycle ergometer (Life Fitness, Schiller Park, IL) at a cadence of 70-80 RPM and a target rating of perceived exertion of \geq 3-4 on a Borg CR-10 RPE scale (Borg, 1998). All testing was performed on the left leg, which corresponded with the dominant leg (self-reported stance leg when kicking a ball) in 68 of our 80 participants.

Anterior Knee-Joint Laxity Assessment. Anterior knee laxity (AKL) – defined as the anterior displacement (mm) of the tibia relative to the femur when subjected to an anteriordirected force of 133 N – was assessed using an instrumented knee arthrometer (KT-2000TM; MEDmetric® Corp; San Diego, CA, USA). Participants were positioned supine with: (1) the thighs supported by a bolster placed just proximal to the popliteal fossa, (2) the knees flexed to $25^{\circ} \pm 5^{\circ}$, (3) the foot and angle neutrally aligned in a manufacturer-provided foot cradle, and (4) a strap secured around the thighs to prevent any lower-extremity rotation. Once positioned, the arthrometer was secured to the tibia in alignment with the medial and lateral joint lines of the knee. With the participant relaxed, a stable neutral joint position was then obtained by applying three anterior- to posterior-directed forces at proximal tibia. Next, an anterior-directed force of 133 N was applied to the posterior tibia, and AKL was measured to the nearest half-mm. A bubble level was affixed to the arthrometer to ensure that an anterior-directed force was achieved. A total of 3 trials were recorded and subsequently averaged for use in statistical analyses. All AKL assessments were performed by a single investigator who had previously established good intra-rater reliability (ICC_{2,3} = 0.83) and measurement precision (SEM = 0.25 mm) using the methods described.

Maximal Voluntary Isometric Contraction Testing. Maximal voluntary isometric contraction (MVIC) testing was performed for surface electromyography (sEMG) normalization purposes (see data sampling and reduction), and for determining each participant's loading assignment for the hamstring musculo-articular stiffness assessment (see stiffness methods located below). Prior to MVIC testing, participants were instrumented with wireless sEMG sensors (Delsys Trigno; Delsys Inc., Boston, MA, USA) placed over the muscle bellies of the medial and lateral quadriceps (vastus medialis and lateralis, respectively) and hamstring (semitendinosus/semimembranosus and biceps femoris long-head, respectively) muscles using

double-sided adhesive. To reduce impedance, sensor sites were shaved using a disposable razor, and cleaned with isopropyl alcohol, prior to sensor placement. Once sensor placement was confirmed via standardized manual muscle testing, cohesive athletic tape was wrapped around the thigh to minimize movement artifact.

Quadriceps and hamstring MVIC testing was performed using a Biodex System 3 isokinetic dynamometer (Biodex Medical Systems, Shirley, NY, USA). Quadriceps MVIC testing was performed with participants positioned supine and the hip and knee fixed in 30° of flexion (Figure 6.1A). Hamstring MVIC testing was performed with participants positioned prone and the hip and knee fixed in 30° of flexion (Figure 6.1B). To ensure a consistent body position, straps were secured across the torso, hips, thigh, and distal shank. Participants then completed 4 practice trials (25%, 50%, 75%, and 100% of self-perceived maximal effort) followed by 3 test trials during which sEMG and peak isometric torque data were recorded. All MVIC trials were held for 5 seconds, and 60-second rest intervals were provided between trials to minimize the risk of fatigue. In addition, participants were provided verbal encouragement throughout testing to ensure performance consistency across trials. The testing order was identical for all participants (quadriceps testing performed first).

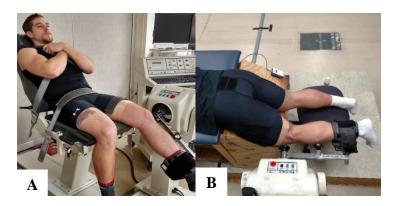


Figure 6.1. Participant Positioning During Maximal Voluntary Isometric Contraction (MVIC) Assessments for the Quadriceps (A) and Hamstrings (B).

Hamstring Musculo-Articular Stiffness Assessment. Hamstring stiffness (K_{HAM}) was assessed via the free-oscillation technique using methods previously described in detail (Waxman et al., 2015). Briefly, participants were positioned prone on a padded table, with the trunk and thigh supported in 30° of hip flexion, and the lower leg and foot segment free to move (Figure 6.2). Participants were then instrumented with a twin-axis electrogoniometer (Biometrics Ltd., Ladysmith, VA) secured to the lateral aspect of the knee joint (Figure 6.2A), a thermoplastic splint and standardized load equal to 30% of mean peak isometric hamstring torque (obtained from hamstring MVIC testing) secured to the distal shank and foot segment (Figure 6.2B), and a triaxial accelerometer (Sensor dimensions: 2.54 x 2.54 x 1.91 cm; NeuwGhent Technology, USA) attached to the thermoplastic splint (Figure 6.2C). Our decision to standardize the load to 30% MVIC was based on a previous study which reported mean hamstring activation amplitudes of ~30% MVIC during the stance phase of gait activities (Ciccotti et al., 1994). Once instrumented, the shank was passively positioned so that the knee was flexed approximately 30°, and the participant was instructed to maintain this position via isometric hamstring contraction. During this time, real-time knee joint angle data were displayed on a monitor, giving participants a visual target to maintain (Figure 6.2D). Within 5 seconds of the participant holding this position, a brief downward perturbation was manually applied to the posterior aspect of the calcaneus, resulting in slight knee extension and subsequent damped oscillatory flexionextension. This damped oscillatory motion was then characterized as the tangential acceleration of the shank and foot segment, and captured via the triaxial accelerometer. Participants were verbally instructed not to interfere with or voluntarily produce the oscillations following the perturbation, and to attempt to keep the hamstring muscles active only to the level necessary to support the mass of the shank and foot segment, and the applied load, in the testing position (Blackburn et al., 2013; Waxman et al., 2015). Each participant performed 3-5 practice trials,

followed by 5 test trials during which data were recorded. Trials were separated by 30-second rest intervals to minimize the risk of fatigue.

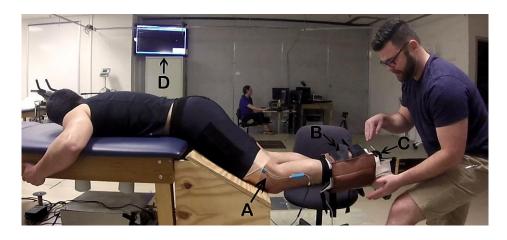


Figure 6.2. Participant Positioning and Instrumentation for the Hamstring Musculo-Articular Stiffness (K_{HAM}) Assessment. Electrogoniometer placement (A); Thermoplastic splint & ankle weights (B); Accelerometer (C); Monitor displaying real-time knee-flexion angle data (D).

Stop-Jump Landing Biomechanics. Landing biomechanics were assessed during the performance of a single-leg vertical stop-jump (SLSJ) task using an 8-camera IMPULSE motion tracking system (Phase Space, San Leandro, CA) and an integrated non-conducting force platform (Type 4060-130; Bertec Corporation., Columbus, OH, USA). Standardized athletic shoes (Adidas, Uraha 2, Adidas North America, Portland, OR, USA) were worn by all participants in order to experimentally control for any potential effects of footwear on landing biomechanics. Participants were then instrumented with optical LED marker clusters (4 markers per cluster; Phase Space, San Leandro, CA, USA) secured to the foot, shank, thigh, pelvis, and trunk (Figure 3). Once instrumented, participants were digitized using MotionMonitor software (Innovative Sports Training, Chicago, IL). Ankle and knee joint centers were determined as the midpoint between the medial and lateral malleoli and medial and lateral femoral epicondyles, respectively. Hip joint centers were determined using the Bell method (Bell, A. L. et al., 1989).

The SLSJ task used in the current study was performed in strict accordance with the methods previously described by Sell et al (2007), with the only exception being that our task was performed on a single leg. The SLSJ consisted of the following: (1) an initial starting position set at a distance equal to 40% of the participant's height behind the rear edge of the force platform; (2) a single-leg broad jump from the starting position, followed by a single-leg landing on the force platform; (3) an immediate single-leg jump for maximum vertical height upon landing; and (4) a secondary single-leg landing on the force platform following the vertical jump (Figure 6.3). To promote performance consistency across trials, and across participants, the following verbal instructions were provided: (1) "starting on your left leg, jump towards the center of the force platform and land on the same leg"; (2) "upon landing, jump straight up into the air as high as you can, and then land again on the same leg". Participants were allowed to use their arms during the task; however, they were instructed to keep their elbows in approximately 90° of flexion in order to minimize marker obstruction. In an effort to prevent any experimenter bias, the investigators did not provide participants with any special instructions regarding their landing biomechanics. All participants were allowed practice trials until they became comfortable with the task (approximately 3-5 trials). Once comfortable, participants performed 5 test trials during which data were recorded. Thirty-second rest intervals were provided between trials to minimize the risk of fatigue. Trials were considered successful if the participant initiated the trial from the proper starting distance, landed on the force platform, jumped for maximum vertical height upon landing, and landed back on the force platform following the vertical jump. Unsuccessful trials were discarded and repeated.

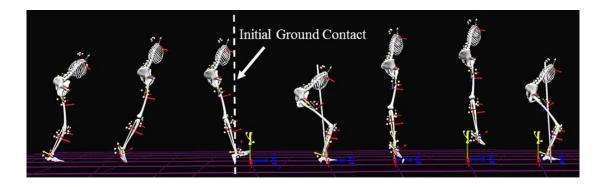


Figure 6.3. Visual Depiction of the Single-Leg Stop-Jump (SLSJ) Landing Task.

Data Sampling and Reduction

Hamstring Musculo-Articular Stiffness. Accelerometer data were sampled at 1000 Hz and collected using MotionMonitor software (Innovative Sports Training Inc., Chicago, IL). These data were then low-pass filtered at 10 Hz, using a fourth-order zero-lag Butterworth filter, and subsequently exported to Matlab (Mathworks, Inc., Natick, MA) for data reduction using a custom-written program. Within Matlab, the time-interval between the first two oscillatory peaks of the accelerometer time series was identified; this time interval was then used to calculate the damped frequency of oscillation for each of the 5 K_{HAM} trials. Once these frequencies were obtained, K_{HAM} was calculated using the equation: $K_{\text{HAM}} = 4\pi^2 m f^2$, where where m is the summed mass of the lower-leg and foot segment (6.1% body mass) (Winter, 1990) and the applied load (30%MVIC), and f is the damped frequency of oscillation. Because K_{HAM} is influenced by anthropometrics (Granata et al., 2002), these values were normalized to body mass (N·m⁻¹·kg⁻¹), and the average of 5 trials was then calculated for use in statistical analyses.

Stop-Jump Landing Biomechanics. Kinetic, sEMG, and kinematic hardware were integrated and time-synchronized with MotionMonitor software for data collection. Kinetic and sEMG data were sampled at 1000 Hz, whereas kinematic data were sampled at 240 Hz and subsequently linearly interpolated to 1000 Hz within MotionMonitor. Quadriceps and hamstring

sEMG data obtained during MVIC testing and the SLSJ task were band-pass filtered from 10 Hz to 350 Hz using a fourth-order zero-lag Butterworth filter, and subsequently processed using centered root mean square (RMS) algorithms with 25- and 100-millisecond time constants, respectively. Kinetic and kinematic data were low-pass filtered at 12 Hz using a fourth-order zero-lag Butterworth filter, whereas peak ground reaction force data were low-pass filtered at 60 Hz. A segmental reference system was defined for all body segments, with the z-axis as the medial-lateral axis (flexion-extension), the y-axis as the longitudinal axis (internal-external rotation), and the x-axis as the anterior-posterior axis (abduction-adduction). Joint motions were then calculated within MotionMonitor using Euler angle definitions with a rotational sequence of Z Y' X" (Kadaba et al., 1989). Joint moments and PTASF were calculated within MotionMonitor using inverse dynamics (Gagnon & Gagnon, 1992). All data were later exported to Matlab for data reduction using a custom-written program.

Within Matlab, the RMS sEMG data recorded from each muscle during each SLSJ trial were normalized to the mean peak RMS sEMG amplitude recorded from each respective muscle during MVIC testing (%MVIC). All joint moment data were normalized to the product of each participant's body weight and body height (BW-1·Ht-1), and all force data were normalized to body weight (BW). Following data filtering and normalization, all neuromuscular and biomechanical variables of interest were then extracted. Specifically, neuromuscular variables of interest included preparatory neuromuscular activation of the medial and lateral quadriceps (MQUAD_{PRE} and LQUAD_{PRE}, respectively), and medial and lateral hamstrings (MHAM_{PRE} and LHAM_{PRE}, respectively). Preparatory neuromuscular activation was defined as the normalized mean RMS sEMG amplitude obtained over a 150-millisecond time interval prior to initial ground contact – the instant at which the vertical ground reaction force first exceeded 10 N.

Biomechanical variables of interest included peak posterior ground reaction force (pGRF_{PK}), and

PTASF, knee-flexion angle, and knee-extension moment (internal) at the instant of pGRF_{PK}. Extracting these variables at the instant of pGRF_{PK} was done in order to stay consistent with the variables previously used by Sell et al (2007) to predict PTASF. All variables were averaged across 5 trials for use in statistical analyses.

Statistical Analyses

All statistical analyses were performed in SPSS (Version 23; IBM Corp., Armonk, NY, USA). A forward, stepwise, multiple linear regression analysis was conducted in order to determine which anatomical, neuromuscular, and biomechanical variables could significantly predict PTASF at the time of maximum deceleration (i.e. pGRF_{PK}). The predictor variables included preparatory activation of the quadriceps and hamstring muscles (i.e. MQUAD_{PRE}, LQUAD_{PRE}, MHAM_{PRE}, and LHAM_{PRE}), pGRF_{PK}, knee-flexion angle at pGRF_{PK}, knee-extension moment at pGRF_{PK}, AKL, *K*_{HAM}, and sex. The criterion (dependent) variable was PTASF at the time of maximum deceleration. Prior to conducting the regression analysis, independent samples *t*-tests compared males and females on all variables of interest in order to evaluate whether there was a potential need for sex-stratified models. Bivariate correlations were also performed in order to examine relationships between the criterion variable and all predictor variables. In all analyses, the a-priori alpha was set at 0.05 to denote statistical significance. Based on a sample size of 80 participants, and maximum of 10 possible predictor variables, we determined that we had over 90% power to detect a multiple R² of 0.25 (Cohen, 1988).

Results

Although 80 participants (40 males, 40 females) completed all testing procedures, six participants (3 males, 3 females) were excluded from analysis due to having insufficient data on

one or more variables of interest. Specifically, two participants were excluded because they were unable to support the weight of the applied load during the K_{HAM} assessment, another two participants were excluded because they were unable to successfully meet performance requirements of the SLSJ, and the last two participants were excluded because of technical problems associated with the acquisition of sEMG data. Thus, our statistical analyses were conducted on a total sample size of 74 participants, which consisted of 37 males and 37 females. Participant demographics are presented in Table 6.1.

Table 6.1. Participant Descriptive Statistics. All Values are Presented as Mean \pm SD.

Variable	Total $(n = 74)$	Male $(n = 37)$	Female $(n = 37)$
Age (years)	21.3 ± 2.0	21.5 ± 2.1	21.2 ± 1.9
Height (cm)	174.5 ± 11.3	182.6 ± 6.9	166.5 ± 8.9
Mass (kg)	73.7 ± 15.9	82.4 ± 13.9	64.9 ± 12.7

Descriptive statistics (mean \pm SD) for all variables of interest are presented in Table 6.2. The results from the independent samples t-tests did not reveal any statistically significant differences between males and females for any of the variables examined (P-value range = .063-.978; Table 6.2). Thus, our decision to include males and females in the same regression analysis was justified.

Bivariate correlations between PTASF and each possible predictor variable are presented in Table 6.3. Proximal tibia anterior shear force (PTASF) was moderately correlated with with pGRF_{PK} (r = .353, P = .001), and strongly correlated with knee-flexion angle at pGRF_{PK} (r = .846, P < .001) and knee-extension moment at pGRF_{PK} (r = -.824, P < .001). The results from the stepwise multiple linear regression model are presented in Table 6.4. From this analysis, it was found that only 3 of the 10 possible predictor variables entered into the final prediction model.

Specifically, the regression analysis revealed that the combination of knee-flexion angle at pGRF_{PK}, knee-extension moment at pGRF_{PK}, and LQUAD_{PRE} collectively explained 78.4% ($R^2 = .784$, P < .001) of the variance in PTASF during the SLSJ task (Table 6.3). The parameter estimates for each of the individual predictors indicate that greater knee-flexion angles (P < .001) and knee-extensor moments (P < .001) at pGRF_{PK}, and lesser LQUAD_{PRE} (P = .044), predicted greater magnitudes of PTASF (Table 6.3).

Table 6.2. Means and Standard Deviations (mean \pm SD) for all Neuromuscular and Biomechanical Variables.

Variable	Total (n = 74)	Males (n = 37)	Females (n = 37)	<i>P</i> -value
pGRF _{PK} (BW)	-0.75 ± 0.16	-0.78 ± 0.18	-0.72 ± 0.14	.109
PTASF @ pGRF _{PK} (BW)	0.27 ± 0.28	0.25 ± 0.30	0.29 ± 0.27	.599
KFA @ pGRF _{PK} (°)	17.93 ± 10.52	17.97 ± 11.10	17.90 ± 10.05	.978
KEM @ pGRF _{PK} (BW ⁻¹ ·Ht ⁻¹)	-0.026 ± 0.048	-0.025 ± 0.051	-0.027 ± 0.046	.914
MQUAD _{PRE} (%MVIC)	25.07 ± 27.23	30.63 ± 31.97	19.51 ± 20.45	.079
LQUAD _{PRE} (%MVIC)	29.27 ± 25.33	33.94 ± 27.00	24.60 ± 22.94	.113
MHAM _{PRE} (% MVIC)	13.912 ± 8.47	12.82 ± 9.05	15.01 ± 7.82	.267
LHAM _{PRE} (%MVIC)	12.49 ± 15.82	10.55 ± 6.89	14.43 ± 21.26	.295
AKL (mm)	7.53 ± 2.50	7.52 ± 2.69	7.54 ± 2.33	.974
$K_{\text{HAM}} (\text{N} \cdot \text{m}^{-1} \cdot \text{kg}^{-1})$	12.66 ± 3.51	13.41 ± 3.82	11.90 ± 3.02	.063

Note. pGRF_{PK} = peak posterior ground reaction force; PTASF = proximal tibia anterior shear force; KFA = knee-flexion angle; KEM = knee-extension moment; MQUAD_{PRE} = preparatory activation of medial quadriceps; LQUAD_{PRE} = preparatory activation of lateral quadriceps; MHAM_{PRE} = preparatory activation of medial hamstring; LHAM_{PRE} = preparatory activation of medial hamstring; AKL = anterior knee laxity; K_{HAM} = hamstring musculo-articular stiffness.

Table 6.3. Bivariate Correlations Between the Criterion Variable (PTASF) and the Predictor Variables.

	r	P-value
pGRF _{PK} (BW)	.353	.001*
Knee-flexion angle @ pGRF _{PK} (°)	.846	<.001*
Knee-extension moment @ pGRF _{PK} (BW ⁻¹ ·Ht ⁻¹)	824	<.001*
MQUAD _{PRE} (%MVIC)	.055	.322
LQUAD _{PRE} (%MVIC)	037	.378
MHAM _{PRE} (%MVIC)	.013	.456
LHAM _{PRE} (%MVIC)	.077	.259
AKL (mm)	107	.183
K_{HAM} (N·m ⁻¹ ·kg ⁻¹)	.164	.081
Sex	.062	.300

Note. pGRF_{PK} = peak posterior ground reaction force; PTASF = proximal tibia anterior shear force; $MQUAD_{PRE}$ = preparatory activation of medial quadriceps; $LQUAD_{PRE}$ = preparatory activation of lateral quadriceps; $MHAM_{PRE}$ = preparatory activation of medial hamstring; $LHAM_{PRE}$ = preparatory activation of medial hamstring; AKL = anterior knee laxity; K_{HAM} = hamstring musculo-articular stiffness.

^{*} Denotes a statistically significant correlation ($P \le .05$)

Table 6.4. Multiple Linear Regression Model Predicting Proximal Tibia Anterior Shear Force (PTASF).

Multipl	le Linear Regression Model				
Source	SS	df	MS	Observations	74
Model	4.553	3	1.518	F(3, 70)	84.46
Residual	1.258	70	0.018	Prob > F	P < .001
Total	5.811	73		R^2	0.784
				R^2 (Adjusted)	.774

		Unstandardized					Correlations		
			Std.	_			Zero-		
	Predictor Variables	В	Error	β	t	P- value	order	Partial	Part
17	Constant	0.006	0.044		0.125	.901			
7	Knee-flexion angle at pGRF _{PK}	0.013	0.003	0.491	5.097	< .001	0.846	0.52	0.283
	Knee-extension moment at pGRF _{PK}	-2.588	0.571	-0.441	-4.532	< .001	-0.824	-0.476	-0.252
	$LQUAD_{PRE}$	-0.001	0.001	-0.116	-2.049	.044	-0.037	-0.238	-0.114

Note. $pGRF_{PK}$ = peak posterior ground reaction force; $LQUAD_{PRE}$ = preparatory activation of lateral quadriceps.

Discussion

The purpose of this study was to conduct a neuromuscular and biomechanical analysis of males and females during the performance of a single-leg stop-jump (SLSJ), and then examine the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics were able to collectively predict PTASF. The impetus for this investigation was a previous study by Sell et al (2007), which demonstrated that preparatory neuromuscular activation of the lateral quadriceps (LQUAD_{PRE}), peak posterior ground reaction force (pGRF_{PK}), knee-flexion angle and moment (external) at the instant of pGRF_{PK}, and sex, collectively predicted 86.1% of the variance in proximal tibia anterior shear force (PTASF) during a double-leg stop-jump (DLSJ). Because noncontact ACL injuries are more likely to occur when landing from a jump on a single leg (Boden et al., 2000; Boden et al., 2009; Koga et al., 2010; Olsen et al., 2004), we ultimately wanted to determine whether the characteristics that are predictive of PTASF during the DLSJ are similarly predictive of PTASF during the SLSJ.

Our primary research hypothesis was that LQUAD_{PRE}, pGRF_{PK}, knee-flexion angle and knee-extension moment (internal) at the instant of pGRF_{PK}, and sex, would be able to collectively predict PTASF during the SLSJ; and that AKL and *K*_{HAM} would explain additional variance in the final prediction model. In partial support of this hypothesis, the main finding of this study was that the linear combination of LQUAD_{PRE}, knee-flexion angle, and knee-extension moment, significantly predicted 78.4% of the variance in PTASF, with lesser LQUAD_{PRE}, and greater knee angles and moments, being predictive of greater PTASF (Table 6.4). While this is the first investigation to our knowledge to examine predictors of PTASF during a SLSJ, our prediction model in large part agrees with the previous prediction model reported by Sell et al. (2007) during a DLSJ. Thus, the characteristics that are predictive of PTASF when performing a stop-jump task on both legs appear to be similarly predictive of PTASF when performing the same task on a

single leg. Our findings are also in general agreement with prediction models that that have been developed using other sagittal-plane landing tasks. During a double-leg drop-vertical jump, for example, Shultz et al (2009) demonstrated that sex, hip- and knee-flexion excursion, peak knee-extension moment (internal), quadriceps and hamstring peak torque, and neuromuscular activation of the quadriceps and hamstrings pre- and post-landing, predicted 56.5% of the variance in PTASF. In addition, Gheidi et al (2014) demonstrated that peak knee-flexion angle and knee-extension moment collectively predicted 30.6% of the variance in PTASF during a single-leg drop landing. Although we are unable to directly compare these findings to those of the current study due to differences in the tasks used, and the predictor variables examined, this previous work helps highlight that quadriceps activation, and sagittal-plane knee angles and moments, are predictive of PTASF across a variety of landing tasks.

The PTASF values calculated during the SLSJ in the current investigation (Table 6.2) are similar to those previously reported by Sell et al (2007) during a DLSJ. Based on a prior study (Wang, L. I., 2011) investigating differences in PTASF between double- and single-leg stop-vertical jumps, we expected that our values would have been larger than those of Sell et al (2007); however, we are unaware of any other work to report PTASF values at the instant of pGRF_{PK}. We chose PTASF as our criterion (dependent) variable because it represents the most direct loading mechanism of the ACL (Butler et al., 1980; Markolf et al., 1995), and because it can be estimated *in-vivo* via inverse dynamics. Before discussing the potential implications of our findings, it is important to note that PTASF, as calculated in the current study, represents the total net force acting at the knee joint; it does not represent the shear force experienced by the ACL, or the shear force applied by the patellar tendon, and therefore is not a direct measure of ACL loading. However, cadaveric studies and musculoskeletal modeling simulations have shown that increases in PTASF are associated with increases in anterior tibial translation, thereby loading the

ACL (Shelburne, Pandy, Anderson, & Torry, 2004; Shelburne, Pandy, & Torry, 2004). Therefore, *in-vivo* studies often rely on PTASF as a biomechanical indicator of ACL loading (Blackburn et al., 2013; Chappell et al., 2002; Gheidi et al., 2014; Sell et al., 2007; Shultz, S. J. et al., 2009; Wang, L. I., 2011; Yu et al., 2006). To this end, PTASF has been proposed as a factor that potentially contributes to females' increased risk for noncontact ACL injury since controlled laboratory studies have observed that females perform dynamic landing tasks with significantly greater PTASF compared to their similarly trained male counterparts (Chappell et al., 2002; Yu et al., 2006). Interestingly, however, both the data of Sell et al (2007) and that of the current study have been unable to replicate these findings.

The parameter estimates for our prediction model indicate that, if all other predictors were held constant, a 1 unit increase knee-extension moment (value becoming more negative) would lead to a 2.59 unit increase in PTASF (Table 6.4). Both the direction and magnitude of this relationship between knee-extension moment and PTASF was expected given that patellar-tendon force has been demonstrated to be a major contributor to PTASF (Laughlin et al., 2011). This finding is also in agreement with the previous PTASF prediction models reported by Sell et al (2007) and Shultz et al (2009). Mechanistically, the ground reaction forces produced upon landing create a flexion moment relative to the knee, which needs to be balanced by quadricepsgenerated knee-extension moment to stabilize the knee and prevent lower-extremity collapse (McNitt-Gray, 1993; Yu et al., 2006). At more extended knee angles (< 30° flexion), contraction of the quadriceps generates PTASF because the patellar tendon's line of action is directed anteriorly with respect to the long axis of the tibia (Draganich, Andriacchi, & Andersson, 1987; Herzog & Read, 1993); and *in-vitro* and *in-vitro* studies have shown that these quadriceps muscle forces are capable of loading the ACL (Beynnon et al., 1995; DeMorat et al., 2004; Li et al., 1999; Withrow et al., 2006).

Participants in the current study were positioned in approximately $18^{\circ} \pm 11^{\circ}$ of knee flexion at the instant of pGRF_{PK} (Table 6.2). Given the mechanistic relationship between ground reaction forces, knee-extension moment, knee-flexion angle, and PTASF, when the knee is closer to extension, we expected that lesser knee flexion and greater quadriceps activation would have predicted greater PTASF. However, our findings contradicted this expectation. The unstandardized regression coefficients for our prediction model indicate that, if all other predictors were held constant, a 1 unit increase in knee-flexion angle at pGRF_{PK} (greater knee flexion) would lead to a 0.013 unit increase in PTASF, whereas a 1 unit increase in LQUAD_{PRE} would lead to a 0.001 unit decrease in PTASF (Table 6.4). As suggested by Sell et al. (2007), the contradictory evidence between knee angle and PTASF in this study may be due to the lack of a clearly established relationship between ACL loading and PTASF during dynamic landing tasks, as many of these studies have examined ACL loading with the knee in a fixed position(s) (Fleming, Renstrom, Beynnon, et al., 2001; Markolf et al., 1995). In addition, although landing with smaller knee-flexion angles, increased quadriceps activation, and higher knee-extension moments and PTASF, have been proposed as noncontact ACL injury risk factors, this theory has been largely based on observed differences in such characteristics between males and females (Chappell et al., 2002; Yu et al., 2006). In this regard, we are only aware of two previous studies (Sell et al., 2007; Shultz, S. J. et al., 2009) that have collectively examined neuromuscular activation along with kinematic and kinetic data obtained during dynamic landing tasks in order to directly make the connection between quadriceps activation, knee flexion angles, kneeextension moments, and PTASF. In combination with this prior work, our findings lend support to the theory that greater knee-extensor moments may increase injury risk due to the associated increase in PTASF. In contrast, however, this is now the third prediction model to indicate that greater knee flexion would actually predict greater PTASF (Sell et al., 2007; Shultz, S. J. et al.,

2009). This is not to suggest that landing with smaller amounts of knee flexion may not be a risk factor for injury, but that additional research examining the relationship between knee-flexion angle, PTASF, and ACL loading, during functional landing tasks is warranted.

Our finding of lesser LQUAD_{PRE} predicting greater PTASF directly opposes that of Sell et al (2007); however, this is not the first study to report an inverse relationship between preparatory neuromuscular activation of the quadriceps and PTASF during a dynamic landing task. Specifically, Shultz et al (2009) demonstrated that a decrease in normalized preparatory neuromuscular activation amplitude of the medial and lateral quadriceps would predict an increase in PTASF during a double-leg drop-vertical jump. In an effort to better understand this relationship, we performed follow-up sex-stratified stepwise regression analyses. Interestingly, when these prediction models were run separately for males and females, LQUAD_{PRE} did not enter into the model for either sex. Instead, the linear combination of only knee-extension moment and knee-flexion angle at the instant of pGRF_{PK} significantly explained 78.4% and 77.5% of the variance in PTASF for males and females, respectively. This suggests that our finding of lesser LQUAD_{PRE} significantly predicting greater PTASF may have been erroneously caused by between-sex differences. To this end, LQUAD_{PRE} was found to be somewhat correlated with sex (r = -0.19, P = .057). Alternatively, landing with lesser LQUAD_{PRE} may have resulted in a less stable joint at initial ground contact, which could potentially increase the anterior acceleration of the tibia following initial ground contact, and thereby result in increased PTASF. Further, this relationship may have been influenced by the combined positioning of the trunk, hip, and knee joints during landing. Specifically, in situations where the trunk is upright or leaning backwards at initial ground contact, it has been hypothesized that the body's center or mass (CoM) would be positioned posterior to the knee joint, and result in greater knee flexion than hip flexion, and ultimately cause the tibia to translate anteriorly due to greater PTASF (Hashemi et

al., 2011). Although we did not include hip and trunk biomechanical variables in this study, the contributions of trunk and hip biomechanics in predicting PTASF deserve consideration in future work.

While our primary research hypothesis was based on the predictors of PTASF previously identified by Sell et al (2007), we also hypothesized that anterior knee laxity (AKL) and hamstring musculo-articular stiffness (K_{HAM}) would significantly predict an additional proportion of the variance in the final model. Our decision to include these variables in the pool of potential predictor variables was based on previous work that has reported these variables to play a role in sagittal plane knee-joint loading. For example, higher amounts of AKL have been shown to associated with higher knee-extension moments and peak knee-flexion angles, and decreased preparatory neuromuscular activation of the hamstrings, during a double-leg drop-vertical jump (Shultz, S. J. et al., 2010a). Similarly, individuals with higher K_{HAM} have been shown to display less anterior tibial translation during controlled perturbations (Blackburn et al., 2011), and less PTASF and greater knee-flexion angles during double-leg jump landings (Blackburn et al., 2013), compared to individuals with lower K_{HAM} values. Thus, although AKL and K_{HAM} were not able to explain an additional proportion of the variance in PTASF in the current study, these variables may have potentially influenced the predictors that entered into our final prediction model (i.e. knee angle and moment, and LQUAD_{PRE}).

Limitations

We acknowledge that this study is not without limitations. First and foremost, this study was based on the assumption that PTASF is a biomechanical indicator of ACL loading.

Therefore, caution should be taken when considering the implications of our findings as they relate to noncontact ACL injury risk. Second, noncontact ACL injuries most commonly occur in athletic populations between 15 and 25 years of age (Griffin et al., 2000). In contrast, the

participants included in our sample were healthy, physically-active, college-aged men and women, who regularly participated in activities that involved running, cutting, jumping and landing (e.g. basketball, soccer, tennis, rugby, and volleyball). Thus, the results of this study are most generalizable to this type of population. Third, this study was performed in a controlled laboratory setting, which may elicit different neuromuscular and biomechanical characteristics than what might be observed in a more game- or practice-like setting. Finally, while the combination of lateral quadriceps activation, knee-flexion angle, and knee-extension moment, significantly accounted for 78.4% of the variance in PTASF, the remaining 21.6% of the variance in PTASF could not be explained by our prediction model. Hence, other factors that were not examined in this investigation likely contributed to the variance in PTASF during the SLSJ task. We chose to investigate only kinematics and kinetics at the knee joint in order to stay consistent with the methods previously used by Sell et al (2007) and maintain statistical power; however, we acknowledge that the knee is not an isolated joint, but rather a single part of the body's kinetic chain. That said, the proximal (i.e. trunk and hip) and distal (i.e. ankle) segments of the kinetic chain have previously been shown to have significant effects on knee-joint biomechanics (Griffin et al., 2006; Hewett, Ford, & Myer, 2006). Thus, some of the unexplained variance in PTASF could likely be accounted for by including contributions from the trunk, hip, and ankle, in future work. This is also true from a neuromuscular standpoint since the gastrocnemius, rectus femoris, and gluteal, muscles have also been shown to influence knee-joint biomechanics (Fleming, Renstrom, Ohlen, et al., 2001; Homan, Norcross, Goerger, Prentice, & Blackburn, 2013; McLean, Scott G., Borotikar, & Lucey, 2010; Wojtys, Wylie, & Huston, 1996). Furthermore, the passive restraint system (i.e. ligaments, menisci, and surrounding tissue) and bony joint geometry (e.g. the slope of the tibial plateau, joint congruency, etc.) have previously been demonstrated to influence the loading response of the knee (Beynnon, Hall, et al., 2014; McLean, S. G. et al.,

2011). Therefore, these factors should be considered when designing studies to further explore the characteristics that are predictive of PTASF and ACL loading in future work.

Conclusion

The findings of this study indicate that greater amounts of knee-flexion, higher knee-extension moments, and lesser LQUAD_{PRE}, would all predict an increase in PTASF when landing on a single leg, potentially increasing the forces experienced by the ACL. These findings are in general support of previous studies investigating predictors of PTASF during double-leg jump landing tasks, indicating that preparatory quadriceps activation, knee angle, and knee moment, are predictive of PTASF across a variety of landing tasks. While additional work is needed to better understand the relationship between PTASF and ACL loading in-vivo during dynamic tasks, the fact that this is now the third study to show that preparatory quadriceps activation, knee angle, and knee moment, are predictive of PTASF, suggests that these variables should be targeted by injury prevention efforts aimed at reducing sagittal plane knee-joint loading. Additionally, we encourage these variables to be used as potential predictor variables in future studies aimed at prospectively identifying risk factors for noncontact ACL injury, or as outcome variables in future studies examining adaptations elicited by current injury prevention efforts.

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CHAPTER VII

EXECUTIVE SUMMARY

Noncontact ACL injuries commonly occur as the relatively extended knee (< 30° flexion) initially transitions from non-weight bearing to weight bearing following initial ground contact during cutting and jump-landing maneuvers. Although these injuries likely result from multiplanar knee-joint loading, it is well accepted that the ACL is most directly loaded via sagittal-plane biomechanics, such as impact-induced anterior tibial acceleration (ATA), proximal tibia anterior shear force (PTASF), and anterior tibial translation (ATT). As such, any factors capable of effectively resisting these ACL-loading characteristics could theoretically help protect the ACL from deleterious loading and reduce noncontact ACL injury risk. In this regard, a property of the hamstring muscle group that may play a critical role in helping resist sagittal plane ACL-loading characteristics is musculo-articular stiffness.

Hamstring musculo-articular stiffness is a neuromechanical property that simply describes the resistance of the hamstring muscle-tendon unit to lengthening in response to an applied load. As such, it is theorized that, for a given load, stiffer hamstrings will allow less anterior-directed motion of the tibia relative to the femur compared to more complaint hamstrings, thereby limiting the loads experienced by the ACL. To this end, healthy individuals with higher hamstring stiffness have been shown to display lesser ATT and PTASF during controlled non-weight bearing perturbations and double-leg jump-landing tasks, respectively. Additionally, females have been shown to display less stiffness than males. Given that females have also been shown to perform cutting and jump-landing maneuvers with characteristics

indicative of greater sagittal-plane ACL loading, and that females are at substantially increased risk for noncontact ACL injury, compared to males, it has been suggested that insufficient hamstring stiffness may help explain, at least in part, why females are at increased risk for injury.

While higher magnitudes of hamstring stiffness have been associated with characteristics indicative of lesser ACL loading, the injury models from which these relationships have been established are limited to non-weight bearing perturbations and double-leg jump-landing tasks. This is problematic because retrospective video analyses of actual noncontact ACL injuries have shown that such injuries are more likely to occur during single- versus double-leg landings. Thus, the injury models used previously may not adequately represent the situations in which noncontact ACL injuries typically occur. In addition, these relationships have been established with males and females included in the same statistical analyses and without equal sexstratification. This is also problematic because hamstring stiffness, PTASF, and several other biomechanical variables (e.g. initial knee flexion angles, knee flexion excursion, internal kneeextension moment, etc.), have been shown to be correlated with sex. Thus, grouping males and females in the same analyses makes it difficult to tease out the unique contribution of stiffness to ACL-loading characteristics versus other sex-dependent factors. Further, using a double-leg stopvertical jump task as a model for injury, it has previously been demonstrated that approximately 86% of the variance in PTASF could be predicted by a select combination of neuromuscular and biomechanical characteristics, where sex (being female), greater preparatory neuromuscular activation of the lateral quadriceps, greater knee-extension moments and posterior ground reaction forces, and greater knee flexion angles were predictive of greater PTASF. It remains unknown, however, whether these same factors are similarly predictive of PTASF when performing the same task on a single leg, and whether hamstring stiffness adds any predictive ability to the final model when included in the pool of possible predictors. Therefore, the

purposes of this dissertation were to: 1) compare the neuromuscular and biomechanical demands of a double- and single-leg stop-vertical jump (DLSJ and SLSJ, respectively) in males and females; 2) determine, within each sex, the extent to which hamstring stiffness uniquely predicts biomechanical characteristics of ACL loading during the SLSJ; and 3) examine the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics are able to collectively predict PTASF during the SLSJ.

When comparing the neuromuscular and biomechanical demands of the DLSJ to the SLSJ in males and females, it was hypothesized that the SLSJ would elicit a landing style considered to be more "risky" in terms of ACL loading and noncontact injury compared to the DLSJ, as evidenced by greater preparatory neuromuscular activation, a more upright body position at initial ground contact, smaller sagittal plane joint excursions and slower angular velocities, greater ground reaction forces and resultant joint moments, and characteristics indicative of greater sagittal plane ACL loading (i.e. greater PTASF, ATT, and ATA). It was also hypothesized that these aforementioned characteristics would be more pronounced in females compared to males. In general support of these hypotheses, our findings revealed that both males and females performed the SLSJ with a more posteriorly-oriented trunk center-of-mass position and smaller knee flexion angles at initial ground contact, less knee-flexion excursion, and greater PTASF, posterior and vertical ground reaction forces, and knee-extension moments, compared to the DLSJ. Thus, the SLSJ elicited characteristics associated with increased ligamentous loading, and a landing posture that was more representative of what has been observed during injurious situations. Additionally, although females performed both the DLSJ and SLSJ with a more "risky" landing style compared to males, they performed the SLSJ using a different biomechanical "strategy" at the hip, which suggests that the demands of performing the stopjump task on a single leg were likely greater for females. Collectively, these finding clearly

demonstrate that performing a stop-vertical jump task on a single leg (i.e. SLSJ) elicits different biomechanical outcomes than performing the same task on both legs (i.e. DLSJ), and that the demands of jumping and landing on a single leg are different for males and females. As such, these findings helped make an informed decision to use the SLSJ task as a model for injury, and sex-specific regression models when examining the extent to which hamstring stiffness was a unique predictor of ACL-loading characteristics.

When examining the unique contribute of hamstring stiffness to ACL-loading characteristics, it was hypothesized that, after statistically controlling for body positioning at initial ground contact (i.e. initial trunk center-of-mass position and hip and knee flexion angles), higher stiffness would be predictive of lesser ACL loading (i.e. less PTASF, ATT, and ATA) within each sex. Contrary to this hypothesis, however, stiffness was not found to be a significant predictor of PTASF, ATT, or ATA during the SLSJ in either sex. Given that higher stiffness has previously been associated with biomechanical characteristics indicative of lesser ACL loading, our conflicting findings suggest that hamstring stiffness may not be as effective at controlling sagittal-plane knee-joint loading when landing on a single leg, potentially due to a more upright landing style. Specifically, landing with the trunks center-of-mass positioned posteriorly, and the hip and knee relatively extended – as observed during the SLSJ, may have altered the lengthtension relationship of the hamstring muscles and their line-of-action on the proximal tibia, thereby limiting their ability to generate an adequate posteriorly-directed shear force at the knee and protect the ACL from sagittal-plane loading. This was also the case when examining the extent to which a select group of anatomical, neuromuscular, and biomechanical characteristics could collectively predict PTASF during the SLSJ. Specifically, it was found that the combination of preparatory neuromuscular activation of the lateral quadriceps, knee-flexion angle, and knee-extension moment, collectively predicted approximately 78% of the variance in

PTASF. Although hamstring stiffness did not enter into the final prediction model, the variables that were found to be significant predictors of PTASF during the SLSJ in large part agreed with those that have been shown to predict PTASF during other tasks in prior work. Thus, lateral quadriceps activation, knee-flexion angle, and knee-extension moment, appear to be important factors to consider when designing future intervention strategies or attempting to examine the effectiveness of current injury prevention programs.

The collective findings of this dissertation are expected to impact current noncontact ACL injury-prevention strategies as well as future laboratory-based studies aimed at identifying potential risk factors for injury. For example, the more upright landing style, and biomechanical characteristics associated with increased ligamentous loading, elicited by the SLSJ suggests that current injury prevention efforts should place a greater emphasis on single-leg jumping and landing activities and focus on teaching individuals to perform such activities with safer landing strategies (e.g. greater amounts of trunk-, hip-, and knee-flexion at initial ground contact, and greater amounts of joint excursion throughout landing). Our finding that preparatory neuromuscular activation of the lateral quadriceps, knee-flexion angle, and knee-extension moment, were significant predictors of PTASF further supports this recommendation. In addition, because the landing style displayed by participants during the SLSJ was more in line with what has been observed during injurious situations, this suggests that single-leg jump-landing tasks, that include both horizontal and vertical deceleration components, may be more ecologically valid injury models compared to double-leg tasks. As such, the findings of this dissertation may help future laboratory-based studies select a task that best helps answer the research question at hand. That said, the between-sex differences identified in this dissertation provide sufficient evidence to highlight the fact that it would be ill-advised to simply lump males and females into the same analyses in future correlational-type studies. Instead, researchers should be encouraged

to employ sex-specific models in future work to eliminate any potential for spurious findings due to between-sex differences. Furthermore, although hamstring stiffness was not found to uniquely contribute to sagittal-plane ACL-loading characteristics during the SLSJ, irrespective of sex, it remains unclear whether this lack of a relationship was due the a more upright landing style placing the hamstrings in a position in which they are unable to effectively resist anterior-directed forces and motion at the proximal tibia, and thus ACL loading. Therefore, additional studies are needed to better understand the functional role of the hamstrings in effectively resisting ACL loading when landing in a more extended position.

In addition to their expected impact on current injury prevention efforts and risk-factor identification studies, the findings of this collective work have also revealed several directions for future research. First, many of the biomechanical and neuromuscular characteristics currently thought to contribute to females' increased risk for noncontact ACL injury are based on observed differences between males and females. However, the evidence that different tasks can affect biomechanical outcomes, and that the task demands are different for males and females, suggests that between-sex differences in landing mechanics are task dependent. As such, a systematic review and meta-analysis examining between-sex differences in landing mechanics, as a function of task, is planned to better identify what is truly known about biomechanical differences between males and females. Identifying the true evidence for biomechanical and neuromuscular differences between males and females across a variety of tasks would provide researchers and clinicians with a clearer understanding of the factors that most likely contribute to females' increased risk for injury. Second, although the hamstrings have been shown to effectively resist anterior and rotary tibiofemoral motion when the knee is flexed beyond ~15°, the more upright landing posture, and characteristics indicative of increased ACL loading, elicited by the SLSJ raises the question of whether the hamstrings are positioned in a way in which they can

effectively protect the ACL from deleterious loading when landing on a single leg. To this end, although there are inherent difficulties associated with measuring muscle forces and ACL loading during dynamic tasks in-vivo, the biomechanical and neuromuscular data recorded during the SLSJ could be used to drive musculo-skeletal modeling simulations to better determine the hamstrings influence on ACL loading when landing in a position that is more representative of the situations in which such injuries commonly occur. Third, while it is theorized that insufficient hamstring stiffness may increase injury risk due to increased ligamentous loading, this is largely based on reports of females displaying less stiffness than males. However, a unique finding of this dissertation was that females actually displayed either greater, or equal, stiffness values compared to males depending on the method used to standardize the assessment load. Specifically, females displayed greater normalized stiffness values than males when assessed using a 10% body mass load, but similar values when assessed using a 30% MVIC load, which suggests that between-sex differences in stiffness may be assessment-method dependent. Given that stiffness is shown to increase as neuromuscular effort increases, this suggests that the MVIC load-assignment method should be used in future studies aimed at identifying the unique contribution of hamstring stiffness on ACL loading characteristics. Finally, although hamstring stiffness was not a predictor of ACL loading characteristics during the SLSJ, this relationship was examined using a sample of physically active males and females. Given that athletic populations are more vulnerable to such injuries, and that athletes likely display different landing mechanics compared to physically active individuals, future studies examining more homogenous athletic populations are planned.

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APPENDIX A

APPROVED INSTITUTIONAL REVIEW BOARD ADULT CONSENT FORM

UNIVERSITY OF NORTH CAROLINA AT GREENSBORO CONSENT TO ACT AS A HUMAN PARTICIPANT

Project Title: The Influence of Lower-Extremity Stiffness Measures on Knee Joint Biomechanics during Double-Leg and Single-Leg Jump Landing Tasks

Principal Investigator and Faculty Advisor: Justin P. Waxman, MSc (Principal Investigator) and Sandra J. Shultz, PhD. ATC, CSCS (Faculty Advisor) Participant's Name: What are some general things you should know about research studies?

You are being asked to take part in a research study. Your participation in the study is voluntary. You may choose not to join, or you may withdraw your consent to be in the study, for any reason, without

Research studies are designed to obtain new knowledge. This new information may help people in the future. There may not be any direct benefit to you for being in the research study. There also may be risks to being in research studies. If you choose not to be in the study or leave the study before it is done, it will not affect your relationship with the researcher or the University of North Carolina at Greensboro. Details about this study are discussed in this consent form. It is important that you understand this information so that you can make an informed choice about being in this research study.

You will be given a copy of this consent form. If you have any questions about this study at any time. you should ask the researchers named in this consent form. Their contact information is below.

What is the study about?

This is a research project. Your participation is voluntary. The purpose of this study is to examine the extent to which measures of lower-extremity (leg) muscle tightness are predictive of motions and forces at the knee joint during double-leg and single-leg jump landing tasks.

You are being asked to participate in this study because you are a healthy individual between the ages of 18 and 26 years, you are physically active for a minimum of 300 minutes per week, and you regularly participate in activities that involve running, cutting, jumping and landing. You should not participate in this study if you have ever had a serious knee injury, have ever had lower-extremity (leg) surgery, have had a lower-extremity injury within the past 6 months, or if you have any known medical conditions that would affect your joints, hearing, or balance. Additionally, you should not participate in this study if you are currently pregnant, are attempting to become pregnant, or if you are allergic to adhesives.

What will you ask me to do if I agree to be in the study?

If you agree to participate in this study, you will be asked to visit the Applied Neuromechanics Research Laboratory (238 Coleman Building) for a single testing session that will last approximately 2.0 hours, and visit the Human Nutrition Assessment Laboratory (341 Stone Building) for a body composition assessment that will last approximately 15 minutes. Thus, your total time commitment will be approximately 2 hours and 15 minutes. Prior to any testing, you will be asked to complete intake surveys and questionnaires pertaining to your physical activity history, health and injury history, and selfperceived knee function, in order to ensure that you meet the inclusion criteria for this study. If you are a female, you will also be asked to complete a questionnaire pertaining to your menstrual cycle history.

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> > 4/27/16 to 12/8/16

During the testing session in the Applied Neuromechanics Research Laboratory, the following measurements will be collected: 1) body size measures of height, weight, leg length, and leg girth, 2) looseness of your joints, 3) single-leg hop for distance, 4) lower-extremity strength, 5) hamstring and leg muscle tightness, and 6) knee joint motions and forces during jumping and landing tasks. The joint looseness measures will consist of pushing and pulling on your lower leg to measure the looseness of your knee joint and having you manually extend your pinky fingers, elbows, and knees, manually flex your wrists, and bend at the waist. You will then have sensors placed on your skin over the front and back of your thigh muscles in order to monitor how hard your muscles are working. You will then complete a 5 minute warm-up on a stationary bike followed by performing a single-leg hop for distance which involves standing on one leg, jumping forward as far as you can, and then landing on that same leg. Lower-extremity (leg) strength will be measured by having you maximally contract your leg muscles against an unmovable resistance. Hamstring muscle tightness will be assessed by having you lay face down on a padded table with a weight secured around your ankle. You will contract your hamstring muscles in order to hold your lower leg in the testing position while the investigator applies a gentle tap to the back of your heel. Leg muscle tightness will be assessed by having you hop barefoot on one leg, and on two legs. We will then assess the motions and forces at your knee joint during double and single-leg jumping and landing tasks. For the double-leg task, you will be asked to jump forward, immediately perform a jump for maximal height, and then land. The single-leg task will be carried out in identical fashion to the double-leg task, but only on the left leg.

During the body composition assessment in the Human Nutrition Assessment Laboratory, your body composition will be assessed using small doses of X-rays. This assessment only requires that you wear loose clothing (sweatpants or gym shorts and a loose top) and lay still and flat on an x-ray table while the scanner passes over your body (without touching you) from head to toe. Because this is not considered to be safe during pregnancy, females will be asked to undergo a pregnancy test prior to completing this assessment.

Is there any audio/video recording?

The researchers may take still photographs or video recordings during the study so that they can use these materials for future conference presentations and publications only if you give your permission to do so. Because your voice/ image will be potentially identifiable by anyone who hears the tape, your confidentiality for things you do and say on the tape cannot be guaranteed although the researcher will try to limit access to the tape as described below (please refer to the section below titled: How will you keep my information confidential?). Please indicate your decision below by initialing in the space provided.

I give the researchers my permission to take still photographs and/or video
recordings of me during my testing session.
I do not give the researchers my permission to take still photographs and/or video
recordings of me during my testing session.

What are the risks to me?

The Institutional Review Board at the University of North Carolina at Greensboro has determined that participation in this study poses minimal risk to participants. There is a rare (< 1% incidence) risk of discomfort or injury due to joint and leg muscle strain during the joint looseness, hamstring and leg muscle tightness, lower-extremity strength, single-leg hop for distance, and jumping and landing assessments. In order to minimize these risks, you will be screened for pre-existing injury prior to participation in this study. There is also a minimal risk of mild skin irritation that may result from the adhesive tape used to secure the muscle sensors to the skin during testing. To minimize the risk of skin irritation due to the use of adhesive tape, allergy to adhesives has been included as exclusion criteria for participation in this study. Lastly, the body composition assessment involves exposure to mild radiation.

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4/27/16 to 12/8/16

This radiation poses minimal risk and is equivalent to 1/10th of the radiation that is emitted during a routine X-ray; however, it is considered unsafe to be exposed to radiation during pregnancy. As such, a trained technician will perform all body composition assessments, and a pregnancy test will be administered to all female participants, in order to ensure participant safety. This study will be stopped in the unlikely event that any of the measurements become painful or emotionally distressing.

If you have questions, want more information or have suggestions, please contact Justin P. Waxman at ipwaxman@uncg.edu OR 336.334.3031 or Dr. Sandra J. Shultz at sishultz@uncg.edu OR 336.256.1429. If you have any concerns about your rights, how you are being treated, concerns or complaints about this project or benefits or risks associated with being in this study please contact the Office of Research Compliance at UNCG toll-free at (855)-251-2351

Are there any benefits to society as a result of me taking part in this research?

Society may benefit from a greater understanding of how lower-extremity stiffness measures are related to biomechanical factors that are known to influence anterior cruciate ligament loading and thus, noncontact anterior cruciate ligament injury risk.

Are there any benefits to me for taking part in this research study? There are no direct benefits to you for participating in this study.

Will I get paid for being in the study? Will it cost me anything? If you agree to participate in this study, you will receive a payment in the amount of \$10 as long as you complete both the testing session and the body composition assessment. Alternatively, you may forfeit this \$10 payment in return for extra credit in one of your courses if applicable. There are no costs to you for participating in this study. ______ I choose to receive \$10 compensation for my participation in this study. ______ I choose to forfeit the \$10 compensation for my participation in this study and elect to receive extra credit for one of my courses offering this opportunity instead. Course name: ______

How will you keep my information confidential?

All information obtained in this study is strictly confidential unless disclosure is required by law. In order to preserve confidentiality, written information will be kept in a locked filing cabinet in secured office and electronic information will be stored on a password protected computer. Data will be coded to ensure that no personally identifiable information is attached to any data. A file linking the subject name and code number will be kept secure in a password protected computer, and will be kept separate from collected data. Additionally, a hard copy of the master file linking subject names and code numbers will be stored in a locked file cabinet separate from collected data. Participants will not be identified by name when data are disseminated. De-identified data will be kept indefinitely. All audio/video/still photography will be stored on a password protected computer. No audio data will be disseminated. The consent form and other identifiable information will be destroyed after 3 years by being physically shredded and/or electronically deleted via the use of Eraser software.

What if I want to leave the study?

You have the right to refuse to participate or to withdraw at any time, without penalty. If you do withdraw, it will not affect you in any way. If you choose to withdraw, you may request that any of your data which has been collected be destroyed unless it is in a de-identifiable state. The investigators also have the right to stop your participation at any time. This could be because you have had an unexpected reaction, or have failed to follow instructions, or because the entire study has been stopped.

Page 3 of 4 UNCG IRB
Approved Consent Form
Valid from:
4/27/16 to 12/8/16

What about new information/changes in the study? If significant new information relating to the study becomes available which may relate to your willingness to continue to participate, this information will be provided to you.
Voluntary Consent by Participant: By signing this consent form, you are agreeing that you read, or it has been read to you, and you fully understand the contents of this document and are openly willing consent to take part in this study. All of your questions concerning this study have been answered. By signing this form, you are agreeing that you are 18 years of age or older and are agreeing to participate, or have the individual specified above as a participant participate, in this study described to you by Would you be willing to be contacted for recruitment in future studies? Yes No

Signature: _____ Date: _____

UNCG IRB Approved Consent Form Valid from:

4/27/16 to 12/8/16

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APPENDIX B

PARTICIPANT INTAKE FORMS

PHYSICAL ACTIVITY AND HEALTH HISTORY

Do you have any General Health Problems or Illnesses? (e.g. diabetes, respiratory disease) Yes No							
Do you have any vestibular (inner ear) or balance disorders? Yes No							
Do you smoke? Yes No							
Do you drink alcohol? Yes No If yes, how often?							
Do you have any history of connective tissue disease or disorders? (e.g. Ehlers-Danlos, Marfan's Syndrome, Rheumatoid Arthritis) Yes No							
Has a family member of yours ever been diagnosed with breast cancer? Yes No (if no, please skip next question.)							
If yes, please put a check next to the types of relatives that have been diagnosed. You may check more than one box:							
Mother Sister Grandmother Aunt							
Male relative (father, brother, grandfather, or uncle)							
Other type of relative (please write in)							
Please list any medications you take regularly:							
Please list any previous injuries to your lower extremities. Please include a description of the injury (e.g. ligament sprain, muscle strain), severity of the injury, date of the injury, and whether it was on the left or right side.							
Body Part Description Severity Date of Injury L or R							
Hip							
Thigh							

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Body Part	Description	Severity	Date of Inj	игу	L or R
Knee					
Lower Leg					
Ankle					
Foot					
	nny previous surge date of the surger				
Body Part	Description	on [ate of Surgery		L or R
please indic activity (i.e.	all physical activitie ate how much time competitive or rec g in the activity.	you spend each v	veek in this activ	ity, the i	ntensity of the
Activity	#Days/week	#Minutes/Day	Intensity.	Activit	y Began When?

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t time of day do you generally engage in the above activities?
se list other conditions / concerns that you feel we should be aware

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The Activity Rating Scale

Please indicate how often you performed each activity in your healthiest and most active state, in the past year.

	Less than one time in a month	One time in a month	One time in a week	2 or 3 times in a week	4 or more times in a week
Running: running while playing a sport or jogging					
Cutting: Changing directions while running					
Decelerating: coming to a quick stop while running					
Pivoting: turning your body with your foot planted while playing a sport; For example: skiing, skating, kicking, throwing, hitting a ball (golf, tennis, squash), etc.					

Investigator Comments:

Female Hormonal History

This questionnaire asks questions about your menstrual cycle. As a reminder, this information is strictly confidential. None of this information will be shared with anyone besides the study investigators. For research purposes, your survey uses a coded identification number in substitution for your name. If you have any questions, or do not understand any of the questions, please let us know.

Subject Code:		Date:		A	.ge:		
1. How old were you	How old were you when you started your menstrual periods (Age)?						
2. When was the first	2. When was the first day of your last period (month/day)?						
	3. On average, how many days are there between your menstrual periods (from Day 1 of your period to Day 1 of the next period)?						
4. How many menstro	aal periods have y	you had in the	last 12 mo	nths?			
5. Have you missed a	ny menstrual per	iods within th	e last 12 m	onths? (Please c	ircle) YE	s No	
6. Since starting your have a menstrual p					where you	did not	
	was the most rec at last?				how many	· ——	
7. Do you know when	ı your next mens	trual periods	will start? (Please Circle)	YES	NO	
When will thi	s be (month/day)	?					
8. Do you exhibit pre	menstrual sympto	oms? (Please	Circle)	YES	NO		
If you experie symptoms on	nce premenstrual a scale of			ate the severity o 10= severe symp		f your	
	ng: llity: Cravings: list any other syn	_ Mood : Cramp	Swings: s:				
9a. Are you currently	using birth contr	ol medication	ı/estrogen t	herapy for any r	eason? YE	S NO	
9b. Type of medication	on: (Circle One)	Pill Patch	Injection	Vaginal Ring	Other:		
9c. Please list the bran	nd name and dos	age:					

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9d. When did you start using this type?	(month/year)					
10. Have you <i>previously</i> used birth cont	trol medication/estrogen therapy fo	or any reason? YES/NO				
If YES, please list the brand name and dosage:						
Please list dates of use:	(date/year) to	(date/year)				

INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE

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

Think about all the vigorous activities that you did in the last 7 days. Vigorous physical activities refer to activities that take hard physical effort and make you breathe much harder than normal. Think *only* about those physical activities that you did for at least 10 minutes at a time.

1.	During the last 7 days, on how many days did you do vigorous physical activities like heavy lifting, digging, aerobics, or fast bicycling?						
	days per week						
	No vigorous physical activities → Skip to question 3						
2.	How much time did you usually spend doing vigorous physical activities on one of those days?						
	hours per day						
	minutes per day						
	Don't know/Not sure						
activiti somev	about all the moderate activities that you did in the last 7 days. Moderate ies refer to activities that take moderate physical effort and make you breathe what harder than normal. Think only about those physical activities that you did least 10 minutes at a time.						
3.	During the last 7 days, on how many days did you do moderate physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis? Do not include walking.						
	days per week						
	No moderate physical activities Skip to question 5						

SHORT LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised August 2002.

4.	How much time did you usually spend doing moderate physical activities on one of those days?
	hours per day
	minutes per day
	Don't know/Not sure
home,	about the time you spent walking in the last 7 days. This includes at work and at walking to travel from place to place, and any other walking that you have done for recreation, sport, exercise, or leisure.
5.	During the last 7 days, on how many days did you walk for at least 10 minutes at a time?
	days per week
	No walking → Skip to question 7
6.	How much time did you usually spend walking on one of those days?
	hours per day
	minutes per day
	Don't know/Not sure
days. time.	ist question is about the time you spent sitting on weekdays during the last 7 Include time spent at work, at home, while doing course work and during leisure This may include time spent sitting at a desk, visiting friends, reading, or sitting or down to watch television.
7.	During the last 7 days, how much time did you spend sitting on a week day?
	hours per day
	minutes per day
	Don't know/Not sure
This	is the end of the questionnaire, thank you for participating.

SHORT LAST 7 DAYS SELF-ADMINISTERED version of the IPAQ. Revised August 2002.

Knee injury and Ostcoarthritis Outcome Score (KOOS), English version LK1.0

	KOOS KNEE SURVEY						
Today's date:/ Date of birth:/							
Name:							
INSTRUCTIONS: This survey asks for your view about your knee. This information will help us keep track of how you feel about your knee and how well you are able to perform your usual activities. Answer every question by ticking the appropriate box, only one box for each question. If you are unsure about how to answer a question, please give the best answer you can.							
Symptoms These question the last week.	ns should be ans	swered thinking	g of your knee	symptoms during			
S1. Do you have Never	swelling in your k Rarely	nee? Sometimes	Often	Always			
S2. Do you feel g moves? Never	grinding, hear click Rarely	sing or any other	type of noise wh	Always			
S3. Does your kr	nee catch or hang u Rarely	p when moving? Sometimes	Often	Always			
S4. Can you stra Always	ighten your knee fo Often	ully? Sometimes	Rarely	Never			
S5. Can you bendalways	d your knee fully? Often	Sometimes	Rarely	Never			
Stiffness The following questions concern the amount of joint stiffness you have experienced during the last week in your knee. Stiffness is a sensation of restriction or slowness in the ease with which you move your knee joint.							
S6. How severe i	is your knee joint s Mild	tiffness after firs Moderate	t wakening in the Severe	e morning? Extreme			
S7. How severe None	is your knee stiffne Mild	ess after sitting, l Moderate	ying or resting la Severe	ter in the day? Extreme			

Pain P1. How often do you Never	experience kne Monthly	e pain? Weekly	Daily	Always					
What amount of knee pain have you experienced the last week during the following activities?									
P2. Twisting/pivoting None	on your knee Mild	Moderate	Severe	Extreme					
P3. Straightening knee None	e fully Mild	Moderate	Severe	Extreme					
P4. Bending knee full None	y Mild	Moderate	Severe	Extreme					
P5. Walking on flat su None	nrface Mild	Moderate	Severe	Extreme					
P6. Going up or down	stairs Mild	Moderate	Severe	Extreme					
P7. At night while in t	bed Mild	Moderate	Severe	Extreme					
P8. Sitting or lying None	Mild	Moderate	Severe	Extreme					
P9. Standing upright None	Mild	Moderate	Severe	Extreme					
Function, daily living The following questions concern your physical function. By this we mean your ability to move around and to look after yourself. For each of the following activities please indicate the degree of difficulty you have experienced in the last week due to your knee.									
A1. Descending stairs None	Mild	Moderate	Severe	Extreme					
A2. Ascending stairs None	Mild	Moderate	Severe	Extreme					

Knee injury and Ostcoarthritis Outcome Score (KOOS), English version LK1.0

For each of the following activities please indicate the degree of difficulty you have experienced in the last week due to your knee.

A3. Rising from None	Mild	Moderate	Severe	Extreme
A4. Standing None	Mild	Moderate	Severe	Extreme
A5. Bending to f	floor/pick up an e Mild □	object Moderate	Severe	Extreme
A6. Walking on None	flat surface Mild	Moderate	Severe	Extreme
A7. Getting in/or None	ut of car Mild	Moderate	Severe	Extreme
A8. Going shopp None	ping Mild	Moderate	Severe	Extreme
A9. Putting on so None	ocks/stockings Mild	Moderate	Severe	Extreme
A10. Rising from None	n bed Mild	Moderate	Severe	Extreme
A11. Taking off None	socks/stockings Mild	Moderate	Severe	Extreme
A12. Lying in be None	ed (turning over, Mild	maintaining knee p Moderate	position) Severe	Extreme
A13. Getting in/o	out of bath Mild	Moderate	Severe	Extreme
A14. Sitting None	Mild	Moderate	Severe	Extreme
A15. Getting on/ None	off toilet Mild	Moderate	Severe	Extreme

For each of the following activities please indicate the degree of difficulty you have experienced in the last week due to your knee.

A16. Heavy domestic duties (moving heavy boxes, scrubbing floors, etc)							
None	Mild	Moderate	Severe	Extreme			
A17. Light domes None	tic duties (cool Mild	king, dusting, etc) Moderate	Severe	Extreme			
The following qu higher level. Th	estions conc e questions	itional activities em your physical should be answe d during the last v	red thinking o	of what degree of			
SP1. Squatting None	Mild	Moderate	Severe	Extreme			
SP2. Running None	Mild	Moderate	Severe	Extreme			
SP3. Jumping None	Mild	Moderate	Severe	Extreme			
SP4. Twisting/piv	oting on your i Mild	injured knee Moderate	Severe	Extreme			
SP5. Kneeling None	Mild	Moderate	Severe	Extreme			
Quality of Life							
Q1. How often are Never	you aware of Monthly	your knee problem? Weekly	Paily	Constantly			
		style to avoid poter	ntially damaging	g activities			
to your knee? Not at all	Mildly	Moderately	Severely	Totally			
Q3. How much are Not at all	e you troubled Mildly	with lack of confide Moderately	ence in your kno Severely	ee? Extremely			
Q4. In general, ho None	w much diffict Mild	ulty do you have wit Moderate	th your knee? Severe	Extreme			

Thank you very much for completing all the questions in this questionnaire.

APPENDIX C
DESCRIPTIVE STATISTICS OF SECONDARY DATA

Appendix C1. Unadjusted Means and Standard Deviations (M \pm SD) for All Dependent Variables of Interest.

	Males (n = 34)		Females (n = 32)				
	DLSJ	SLSJ	DLSJ	SLSJ			
Kinematics							
$TrunkCOM_{IC}\left(cm\right)$	-10.4 ± 4.2	-12.7 ± 4.1	-10.1 ± 5.2	-12.6 ± 4.9			
$TrunkCOM_{EXC}(cm) \\$	12.1 ± 5.1	11.4 ± 4.4	12.2 ± 5.7	11.4 ± 6.0			
$\mathrm{HF}_{\mathrm{IC}}(^{\circ})$	34.56 ± 31.23	17.11 ± 20.35	34.95 ± 31.51	20.30 ± 37.44			
HF_{EXC} (°)	60.20 ± 23.64	52.56 ± 19.92	48.62 ± 22.42	35.36 ± 25.34			
HFV ($^{\circ}$ ·s ⁻¹)	191.63 ± 116.85	190.95 ± 76.81	165.69 ± 133.55	105.89 ± 135.24			
KF_{IC} (°)	12.59 ± 7.26	3.12 ± 7.05	9.51 ± 7.59	-0.96 ± 8.25			
KF_{EXC} (°)	68.54 ± 12.84	52.75 ± 10.20	64.01 ± 10.36	51.50 ± 9.51			
KFV (°·s⁻¹)	261.07 ± 36.12	201.26 ± 34.74	269.94 ± 40.98	216.85 ± 28.37			
Kinetics							
$vGRF_{Pk}\left(BW\right)$	1.66 ± 0.43	2.97 ± 0.52	1.57 ± 0.36	2.71 ± 0.40			
$pGRF_{Pk}\left(BW\right)$	-0.46 ± 0.13	-0.76 ± 0.16	-0.43 ± 0.13	-0.72 ± 0.14			
$HFM_{Pk}(BW^{\text{-}1}{\cdot}Ht^{\text{-}1})$	-0.15 ± 0.03	-0.20 ± 0.05	-0.12 ± 0.04	-0.14 ± 0.05			
$KEM_{Pk}\left(BW^{\text{-}1}{\cdot}Ht^{\text{-}1}\right)$	-0.08 ± 0.02	-0.08 ± 0.04	-0.08 ± 0.02	-0.08 ± 0.03			
Neuromuscular							
$QUAD_{PRE}\left(\%MVIC\right)$	27.07 ± 22.39	33.11 ± 28.70	20.09 ± 12.67	23.13 ± 20.58			
HAM_{PRE} (%MVIC)	5.96 ± 3.58	11.51 ± 7.13	7.03 ± 4.19	14.53 ± 12.64			
ACL Loading Characteristics							
$ATT_{Pk}\left(mm\right)$	26.26 ± 27.53	17.56 ± 47.88	56.07 ± 42.14	29.98 ± 31.82			
$ATA_{Pk}(m\!\cdot\! s^{\text{-}2})$	17.36 ± 6.50	16.44 ± 5.92	21.90 ± 9.82	17.86 ± 6.49			
$PTASF_{Pk}\left(BW\right)$	0.53 ± 0.13	0.66 ± 0.19	0.55 ± 0.15	0.71 ± 0.19			