The Effects of Functional Fatigue on the Hoffmann's Reflex in Female Soccer Players to Predict Anterior Cruciate Ligament Injury

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Authorization to Submit Thesis

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Abstract

Anterior Cruciate Ligament injuries are debilitating and have a time-consuming rehabilitation process. Females are 2 to 8 times more likely to sustain non-contact ACL injuries than men, particularly in sports that require frequent cutting, accelerating, jumping, and landing. Although there are many suggestions as to why this high incidence of injury occurs in women, there are no real explanations. The present study used a within subjects design to investigate the effect of functional fatigue on the Hoffmann's Reflex in female Division I soccer players. The H-Reflex was used to test the action of the stretch reflex within the quadriceps to determine how human reflexes are affected due to fatigue. The onset, duration, and peak of H-reflex were measured for each subject before and after fatigue. A significant change in stimulus required to produce the onset of H-reflex was found when comparing pre and post fatigue results.

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Dedication

I dedicate my thesis to my many friends who have supported me throughout my thesis, new and old. I will always appreciate all they have done, especially the athletics department at the University of Idaho, who gave me this great opportunity to complete my masters.

A special thanks goes out the Idaho Vandals Soccer Program. To all the players who made this research possible, to the assistant and head coaches that have come and gone but especially to Derek Pittman, who made me believe in my passions again. That through hard work and dedication, in all realms of life, you make life what you want it to be.

Most of all, I dedicate my thesis to my family. A special feeling of gratitude goes out to my loving parents whose words of encouragement and push for excellence will always go with me. I hope that the completion of this thesis not only makes them proud, but also goes on to motivate my brother, James, in his own path for academic success. To all my family members, both to those who are still here and those who are no longer with us, this is for you

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Chapter One – Introduction

Introduction

Anterior Cruciate Ligament (ACL) injuries are debilitating and often season-ending injuries. Recovery from an ACL injury typically is an expensive process commonly involving surgery and a time-consuming rehabilitation process. Females are 2 to 8 times more likely to sustain non-contact ACL injuries than males, particularly in sports that require frequent cutting, accelerating, decelerating, jumping, and landing (Mandelbaum & Silvers, 2005). Although there have been many suggestions as to why this high incidence of injury occurs, there is not a consensus on the underlying causes (Effectiveness of a Neuromuscular and Proprioceptive Training Program in Preventing Anterior Cruciate Ligament Injuries in Female Athletes).

The ACL is one of the 5 structural ligaments in the knee, the others being the posterior cruciate ligament, medial collateral ligament, anterolateral ligament and lateral collateral ligament (Claes, Vereecke, Maes, Victor, Verdonk, & Bellemans, 2013). The ACL functions as the primary static restraint to anterior tibial translation (Butler, Noyes, & Grood, 1980). Excessive tibial translation can lead to anything from a mild sprain to a complete rupture. In addition to the ligamentous support, muscles also act to protect the tibia and femur from separating during activity. Some studies indicate that the quadriceps are a primary muscular restraint to anterior tibial translation during closed kinetic chain activities such as running, jumping, walking and standing (Bodor, 2001). Other studies focus on the importance of the hamstring muscles in the posterior thigh, and it's role in stabilizing the knee along with the quadriceps. The strength relationship between the quadriceps and hamstrings are of importance, whereby female athletes after menarche who have quadriceps strength greater to their hamstring strength, have exhibited a higher risk of ACL injury (Ahmad, Clark, Heilmann, Schoeb, Gardner, & Levine, 2006).

Possible risk factors associated with noncontact ACL injuries include fatigue, the environment, hormones, anatomy and the neuromuscular system, as well as various other factors that may contribute. In particular, females are far more likely to sustain a noncontact ACL injury than males during activities that require frequent acceleration, deceleration, jumping, landing and changes of direction. Irrespective of the athlete's sex, an ACL rupture is more likely to occur from a non-contact mechanism of injury (MOI) (Remer, Fitzgerald, Friedman, Rogers, Hendrix, & Schafer, 1992). An example of such an injury is when the foot is planted and the athlete changes direction quickly, causing a quick rotation of the knee where the tibia rotates towards the midline of the body, causing an over stretch of the ligament (Griffin, 2001).

While ACL tears are common in male and female athletes, multiple intrinsic and extrinsic factors play a role in the injury risk patterns. Intrinsic factors include factors such as height, weight, limb alignment, strength, generalized laxity, and hormonal differences, whereas extrinsic factors include variations in equipment, as well as practice and game facilities (Deitch, Starkey, Walters, & Moseley, 2006). The overall focus of research on ACL tears have been on intrinsic factors, which were separated into three categories for the purpose of this study: anatomical differences, neuromuscular differences, and hormonal differences. Anatomical differences that have been studied include: lower extremity alignment (Q-angle), intercondylar notch size and shape, ligament size and ACL strength, ligament laxity, and range of motion of the knee. Neuromuscular differences are muscle strength, neuronal firing and flexibility; hormonal differences include changes in estrogen, progesterone, testosterone, and relaxin throughout the menstrual cycle. (Pantano, White, Gilchrist Louise, & Leddy, 2005).

Some of the most prominent factors theorized to result in ACL injury are anatomical differences, such as the Q angle and intercondylar notch size. The Q angle represents the direction of pull of the quadriceps muscles through the patella to its insertion on the tibial tuberosity and is defined as the angle between the line connecting the anterior superior iliac spine (ASIS) to the center of the patella, and the extension of a line from the tibial tubercle to the same reference point on the patella (Brattstroem, 1964); the angle is strongly influenced by the position of the patella and the tibial tubercle. A large

Q-angle, which is greater than 20 degrees in females, may be associated with increased knee valgus movement resulting in anterior cruciate ligament strain (Horton & Hall, 1989). Investigators have found that generally males with Q angles above 15 degrees and females with Q angles above 20 degrees are clinically abnormal. Normal Q angles have been reported between 8 and 17 degrees in females, with females consistently having higher Q angles are than males when compared (Woodland & Rulon, 1992; Nguyen & Shultz, 2007). Forces occurring during sports, while changing direction for example, are concentrated on the ligament each time the knee twists, causing an anatomical difference in the joint compared to when the athlete is standing quietly. Although more recently Posthumus et al. (2011) stated that the certainty of a large quadriceps angle increasing the risk of ACL injury is inconclusive and needs further research, pointing towards multifactorial reasons for ACL injuries, some of which will be explained (Posthumus, Collins, September, & Schwellnus, 2011).

The second prominent anatomical difference between males and females is the size and shape of the intercondylar notch, where the ACL attaches to the femur. Shelbourne reported this notch as being significantly narrower in females using the smaller surface of attachment as an explanation as to why females are at greater risk of ACL injury, compared to male counterparts. Shelbourne et al. (1998) stated that the smaller area for attachment increases the chances of detachment of the ACL from the femur due to the smaller surface area of the ligament attached to the bone. In addition, van Eck et al. (2010) recorded significantly different shapes of the intercondylar notch between males and females, with females having more of an "A" shape compared to the "U" shape seen in males. The most important finding in this study was that ''A-shaped'' notches were narrower at the base, middle, and top of the notch entrance than ''U-shaped'' notches, implying predispositions for ACL injury (van Eck, et al., 2010). Lastly, Chandrashekar et al. (2005) reported the female ACL as having a smaller cross-sectional area, a shorter length, and smaller volume than the male ACL.

A controversial factor that is under much review is the hormone differences between males and females and how hormones can change aspects about the knee joint, and that can increase the chance of ACL injury. Female hormones such as estrogen, progesterone and relaxin, allow for greater joint laxity due to the hormone receptor sites on connective tissues such as the ACL. Depending on the amounts of each hormone released into the bloodstream, studies have indicated females may be more susceptible to joint injury at different times in the menstrual cycle (Faryniarz, Bhargave, Lajam, & al, 2006).

Fatigue is another factor that has been examined to determine why both males and females tear their ACL, and is a key factor that this study focuses on. Specific studies have focused on the role of fatigue in injuries as a whole, some focusing explicitly on the relationship between fatigue and ACL injuries. Harris et al. (2013) found that in professional basketball players, 40% of all ACL injuries occurred in the fourth quarter, 62% in the second half overall, implying a role of fatigue in the injury. Further, a study focusing on professional females soccer players indicated that there was a statistical significance with all injuries, including ACL tears, increasing towards the end of the season (Giza, Mithöfer, Farrell, Zarins, & Gill, 2005). These studies suggest that fatigue could have some affect on the risk of ACL injuries towards the end of games and seasons. A risk that can be connected to neuromuscular control, the area that this thesis will be focusing on.

Problem Statement

ACL injuries are an issue in female sports, especially those that require cutting, jumping, landing, and deceleration and acceleration. While numerous factors contribute to the potential for ACL injury, one of the more recent is fatigue. Given that the injury is most commonly occurs as a non-contact injury, one of the emerging hypotheses is that fatigue plays a part in this statistic (Harris, et al., 2013).

Females who have experienced ACL tears and therefore ACL reconstruction have had to deal with the mental, psychological and physical pain of such an injury. Action is needed if we are to keep females healthy and safe in one of the most popular female sports in the world.

Dependent Variables

1. Spinal Reflexive Excitability measured by the Hoffman Reflex
    a. Maximum H-Reflex amplitude measured in the Vastus Medialis muscle supplied by the femoral nerve.

Independent Variables

2. Participants
    a. NCAA Division 1 female soccer players.
3. Femoral nerve stimulation
    a. Reflex evoked through different voltage stimulations using the BioPac system.

Research Questions

Does the excitability of the spinal reflexive pathways of the Vastus Medialis muscle change once the target muscle of a female NCAA division I soccer player is fatigued.

Null Hypothesis

There will be no difference in the onset or magnitude of the H-Reflex between pre and post fatigue conditions.

Research Hypothesis

A higher stimulus will be required to elicit a post-fatigue H-reflex and the magnitude of the H-reflex will be decreased post fatigue.

Operational Definitions

- H-reflex – Hoffmann reflex. Testing method used to determine spinal reflexive contributions of a muscle
- VM – Vastus Medialis. One of the quadriceps muscles involved in alignment of the patella in the last 10-15 degrees of extension and a stabilizer to lateral patellar pull. (Fourie, 2010)

- Fatigue - decline in the maximal force or power capacity of muscle (Enoka & Duchateau, 2008)
- EMG - is an electrodiagnostic medicine technique for evaluating and recording the electrical activity produced by skeletal muscles (Kamen, 2004).
- NCAA - The National Collegiate Athletic Association (NCAA) is a non-profit association that regulates athletes of 1,281 institutions, conferences, organizations, and individuals (National Collegiate Athletics Association).
- Division I – Division I (D-I) is the highest level of intercollegiate athletics sanctioned by the National Collegiate Athletic Association (NCAA) in the United States. D-I schools include the major collegiate athletic powers, with larger budgets, more elaborate facilities, and more athletic scholarships than Divisions II and III as well as many smaller schools committed to the highest level of intercollegiate competition (About the NCAA, 2015).

Delimitations

1. The participants recruited for this study were healthy young females, all of whom were current NCAA Division 1 Soccer Players at the University of Idaho. The explanations and implication of the outcomes were based on this population.
2. This research examined the H-Reflex of the VM, supplied by the femoral nerve, in the participant’s dominant leg before and after strenuous exercise.

Limitations

1. This research was limited to investigation of the H-reflex modulation in the vastus medialis oblique muscle. The outcomes may not explain changes in reflexive function of other supporting muscles such as the hamstrings or gastrocnemius.
2. The H-reflex is sensitive to the position of the knee, position of the body, level of contraction of surrounding muscles and posture of the subject. Although every effort was taken to control every factor, it is not possible to maintain all factors at all times, across all subjects. To mitigate the effects of variation, 3 readings were taken at each stimulation voltage to produce an average.

3. EMG data collection may also be a limiting factor due to the likelihood in variance if not collected under the same conditions. Inability of the subject to relax the target muscle as well as surrounding muscles, along with other body parts is a potential factor which may modify results.

Study Design

This study used a within subjects design to investigate the effect of functional fatigue on the H-Reflex in NCAA female division I soccer players. Prior to muscular fatigue the H-Reflex of each participant was elicited through direct stimulation of the femoral nerve and the magnitude and timing of the reflex were measured via EMG from the VM. The participants were then fatigued via a Wingate cycling protocol, a standardized test that causes fatigue, and the H-Reflex was elicited again immediately following the Wingate test. The procedure was repeated twice to ensure that pre and post H-reflex testing could be compared to see the effects of fatigue.

Subjects

All participants were between the ages of 18 and 22 years old and were screened for current musculoskeletal injury of lower extremities or back, lingering symptoms of musculoskeletal injury in lower extremities or back, current musculoskeletal pain in the lower extremities or back, chest pain with exertion, shortness of breath, known hypertension, history of heart attack, or pregnancy. All subjects were also examined by a Certified Athletic Trainer employed at the University of Idaho due to NCAA division I rules for student athletes requiring a physical checkup by July. Therefore, any participants disqualified for soccer activity due to lower extremity/back pain or dysfunctions were disqualified for the study also. All participants were also current members of the University of Idaho Division I female Soccer team at the time of testing.

Instrumentation

The BIOPAC MP150 (BIOPAC Systems Inc., Goleta CA, USA) was used to convert analog EMG signals to a digital signal that was used for data processing in AcqKnowledge, an interactive program allowing us to instantly view, measure, transform, replay and analyze data from the BIOPAC MP150 (BioPac - AcqKnowledge Software). The STMISOC stimulator module was used in tandem with the BIOPAC system to provide pulse and waveform stimulus outputs for nerve conduction in the femoral nerve (BioPac - Stimulator Module).

Ethical Approval

The research presented in this thesis was conducted in compliance with the University of Idaho procedures that require Institutional Review Board (IRB) review and approval of projects involving humans. Official approval from the IRB was given before the research begun (Approval No. 14-310, July 14th, 2014).

Significance of Research

Studies have indicated that females are up to 8 times more likely to suffer from non-contact ACL injuries compared to males (Mandelbaum & Silvers, 2005). ACL injuries require consumables, drugs, equipment, surgery, doctors, regular rehabilitation and facilities costing up to $40,000 per surgery. An estimation that does not include the cost for re-occurring health issues with the knee such as osteoarthritis or re-tearing of the reconstructed ligament (Mather III, et al., 2013). Although this study does not compare the H-Reflex responses of males and females, it is clear that females are at a higher risk, requiring a higher expense; therefore, females were focused on for the purposes of this study.

If fatigue is shown to have an impact on the H-reflex, it can be inferred that stability of the knee joint may be compromised due to the change in coordination of the reflex, and not simply the inherent strength of the muscle. Once completed, this research is aiming to determine if reflexes have some involvement in knee stability, ultimately helping to promote the introduction of neuronal conditioning into regular training programs of female athletes. Particularly those most at risk who participate in sports with high ACL risk, such as soccer and basketball.

It is important to recognize that there are many factors involved in higher incidence of ACL tears in female athletes, and it is hard to predict ACL injuries or totally prevent them. However, in order to minimize the risk it is important to use our knowledge to help with prevention; whether that be strengthening of the muscles of the lower extremities, or identifying different nerve training techniques to keep the nervous system firing at normal speeds, and even through muscular fatigue.

Chapter Two – Literature Review

Introduction

This chapter provides background and analysis of the current literature on the relationship between the incidence of ACL tears in female athletes and its connection with neuronal control analyzed through H-Reflex elicitation. This review has explanations of knee joint anatomy, the reflex arc; the Hoffman's reflex to explain why this study was created the way it has been described. Furthermore the literature review will provide an analysis of current research in both the H-Reflex as well as studies performed on females to explain the high incidence of ACL ruptures.

Knee Joint Anatomy

Structure of the Knee

A joint is a structure in which two or more bones meet, and where muscles, ligaments, and tendons stabilize actively and passively to secure the joint, respectively. Tendons help facilitate movement at joints by connecting muscle to bone. In the knee there are two tendons anterior to the femur and four posterior. Anteriorly, the patellar tendon attaches the patella to the tibia below the knee, and the quadriceps tendon attaches the quadriceps to the patella above the knee. These tendons help produce movement of the patella, when the muscles contract, producing extension of the knee. Posteriorly, the four hamstrings tendons attach to the tibia and fibula. The "hamstring" is often a term used to refer to the three muscles posterior to the femur: the semitendinosus, semimembranosus and biceps femoris; however, it is actually the name of any of the four tendons contracted by three posterior thigh muscles (Tortora & Derrickson, 2011).

Ligaments provide passive stability to the knee. Ligaments are connective tissues that connect bone to bone and do not produce movement but rather facilitate and restrict it. There is a ligament on both the medial and lateral surfaces outside of the knee joint; the lateral collateral ligament (LCL) that attaches the femur to the fibula on the lateral side of the knee and medial collateral ligament (MCL), attaches the femur to the tibia on the medial side of the knee (Tortora & Derrickson, 2011). The Anterolateral ligament originates at the lateral epicondyle of the femur, and inserts at the anterolateral aspect of the proximal tibia (Claes, Vereecke, Maes, Victor, Verdonk, & Bellemans, 2013). There are two additional ligaments that are intra-capsular but extra synovial; the posterior cruciate ligament, connecting the posterior tibia to the medial condyle of the femur, and the ACL, that crosses anterior to the PCL, attaching the lateral condyle of the femur to the intercondyloid eminence of the tibia. The LCL and MCL restrict movement of the knee mediolaterally and rotational movement; however injuries to these ligaments are most commonly damaged during a contact injury. The PCL restricts posterior tibial translation, anterior femoral translations, with secondary restraint of external rotation of tibia, whereas the ACL resists hyperextension, lateral rotation of the tibia, valgus stress, and anterior translation and medial rotation of the tibia in relation to the femur (Stevens MD, Jarbo MD, Economopoulos MD, & Chhabra MD, 2015).

Although all five ligaments are involved in stabilizing the knee in different ways, the present study was focused on the ACL alone due to the fact that this ligament is more susceptible to be ruptured through non-contact movements (Renstrom, et al., 2008). Convincingly, research results, that will be mentioned, have shown the ACL to be the most susceptible in females as well as having the most debilitating rehabilitation causing season ending injuries in athlete.

Differences between males and females

The possible risk factors increasing the chances of a non-contact ACL injury in female athletes have been studied in great detail. There are extrinsic and intrinsic factors as well as a mixture of both that have been used as explanations including: hyperextension, notch size and shape, hormonal changes, ACL size, coordination of muscle firing, proprioception, footwear, and playing surface, among many others (Ireland, 2003).

Many studies have focused on intrinsic factors as a cause of ACL injury, primarily because these data are more easily collected. Safe landing studies researching the importance of proper landing technique have examined the biomechanics of landing (which is important in sports involving aggressive jumping, landing and changing direction) and how a "safe position" helps to decrease risk factors such as increased Q angle. While in this position, muscles of the lower extremity decrease tibial rotation and therefore decrease risk of injury (Ireland, 2003). Research into injury prone landing positions found that the hip angle (measured as the angle between the trunk and the thigh) was significantly more flexed at foot contact during a one-legged landing manoeuvre that resulted in an ACL disruption, as compared to a safe landing position which did not produce these problems (Sipprell, Boden, & Sheehan, 2012).

Andrews and Axe (1985) introduced the concept of ligament dominance whereby the musculature in the lower extremities does not adequately absorb the forces produced during sports manoeuvres, resulting in excessive loading on the ligaments of the knee, especially the ACL. They reported that this ligament dominance often resulted in valgus knee movements and excessive knee valgus motion. Further, another type of dominance encountered in athletes was quadriceps dominance, an imbalance in the recruitment patters of the hamstrings and quadriceps. Hewett (1996) and Huston (1996) both described quadriceps dominance more commonly in females, who rely more on their quadriceps than their hamstrings to produce dynamic knee stability during jumping and landing, two movements that increase the chance of ACL injury. Along with leg dominance, whereby an athlete has one leg stronger than the other, which is commonly seen in soccer due to the occurrence of a standing and a kicking foot, these two types of dominance increase the risk of ACL tears. Within the literature, strength and imbalance differences reported between males and females provide possible answers as to why females attain more ACL injuries, in comparison to males.

Research has also indicated differences between males and females performing landing and cutting tasks, which may predict ACL injuries. Zazulak et al. (2007) examined how the gluteus maximus and gluteus medius act synergistically to stabilize the pelvis in all planes of motion and control rotation at the hip, especially in single-leg stance (the stance most common in the tearing of the ACL). While the gluteus medius is the primary abductor of the hip, the gluteus maximus functions primarily as an extensor and, secondarily, as an external rotator of the hip. Decreased activation of proximal stabilizing muscles may lower load-bearing capacity of the knee joint and predispose it to injury (Zazulak, Hewett, & Reeves, 2007). This study found decreased gluteus maximus activity during landing in female soccer, basketball and volleyball players compared with males, using EMG (Mendiguchia, 2011). A difference that could possibly account for increased knee instability within female athletes.

Additional studies have gone into depth investigating different types of movement such as running and side step cutting (as well as landing maneuvers), and how these movements can increase or decrease the likely hood of injury to the knee as well as how these movements change comparing males and females. Ferber et al. (2003) reported significantly greater hip internal rotation angles and hip negative work in the transverse plane during running in female recreational athletes compared with males. Landry et al. found that females produced a greater overall hip external rotation moment than did male subjects during the early stance phase of the unanticipated straight run, rotations, which may cause stress on the ACL during activities where the femur and tibia may rotate in different directions. Furthermore, during side-step cutting the contribution of the hip musculature in the movement is supported by findings of McLean et al., who showed that the peak knee abduction moment was more sensitive to initial hip internal rotation and knee abduction position in females compared with males. These findings are in agreement with those reported by Sigward and Powers who found significantly greater hip internal rotation and greater internally rotated foot at initial contact in the excessive valgus moment compared with a normal valgus group during side-step cutting (Mendiguchia, 2011), ideas that agree with Ferber and Landry on the instability of the knee joint in females.

Another anatomical factor that has been studied and has shown gender differences is Q angle. The Q-angle approximates the resultant force orientation of the four muscles of the quadriceps group acting on the patella. The Q-angle is defined as the angle between a line connecting the center of the patella and the patellar tendon attachment site on the tibial tubercle and a second line connecting the center of the patella and the anterior superior iliac spine on the pelvis, which the knee is fully extended (Mizuno, et al., 2001).

One study comparing male and female NCAA basketball players found that the greatest magnitudes of force were incurred by the ACL at 30 degrees of knee flexion. Q-angle is often associated with increased tibial internal rotation. The ACL functions to prevent internal tibial rotation; thus, at 30 degrees of flexion, if internal rotation is increased in females and the eccentric hamstrings-to-eccentric quadriceps strength ratio is diminished, during deceleration the knee is incurring two forces that compromise the integrity of the ACL. The combination of structure and strength may predispose females to a greater incidence of ACL injuries (Moul, 1998). However, due to recent research into the Q-angle and the likelihood of multifactorial reasoning for ACL tears, it is unlikely that an anatomical factor, such as the Q-angle, is the only reason as to why females have shown higher risk (Posthumus, Collins, September, & Schwellnus, 2011).

A final anatomical factor that may contribute to differences between males and females is the size of the ACL, which is directly correlated with the size of the intercondylar notch (located on the femur). Many researchers have described a mechanism by which the intercondylar notch of the distal femur can tear the ACL with hyperextension, as well as intercondylar notch stenosis being a factor in ACL injuries. Houseworth et al (1987) and Anderson et al (1987) used computer tracings and CT scans to measure the notch and found the before mentioned correlation between intercondylar notch size and ACL injuries. Furthermore, a study measured intercondylar notch width of 902 athletes and found that across all athletes, those who had previously sustained ACL noncontact injuries had significantly narrowed intercondylar notches (Souryal & Freeman, 1993).

The role of hormones in the susceptibility of female athletes to ACL injuries is an area of much study because while hormonal influences have been extensively researched, this factor has been one of the hardest areas to measure due to all the variables involved. There has been no conclusive evidence to date on how hormones are a factor. For example, some studies have presented a link between hormones and ACL injuries while as others have not. Mendiguchia et al. (2011) conducted a study in which self-reported menstrual histories with salivary sex-hormone profiles at the time of ACL injury were collected. They found that 26 of 37 athletes tore their ACLs during the follicular phase of the menstrual cycle. Among athletes who self-reported their menstrual histories, 10 of these 27 injuries occurred during the few days before and the 2 days after the onset of menses. It was concluded that ACL injuries occurred most frequently on days 1 and 2 of menses, suggesting that ACL injury is not random but occurs more often around the time of menses, when circulating sex-hormone levels are low (Mendiguchia, 2011). However an earlier study (Wojtys, Huston, Boynton, Spindler, & Lindenfeld, 2002) had different results, indicating a significantly greater than expected percentage of ACL injuries during midcycle (ovulatory phase) and a less than expected percentage of those injuries during the luteal phase of the menstrual cycle. Furthermore, both studies reported that those who took oral contraception had completely different results. Clearly there is more research to be done in the area of hormonal research to determine if it is a legitimate risk factor in females.

Motor Units and Muscle Fiber Types

The Central Nervous System (CNS) is responsible for the production of coordinated movement through the recruitment of motor units to activate muscles. A motor unit is made up of a motor neuron and the skeletal muscle fibers that are innervated by those neuron's axonal terminals. Usually groups of motor units work together to coordinate contractions of muscles. A group of motor units in a muscle are labeled the motor pool. The recruitment of motor neurons occurs in an orderly fashion, starting with the smallest motor units and building up to the large ones if necessary, as the force needed increases; this principle is Henneman's size principle.

Skeletal muscles that are recruited by these motor units also have different properties, when comparing across a population. One of the quadriceps muscles of a soccer player may have a larger percentage of fast twitch fibers than slow twitch compared to a different player, and these different muscle fiber types have different characteristics. Type I and II fibers have distinct differences; type I has a higher resistance to fatigue and large blood supply but small twitch force and slow twitch speed, whereas type II has the inverse of these characteristics (MacIntosh, Gardiner, & McComas, 2006). In the VM specifically, a 10-year study of 55 men and 95 women found a wide range of differences in the percentages of these fiber types. Across the testing population there were some individuals with high percentages of type I, and others with high percentages of type II. But individuals with a percentage of Type I fibers less than 35% were found almost twice as often for the men (38%) compared to the women (20%). This study not only exhibited a large variance in fiber type in the VM, but also a statistical difference between males and females (Staron, et al., 2000). With soccer having both a longer aerobic aspect as well as short, explosive sprints it is likely that both fiber types are required for the sport. Further, different positions require different types and amounts of activity. For example, a goalkeeper is predominantly an anaerobic position, compared to a midfielder who will do the most aerobic running on the team. Therefore it can be inferred that each soccer player may have different ratios of fiber types in order to be successful at their position.

Reflex Arc

A reflex arc is a neural pathway that controls an action reflex and is a protective measure animals have developed to minimize damage – in this case, damage of the joints. In vertebrate animals, most sensory neurons do not pass directly into the brain, but synapse in the spinal cord. This characteristic allows reflex actions to occur through monosynaptic action by activating spinal motor neurons without the delay of routing signals through the brain or even interneurons in the spinal cord. Type Ia and Ib are two types of sensory neurons that operate via monosynaptic pathways. Type Ia detects the length changes of muscle spindles, which depolarize the neuron allowing signals to be sent to the spinal cord. Within the spinal cord acetylcholine (ACh) is released at the synapse causing an excitatory postsynaptic potential (EPSP), leading to the excitation of the effector neuron (alpha), which initiates the reflex when overstretching of the muscle occurs. While this is occurring an inhibitory postsynaptic potential (IPSP) in the motor neuron of the antagonistic muscle is produced through GABA neurotransmitters (Purves, Augustine, Fitzpatrick, Hall, LaMantia, & White, 2011). This action relaxes the muscle in opposition to the overstretched muscle, allowing contraction and decreasing risk of overstretching. In the case of protective measures in the knee, any overstretching in the muscles around the joint are unconsciously controlled by the actions of reflexes to correct unwanted changes in joint movement to protect the area from injury. In work by Moore et al. (2002), decreases were noted in females in reflex amplitude of the patellar tendon tap reflex following fatigue. Further, Myers et al. (2003) found similar decreases in muscle protective responses when subjects were injected with a solution to simulate excess fluid in the joint and they concluded that dynamic stability may be compromised due to the decrease in the reflex response, helping to explain the evidence that many injuries occur later in competition, when athletes are fatigued.

As described above, there are many factors that could contribute to why some people are more susceptible to ACL tears than others, fatigue being one of them. Recent research has shown that many of these factors may be exacerbated by fatigue or neuronal factors, for example the Q angle. This may help explain how male athletes with above average lower extremity strength, for example NFL football players, are still capable of obtaining non-contact ACL injuries. This research is focused on the influence of fatigue on the neuronal response and the nerves ability to recruit muscles for protection, while fatigued.

Hoffman's Reflex

In the current study, the Hoffmann's Reflex (H-Reflex) was used to test the action of the stretch reflex within the quadriceps and to determine how human reflexes are affected due to fatigue. The H-Reflex is the electrical equivalent of the stretch reflex in a monosynaptic pathway elicited by electrical stimulation of afferent Ia axons. A stimulus of a low amplitude and long duration (1ms) stimulate the Ia afferent axons but not the efferent, bypassing the muscle spindle. Within 10-40 milliseconds of nerve stimulation, depending on the muscle's distance from the spinal cord, the H reflex can be seen on an EMG recording, which measures the electrical activity of the muscle. The soleus H-reflex has been well researched and consistently appears on EMG recordings at a latency of approximately 30 milliseconds after stimulus delivery, whereas the VM H-reflex, the target muscle in this study, appears after approximately 15 milliseconds (Palmieri, Ingersoll, & Hoffman, 2004).

Action potential conduction within muscle fibers generates a short latency EMG response in the muscle called the M-wave. At a higher voltage stimulus the H reflex disappears due to action potentials generated through sensory fibers and those travelling in the opposite direction colliding preventing reflex signals. More motor fibers are stimulated in this case and more collisions occur so H reflex decreases and M wave increases.

The H-reflex is an estimate of alpha motor neuron (αMN) excitability when presynaptic inhibition and intrinsic excitability of the αMNs remain constant. This measurement can be used to assess the response of the nervous system to various neurologic conditions, musculoskeletal injuries, and application of therapeutic modalities, pain, exercise training, and performance of motor tasks (Knikou, 2008).

This H-reflex has been used clinically and in research for many years. Clinically, the most common way in which this reflex has been used is to evaluate the gastrocnemius-soleus complex in the lower leg. Palmieri et al. (2004) reported that the H-reflex is used to test for bi-lateral differences in the gastrocnemius-soleus complex as this difference is thought to be an early indication of spinal stenosis. In addition to the use of the H-reflex in clinical studies, Misiaszek (2003) describes the used of the reflex as a neural probe. In review they were able to describe numerous ways in which the H-reflex is used; for the investigation of the functional organization of neural circuitry, to monitor change in state of spinal excitability, for modulation of presynaptic inhibition of Ia afferents, to measure motoreuron excitability, and for adaptation in spinal structures in health and disease.

Validity and reliability of H-Reflex measurement

Reliability of H-Reflex

A study performed by Christie et al. examined the intraclass reliability of the latency and amplitude of the H-reflex in the flexor carpi radialis (FCR), a flexor muscle of the wrist. It was demonstrated in this study that the H-reflex can be easily evoked in the FCR and that measures taken from these recordings (amplitude and latency) are reliable. They showed that the H-reflex is an effective means of assessing changes and damage (specific to this study) to the nerves. These results, therefore, have important implications whereby informative recordings can be collected from the H-Reflex with relative ease and consistency. Additionally, the high reliability of the measures suggests that investigators can be confident that any observed changes in amplitude and/or latency are due to changes within the individual and not due to error in the measurement (Christie, Inglis, Boucher, & Gabriel, 2005).

The H-reflex can be utilized to assess modulation of spinal inhibitory interneuronal circuits, but attention to the factors that affect Ia transmission is needed. The H-reflex is not hard-wired but is dramatically modulated during various motor tasks (task dependence) or during different phases of a cyclical movement (e.g. cycling and walking), and can be affected by several factors such as voluntary contraction and background noise, that must be acknowledged to avoid misinterpretation of the data (Knikou, 2008). The H reflex can certainly be used as another useful tool in evaluating the changes in human reflex pathways and the plasticity of the neuromuscular system with considerations in the frequency of stimulation to avoid post activation depression, the behavioral state of the participant and the normalization of data collection. (Zehr, 2002). In other studies peripheral nerve stimulation of the soleus produce reliable results and that collecting data on a curve is adequate in seeing consistent reflex measures. This presents a high reliability allowing us to test changes in motor neuron pool excitability (Palmieri, Hoffman, & Ingersoll, 2002). Reflex magnitude can change dramatically during contraction or stretch of agonist and antagonist muscles. If these limitations are recognized and addressed, the H-reflex will remain one of the major probes for studying sensorimotor integration and training-induced neural adaptation in health and neural pathology (Knikou, 2008).

Methodological concerns / limitations in H-Reflex assessment

Assessment of the H-Reflex requires electrical stimulation to a peripheral nerve and then recording the action potentials on an EMG of the muscle that is being innervated. Many factors may affect the type of data that is collected which include but are not limited to, electrode placement, muscle tension, type f stimulation, intensity and duration of the stimulation, and any other outside distractions. Therefore it is a difficult procedure to standardize due to the differences between subjects in each test. This is why this study has a within subjects design, in order to normalize the data across participants.

One of the major limitations in using the H-Reflex is that an electrically induced reflex does not occur normally within the human body and the muscle spindle is completely bypassed in the process. Muscle spindles are thought to adjust reflex output during movement and therefore are extremely important in determining muscle output during body movements (Palmieri, Ingersoll, & Hoffman, 2004). Although there is a direct connection between Ia afferents and motor neurons, it has been interpreted in the literature that the Hoffman's reflex accurately represents motor neuron excitability. This conclusion is inaccurate, because the synaptic connection between the Ia afferents and motor neurons is subject to presynaptic modification. Presynaptic inhibition alters neurotransmitter release at the Ia-motor neuron synapse and can result in a decrease in the H-Reflex with no change in motor neuron membrane potential and conductance. In humans, changes in presynaptic inhibition have been observed due to joint effusion, voluntary contraction, postural adjustments, and cortical stimulation. In addition, presynaptic control of movement differs with age and training type (Palmieri, Ingersoll, & Hoffman, 2004).

Further, many studies have used the assessment tool of the H-Reflex/M-wave ratio as a standard of describing the H-Reflex response, maximum point etc. However it has been found that various joint positions are critical in control of the M response. Therefore interpretation of any H-Reflex responses is simple to decipher only when the M response is independent of the joint position. Unfortunately, research has shown that joint angle factors (angle changes), such as the change in diameter of a muscle, may change the elicited response (Hwang, 2002). Therefore, this study used the raw data of the H-Reflex, rather than a comparison of its voltage to the M response, to reduce the amount of factors that had to be accounted for.

It is also important to understand that the spinal cord circuitry is oligosynpatic (Pierrot-Deseilligny & Burke, 2005). Any neural input from the supraspinal and peripheral pathways can potentially influence the H-reflex modulation through either direct regulation of the motoneuron membrane potential or indirect regulation of interneuronal circuitry (Misiaszek, 2003). Furthermore the participant's physiological and psychological state during the experiment can affect the H-reflex results. It has been demonstrated that postural anxiety can depress the H-reflex modulation (Sibley, Carpenter, Perry, & Frank, 2007). Further, the H-reflex response may not be observed in all experimental trials for all participants. It is possible that the H-reflex test is unsuccessful in a small proportion of a healthy population under some experimental conditions.

In summary, H-reflex is a short-latency reflex, which has been used to examine the effects of Ia afferent inputs on motor neuron activation. High test-retest reliability of H-reflex has been reported in the literature however a within subjects study is the best way to go about analysing the reflex, due to the variance in outside factors such as electrode placement, distractions etc. that may slightly alter results from subject to subject (Knikou, 2008).

Influence of Fatigue

There are many different reasons why females may be tearing their ACLs; however, several recent studies have tried to pinpoint the effect of fatigue on injuries, and now are commonly trying to use fatigue to explain the gross amount of injuries to the ACL. Research has been aimed towards females, specifically, in order to explain the large difference between males and females and the occurrence of this particular injury.

When a muscle/motor unit is fatigued, there is a failure to maintain the required or expected force (Edwards, 1981). As a muscle is progressively more fatigued, the ability to maintain a constant force is accomplished, at least in part, by the recruitment of additional motor units, which often results in an increase in surface electromyographic (EMG) amplitude. Stock (Stock, Beck, & Defreitas, 2012) showed that quadriceps muscles actually fatigue at different rates, the VM being slightly more resistant to fatigue than the VL. With this research in mind it could be inferred that if muscles fatigue at different rates there is a higher likelihood of instability in the knee. This is one of the reasons why a fatigue test was incorporated into the present study.

Muscle fibers have a decreased capacity to absorb energy when fatigued, thus causing an athlete to land with altered neuromuscular function. Therefore, neuromuscular fatigue can impair the effects of dynamic joint stabilization and the body's inherent protection from injury, regardless of muscular strength and imbalances, or the lack there of. The H-reflex decrease during the recovery period could be related to the presence of metabolites produced by the muscle during fatigue. On the spinal level, the post-synaptic inhibition of the antagonistic muscle increased during a maximal fatiguing contraction (Kukulka, Moore, & Russel, 1986), and decreased during submaximal (20% MVC) (Löscher, Cresswell, & Thorstensson, 1996). In other words, inhibition increases the excitability of the motor neurons during submaximal fatigue and the opposite happens during maximal fatigue. This evidence indicates that while fatigued, the protective measures that stop over-stretching in the target muscle, react differently depending on how the athlete was fatigued.

Fatigue further causes large increases in initial-contact and peak stance-phase knee abduction and knee internal rotation motions and in peak external knee-adduction, abduction, and internal rotation moments (McClean, 2007). Movements that if rotated far enough internally or externally, could put stress on the stabilizing ligaments of the knee. A significant amount of research continues to focus on the likely role of altered or abnormal neuromuscular control, when fatigued, as the underlying injury mechanism. Despite the ever-increasing number and complexity of programs that have aimed to reduce risk of ACL injuries, injury rates and the sex-based disproportion have continued. It appears therefore, that the success of current ACL injury prevention strategies may be limited by an incomplete understanding of the mechanism of injury and a mechanism of fatigue in general (Borotikara, Newcomera, Koppesa, & McLean, 2008). Therefore this is an area that is open for study, and one that could help in the understanding of traumatic non-contact ACL tears.

Furthermore the complexity of the knee joint adds to the difficulty in pinpointing reasons for injury due to the many different muscles that support the joint. In this study we specifically focused on the VM muscle, but in recent studies it has been found that isolated fatigue of both the quadriceps and hamstring musculature in young healthy subjects precipitated potentially hazardous anterior tibial translation increases in the presence of an externally applied anterior loads. And that the order of muscle activation of the quadriceps did not change under fatigue, muscle premotor or reaction phases was noticeably greater in one study, suggesting a possible compromise in their protective role (McClean, 2007). Localized quadriceps and hamstring fatigue also have been found to induce significant changes in female lower-limb control during crossover cutting tasks, a much discussed link to noncontact ACL injury.

More recently, generalized neuromuscular fatigue has been suggested to increase ACL injury risk during stop jump tasks, primarily via promotion of potentially hazardous anterior tibial shear loading. This point seems particularly pertinent because hazardous knee loads are known to stem from altered control elsewhere in the lower limb, particularly in females. Examining neuromuscular fatigue effects across the entire lower limb during high- risk sports movements thus seems crucial to determining the role of fatigue within ACL injury and to enhancing the efficacy of current injury-prevention strategies.

Summary

In summary, it is clear that there is still an uncertainty as to how all of the factors stated factors contribute to the incidence of non-contact ACL injuries. Furthermore, although there is research into why females are particularly susceptible to ACL injuries, it is still uncertain as to how all the factors mention contribute to explain this. There is doubt as to how fatigue and neuromuscular factors provide any explanation to the aforementioned problem; this is why this study took place.

Chapter 3 – General Methodology

Subjects

All participants were between the ages of 18 and 22 years old and were screened for current musculoskeletal injury of lower extremities or back, lingering symptoms of musculoskeletal injury in lower extremities or back, current musculoskeletal pain in the lower extremities or back, chest pain with exertion, shortness of breath, known hypertension, history of heart attack, or pregnancy. All subjects were also examined by a Certified Athletic Trainer employed at the University of Idaho due to NCAA division I rules for student athletes requiring a physical checkup by July. Therefore, any participants disqualified for soccer activity due to lower extremity/back pain or dysfunctions were disqualified for the study also. All participants were also current members of the University of Idaho Division I Women's Soccer team at the time of testing.

Recruitment of Participants

Participants were recruited from the University of Idaho NCAA Division 1 Women's Soccer team. Athletes on the University of Idaho NCAA Division I women's soccer team were asked to volunteer to attend an information session with a brief description of the research study. Those that volunteered attended a session to introduce them to what the procedure entails, practically, and provide informed consent once they agreed to continue (example of flyer attached in the IRB Form).

Methodology

A surface electrode was placed on the femoral nerve at the Scarpa or Femoral Triangle where the nerve was most superficial. The stimulating electrode was attached to a large conducting pad that was moistened with a sponge and placed on the gluteus maximus. Two surface EMG electrodes were placed 2cm from each other on the VM, close to the knee where the muscle was most superficial. These electrodes were used to measure muscle activity in the VM muscle. The ground electrode was placed on the patella.

After some trial and error and the onset of the H-reflex was found, the nerve was stimulated at increments of 0.2 millivolts up to the max H-reflex. The EMG wave was recorded for 3 seconds, with nerve stimulation at 1 second (Figure 1). Three measures for the H-reflex were collected for each voltage in order to calculate the mean, which was used in analysis of the data.

During H-reflex stimulation the participants were told to lay down and relax. The environment in the lab was controlled so that no background noise was present during the activity, as well as minimal talking and voluntary movement. These factors were controlled in order to keep the environment constant across participants, reducing any factors that would affect the H-Reflex.

Participants had their H-Reflex elicited until the maximum was found and then was stimulated in the same increments back down to the original onset. Following this, muscular fatigue in the target muscle (VM) was achieved through one bout of a Wingate test. This standardized test required the participants to pedal as hard as possible on a stationary bike against a resistance of 8.6% of their body weight for 20 seconds. To calibrate these measurements, body weight was measured before testing. Muscle activity was then again recorded via the EMG, post Wingate test, with the stimulating electrode again being used to elicit the femoral nerve H-Reflex. In order to compare across trials all pf the skin electrodes remained on the participant through the Wingate test. This made sure that the EMG was recorded from the same location on the VM, pre and post and that the H-reflex was stimulated at the same location on the nerve.
Post fatigue, the same voltage increments, or as close as possible, were used from pre fatigue for comparison in data analysis. Each voltage increment was repeated three times to find the mean for analysis to help in answering the research question of this study, do changes in excitability of the spinal reflexive pathways of the VM muscle change once the target muscle of a female NCAA division I soccer player is fatigued.

Data Processing and Analysis

H-reflex data from before and after fatigue was compared for each participant to examine differences in response and stimulus strength needed to elicit the response. A script was written in Matlab in order to convert the AcqKnowledge data into excel sheets for each participant, which included information represented in the graph in Figure 1 (Appendix A).

Once the data was analyzed, two graphs were produced for each participant in order to visualize the results. The first graph plotted the voltage at which the nerve was stimulated on the x-axis, against the average magnitude of the positive peak from trials 1 and 2 on the y-axis. For the second graph the data was normalized so a comparison could be made across participants. The graph plotted stimulus voltage/average onset of H-reflex vs. H-reflex magnitude/peak pre H-reflex magnitude. The stimulus voltage/average onset of H-reflex produced a ratio; 1.0 calculated as the onset of stimulation. This value was plotted on the x-axis against the magnitude of the positive peak (Appendix A – figures 5 and 6). Figure 1 gives an example of a representative EMG trace showing how the H-reflex was analyzed. The letters represent the following: a) the stimulus artifact at 1 ms b) the M-wave c) the start of the H-reflex d) the H-Reflex peak e) the end of the H-reflex. The magnitude (mV) and timing (ms) of each time point was collected using MatLab and converted into Excel to produce the graphs.

Pre and Post for trials 1 and 2 were averaged together to produce an overall mean, which was used to graph the results shown in figure 5 (Appendix A). The data was also then normalized, plotting stimulus voltage/average onset of H-reflex vs. H-reflex magnitude/peak pre H-reflex magnitude in order to more easily compare the results (figure 6). A repeated measures ANOVA test was used in data analysis to calculate p-values, providing information about whether our results were significant.

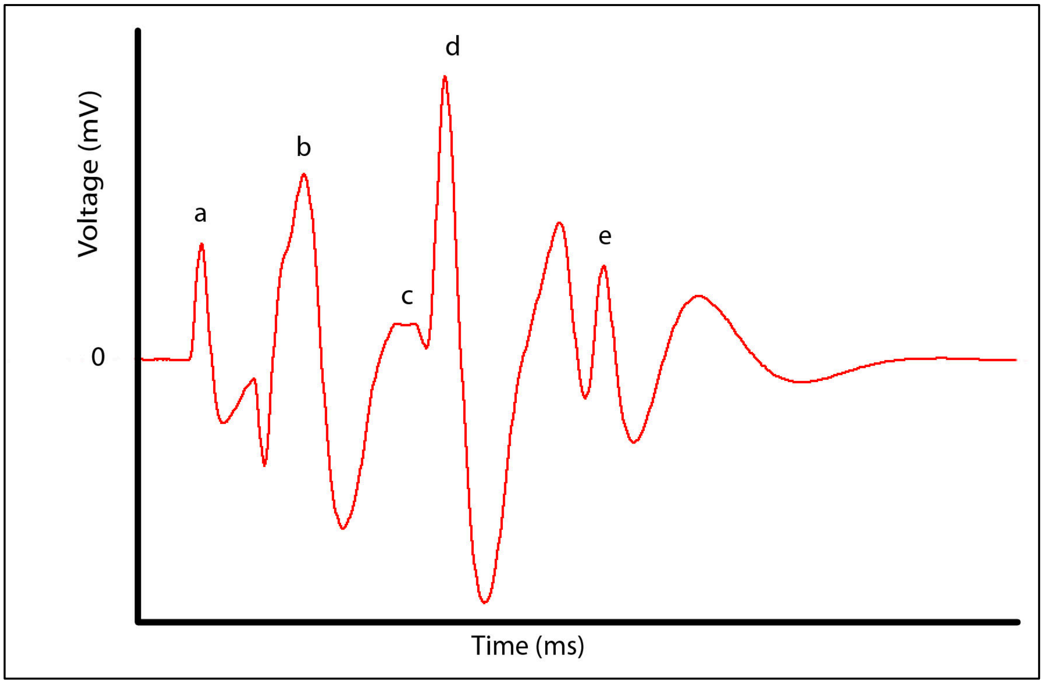

Figure 1: Example of an EMG trace seen during data analysis. The letters represent the following: a) the stimulus artifact at 1 ms b) the M-wave c) the start of the H-reflex d) the H-Reflex peak e) the end of the H-reflex. The magnitude (mV) and timing (ms) of each time point was collected using MatLab and converted into Excel to produce the graphs.

Chapter 4 - Results & Discussion

Relationship of Subject Fatigue to H-Reflex latency

Our results showed that H-Reflex latency is related to the subjects' fatigue. Each participant's raw and normalized data are plotted in figures 5 and 6, respectively (appendix A), but to present this data more effectively, figure 2 was produced. Here each participants H-reflex normalized onset was averaged to produce the bar graph, below. The onset of H-reflex in pre-fatigue is calculated at 1.0, with 1.0 being the ratio of H-reflex magnitude/peak pre H-reflex magnitude. The post H-reflex peak was divided by the same peak pre H-reflex magnitude to produce a ratio for post. The data represented in Figure 2 shows that there was a significant difference in the voltage needed to produce the H-reflex between the pre and post fatigue states. A repeated measures ANOVA test calculated a p-value < 0.001, which presents a significant difference in the onset of H-reflex.

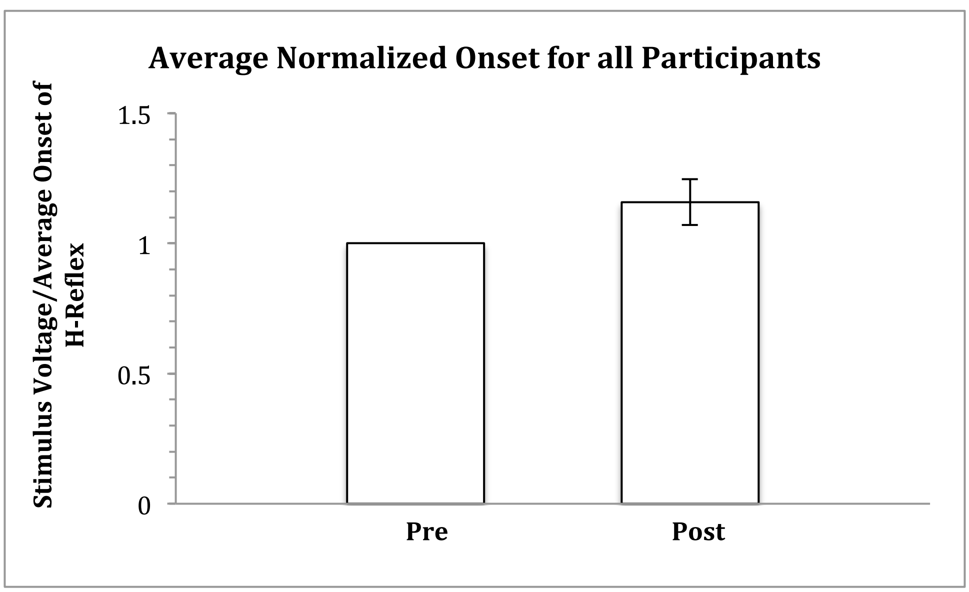

Figure 2: Graph representing the average normalized onset for all participants. Using a repeated measure ANOVA a significant p value of less than 0.05 was calculated between the average onset comparing pre and post trials.

To further illustrate the significant difference in the onset of H-reflex the normalized data for all participants were graphed together, shown in figure 3 below. The red lines represent pre-fatigue results and blue lines represent post-fatigue results. The graph illustrates the significant differences between the two states where the red lines ascend at the onset of 1.0 but the blue lines, on average, ascend around 1.2 (represented in figure 2). The data on this cumulative graph (figure 3) demonstrates an overall decrease in the sensitivity of the H-reflex, post fatigue, but it also represents the variability between the participants. That even though the data is normalized there is an array of maximum H-reflexes.

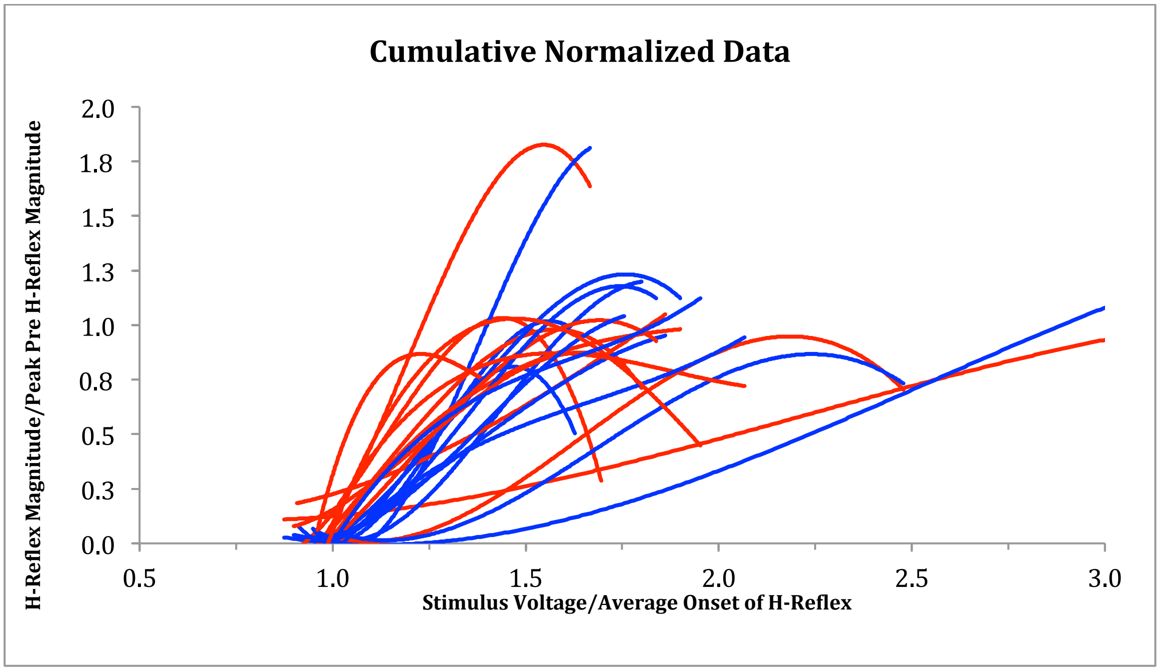

Figure 3: Graph showing the normalized data H-reflex magnitude/peak pre H-reflex magnitude vs stimulus voltage/average onset of H-reflex, for all participants. Red lines represent pre-fatigue results and blue lines represent post-fatigue results.

Changes in the Wave Form

Although there was a significant change in the stimulus needed to elicit the H-reflex between pre and post fatigue, there were no differences when comparing the waveforms of the two conditions. Timing data are represented in table 1. It can be seen that there is no significant difference between the pre and post results for each of these measures indicated by the repeated measures ANOVA producing a p-value higher than 0.05.

There was also no significant difference in the average magnitude of the peak H-reflex when comparing across all participants. The average pre and post magnitudes were plotted in a bar graph (figure 4), where a p value of 0.58 was calculated using a repeated measures ANOVA.

Table 1: Table showing the average time of peak, average start time, average end time and average duration of the H-reflex across all participants, pre and post. The repeated measured ANOVA calculates that between participants there was no significant difference between pre and post for each time point.

	Pre	(s.d.)	Post	(s.d.)	p-value
Time of Peak (secs)	0.021	(0.003)	0.021	(0.002)	0.296
Start Time (secs)	0.020	(0.002)	0.020	(0.001)	0.852
End of H-reflex (secs)	0.032	(0.002)	0.032	(0.001)	0.331
Duration (secs)	0.018	(0.004)	0.019	(0.003)	0.204

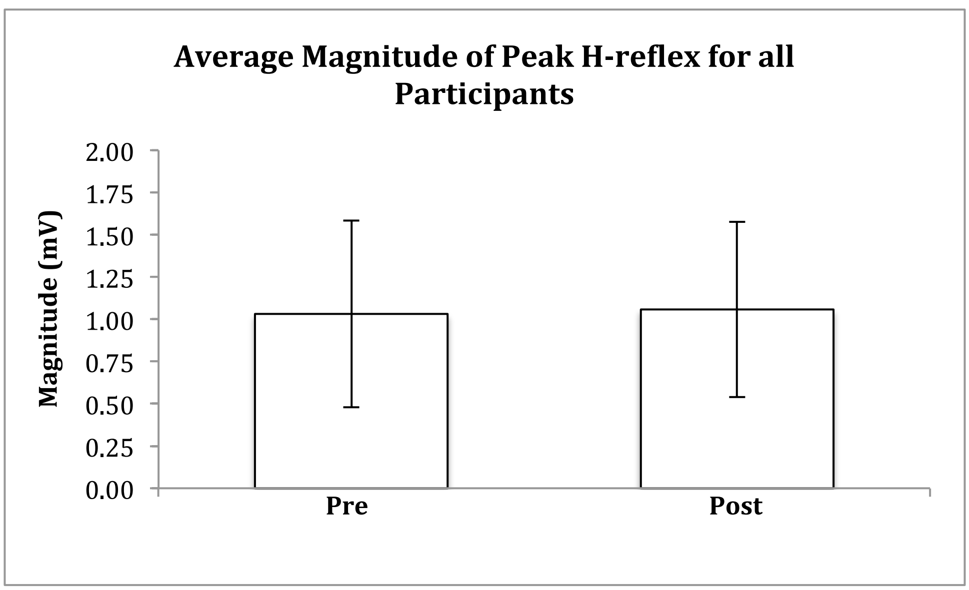

Figure 4: Graph showing the average magnitude of the peak H-reflex for all participants, pre vs. post. A p value >0.05 was calculated showing there is no significant difference between the magnitude of the peak H-reflex pre and post fatigue.

Discussion and Conclusions

Females are 2-8 times more likely to tear their ACLs than their male counterparts (Mandelbaum & Silvers, 2005) indicating a high level of risk for females to participate in sports that include frequent cutting, landing, acceleration, and deceleration. This suggests that there is gender specificity in the risks associated with noncontact ACL injuries, demonstrating a need for research in the area. But what is the mechanism for non-contact ACL injuries? The purpose of this study was to determine if fatigue had an impact on reflexes in order to understand if muscle coordination could play a factor in non-contact ACL injury occurring late in games and/or seasons. The main finding was that the H-reflex onset was delayed in the post trials, meaning that in a fatigued state the nerves require a higher stimulus to elicit the H-reflex.

Timing of the H-reflex

The time between the stimulus and peak H-reflex did not show a significant change when comparing pre and post trials. Furthermore, the start, the end and the duration of the H-reflex between pre and post trials presented no significant difference in data, either. Although these were factors that were predicted to change based on review of the literature (Knikou, 2008), this study has shown that there are other factors that change with fatigue and that are associated with the neuronal control, and therefore changes in these factors cause decreases in stability of the knee, post fatigue. A change in the time of the peak H-reflex relative to the stimulus could indicate changes in the conduction velocity of the nerve or a delay in response from muscle spindle to the sensory nerve. However, the change in onset voltage, reported in this study, can still be used to partially accept the proposed hypothesis that fatigue affects the H-reflex. From this study's findings it can be deduced, for this specific population of trained soccer players, that once the stimulus hits the H-reflex threshold, there was no significant difference in the conduction of the nerve; that the main change in H-reflex was due to the magnitude of the stimulus needed to produce the initial reflex was increased in a fatigued state. This change could be due to many reasons, one of them being a hypo-polarization of the nerve itself due to metabolites within the axon produced during fatigue. This could indicate that more sodium ion channels need to first produce a more positive resting potential and then in order to depolarize it and cause an action potential and contraction. This theory could explain the latency in the reflex with the need of a higher contraction to open more sodium channels, whereby the relationship between sodium channels and the H-reflex conduction velocity has already confirmed (Carp, Chen, Sheikh, & Wolpaw, 2001). A further reason could be because of changes in the synapse in the spinal cord due to fatigue. Neurotransmitters in the spinal cord could have a decreased re-uptake at the synapse due to increased use during the exercise, in this case the 20 second wingate bike test, or again a change in the charge of the pre-synaptic terminal due to metabolites produced during fatigue. These changes could cause a decreased sensitivity to the stimulus in the fatigued state.

Stability

The significant change in the sensitivity of the nerve leading to a different H-reflex onset reported in this study may have implications for stability within the knee joint. Muscles provide active stability to the knee, the quadriceps and hamstrings working in antagonistic action to facilitate movement at the knee joint but also to stop unwanted movements at the knee, which may cause injury. Ligaments and tendons work to aid in the movement and also provide passive stability to the knee, keeping bones and muscles connected. With a decrease in active muscle control, more pressure and strain is put upon the tendons and ligaments to keep the knee joint from twisting, a movement that can cause rupture to the connective tissues at the joint. The muscle spindles help prevent injury by first detecting over stretching and stretch velocity in the muscle it resides in, and eliciting contraction via the stretch reflex. This protective measure helps the muscle by eliciting a contraction to reduce the chance of muscle tearing.

The H-reflex is analogous to the stretch reflex; therefore a change in H-reflex elicitation due to fatigue can help in the understanding of how this protective measure is affected by fatigue.

Once participants were fatigued, our results illustrate that a higher stimulus was needed to elicit the H-reflex relative to the rested state. With a decreased sensitivity of the H-reflex it can be inferred that the muscle spindles residing in the fatigued muscle would require a higher muscle stretch in order to illicit a reflex, contracting the over stretched muscle. If this is the case, and the active stability of the muscles is compromised, it can be inferred that more stress will be put onto the ligaments and tendons to keep the joint stable, when the joint is twisting, over stretching, or over flexing. Therefore the joint will be at risk of rotating further before muscles are activated to re-stabilize the joint. The ACL helps to resist medial rotation of the knee joint; however, if all of the pressure to resist this movement is now placed on the ligament, due to a delay in muscle reflex response, it is likely that increased medial rotation could occur and cause injury to the ligament.

There are other factors that could be affected by a change in the H-reflex, the Q-angle being one of them. This angle is larger in females than males and therefore may be associated with increased knee valgus movement. If this anatomical factor causes movement that results in anterior cruciate ligament strain, and the muscles are also fatigued to the point where the contribution it has to active stability has decreased, an argument can be made as to how females may be more susceptible to ACL injuries than males. With the two factors contributing to higher valgus movement, and less muscle stabilization then, when cutting, jumping, accelerating and, decelerating high force may be put on the supporting ligaments of the knee to resist unnatural movements.

In addition, due to the restraints of the protocol in this study those soccer players that had previously torn their ACLs were disqualified from testing. Only testing athletes that were injury free may have skewed the results. A future study should focus on the comparison of previously injured and healthy soccer players to see if there is a difference in neuronal patterns that may have caused those injured athletes to be susceptible to ACL injury. Further, those healthy athletes who were tested should be monitored over time to see if any acquire ACL injuries. The H-reflex data for the previously healthy athlete who recently tore their ACL could be reviewed to see if any precursors for injury could be seen, such as large changes between their pre and post H-reflex.

Comparison to H-Reflex Studies

The H-reflex has been used as a tool to monitor the neuronal control of a muscle in multiple studies as well as being used to compare how the reflexes are affected by fatigue. Patikas et al. (2006) sub maximally fatigued the soleus muscle of untrained subjects and studied the results in the H-Reflex. Patikas' study of fatigue produced significant increases in magnitude of the H-reflex during exercise but when the protocol was over and fatigue had set in, the H-reflex was depressed for the first 3 minutes, relative to the H-reflex that was recorded before fatigue. The results of the study suggested that the reflex excitability is increased as activity develops, whereas this increase turns to depression for the first minutes of the recovery phase (Patikas, Bassa, & Kotzamanidis, 2006). The study in question shows similar results to this current study whereby during the recovery phase of exercise the H-reflex is altered in a way that it is depressed, potentially compromising knee joint stability. Although the reflex was not monitored during exercise in this study, the results of Patikas's study support those of this study and help to further knowledge in the area, that fatigue has an impact on neuronal control in muscles.

Furthermore, a number of fatigue studies have analyzed the H-reflex research into tibial translation, how far the tibia moves under the femur, as a way of determining the impact of fatigue on neuronal control. An example of such a study by Melnyk and Gollhofer (2006) found that increases in anterior tibial translation are commonly associated with higher risks of injury for athletes. Their results confirmed earlier findings that muscle fatigue induces a decrease in knee joint stability, due to the increase in tibial translation when the thigh muscles are fatigued. They described a widespread agreement that exists between the reviewed studies, in spite of different fatigue protocols, suggesting that thigh muscle fatigue leads to a higher risk of ACL injury. Their findings suggested that an increase in tibial translation is associated with a decrease in motor neuron activity related to the reflex with a resulting reduction in muscle force (Melnyk & Gollhofer, 2006).

Potential Mechanisms

One reason why the nerve needed a larger stimulation to cause a reflex could be because of a change in ion concentration in the nerve due to fatigue, causing hyperpolarization and the need for an increased stimulus to reach the action potential threshold. Because the H-reflex is analogous to the stretch reflex, but bypasses the muscle spindle, it cannot be inferred from this study that there is a change in the muscle spindles ability to detect stretch in the muscle. However, due to the fact that the rest of the pathway elicited is the same as the stretch reflex, the results suggest that there is a change in the reflex pathway between the sensory nerve, and the muscle response. Reasons for this difference could also include changes in the sensory nerve, the synapse, and/or the motor neuron. In general we support our hypothesis that there would be a change in the H-reflex when comparing pre and post fatigue EMG results. However no significant differences were seen in H-reflex peak, duration of the reflex, or the start and end time of the H-reflex.

Gender Differences

Although this study did not compare the differences between males and females, due to the high incidence of injury within the female population this study focused on trained female athletes. However, other studies have compared males and females to determine why females are at such high risk. In a fatigue study, Moore et al. (2002) found that females took significantly longer to produce knee extension force in response to a patellar tendon tap following a bout of fatiguing isokinetic exercise, while males did not change. Changes in force delays appeared to be short-lived, returning to near pre-fatigue levels within 2 and 4 min post cessation of exercise. This change in the H-reflex during recovery supports the results found on H-reflex recovery by Patikas et al. (2006). Although these studies focused on different muscles, the hamstrings and the soleus, there seems to be a pattern in association with the H-reflex and it’s reaction to fatigue, regardless of which muscle is being used. An equally important finding by Moore et al. was that the reflex response magnitude was somewhat diminished in females, yet significantly augmented in males following fatigue. Moore's findings alone are clinically relevant in that it suggests that males appear to have a greater capacity to compensate for contraction force failure and delays than females under similar levels of fatiguing exercise (Moore, Drouina, Gansnedera, & Shultz, 2002). The findings of Moore's study support the current study in the understanding of one reason as to why females tear their ACL more frequently than males. The current study shows that there is a significant difference in the onset of H-reflex, post fatigue, and that this may have implications towards stability of the knee and therefore susceptibility to ACL injuries. Along with other accepted differences between males and females, such as Q angle, that may be worsened when active muscle action is decreased due to fatigue, there seems to be an explanation here as to how females may be more at risk.

Another gender difference that was not addressed in this study is muscle fiber type and how the each type may react to fatigue better than the other. Moore et al. (2002) suggested that if females had a greater distribution of slow twitch fibers compared to males, it is possible their fast twitch fibers may have fatigued more quickly in a maximal effort fatigue protocol such as ours. Moore et al. also suggests that reflexes are predominated by slow twitch fibers, greater reliance on fatigue resistant slow twitch to maintain effort as fatigue progressed may have had a greater impact on post fatigue reflex responses in females (Moore, Drouina, Gansnedera, & Shultz, 2002). These may be attributed to fatigue resistance and should be studied further when comparing males to females.

Implications of the findings and future research

Due to the H-reflex stimulating the afferent nerve after the muscle spindle, a future study could focus on the tendon tap as a means to produce muscle contraction rather than the H-reflex protocol. This as a protocol would find the maximum reflex but indicate to us how the muscle spindle responds to fatigue. There are also a large number of factors that account for non-contact ACL injuries; therefore there are many different future directions that this project could lead into. This project focused on the Hoffman's reflex in Division I Soccer Players and how fatigue can affect the reflexes that control the stabilizing muscles of the joint. These findings could help to explain why a majority of non-contact injuries occur late in games and/or seasons. However, females sustain a disproportionally high number of ACL injuries compared to males; therefore future studies should research into why this is the case. Related to this study, more research should be focused on whether the reflexes in males respond to fatigue better than females. If this difference is presented, injury prevention techniques could be focused in on improving the nerves' response to fatigue in order to decrease the risk of noncontact ACL tears in females. Furthermore, this study focused on highly trained athletes who encounter movements that put the ACL at risk on a daily basis, such as medial translation of the knee, when they are performing cutting, landing, accelerating, and decelerating movements. Due to this study focusing on such a specific group of participants, trained females, future research should focus on the difference between trained and untrained athletes, as well as males and females to be able to compare differences in results. Any and all research should be encouraged to help introduce training regiments into sports where this particular type of injury is prevalent, such as soccer and basketball. Such regiments would help to prevent the physical, emotional and mental damage caused by such a traumatic lower extremity injury.

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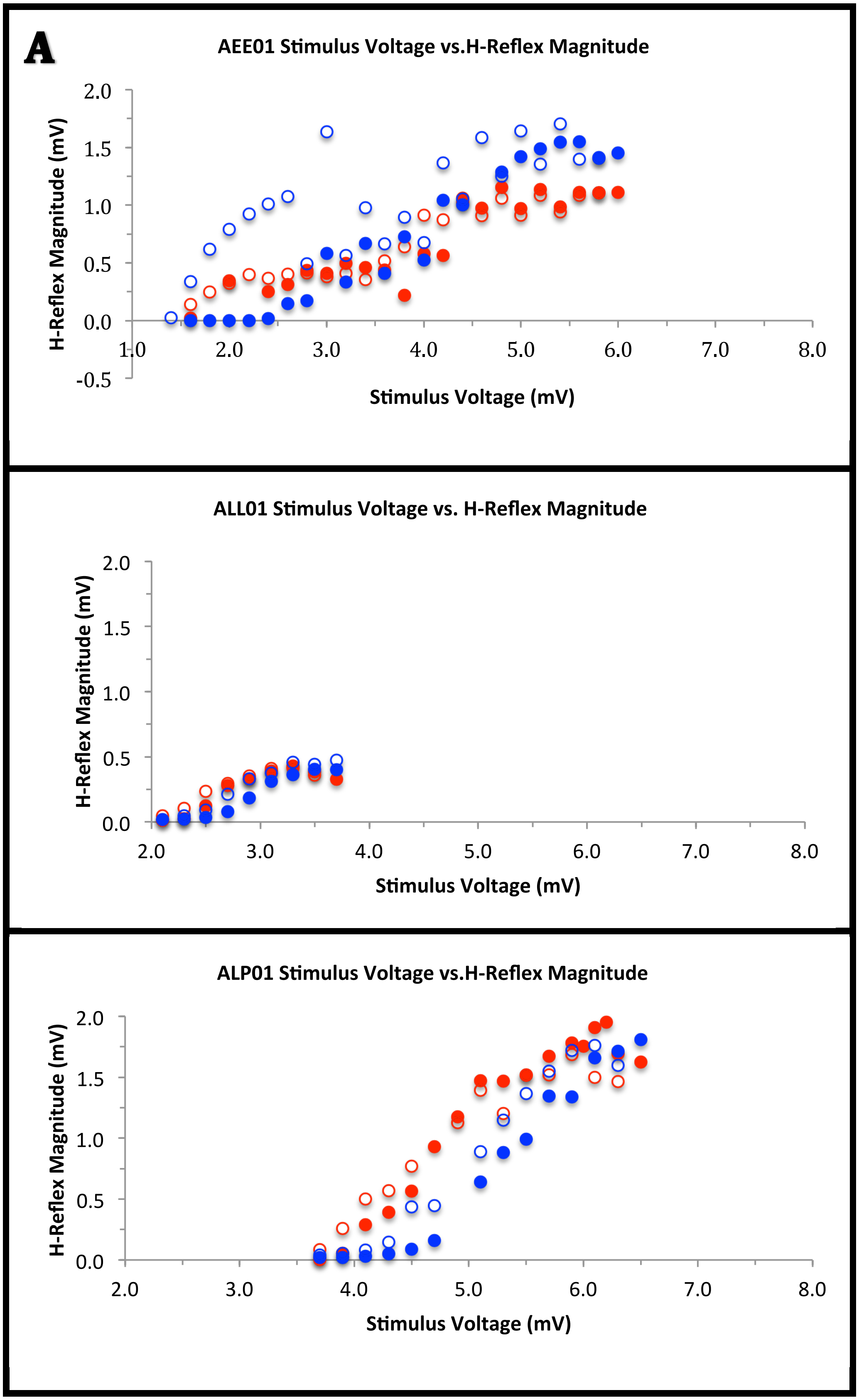
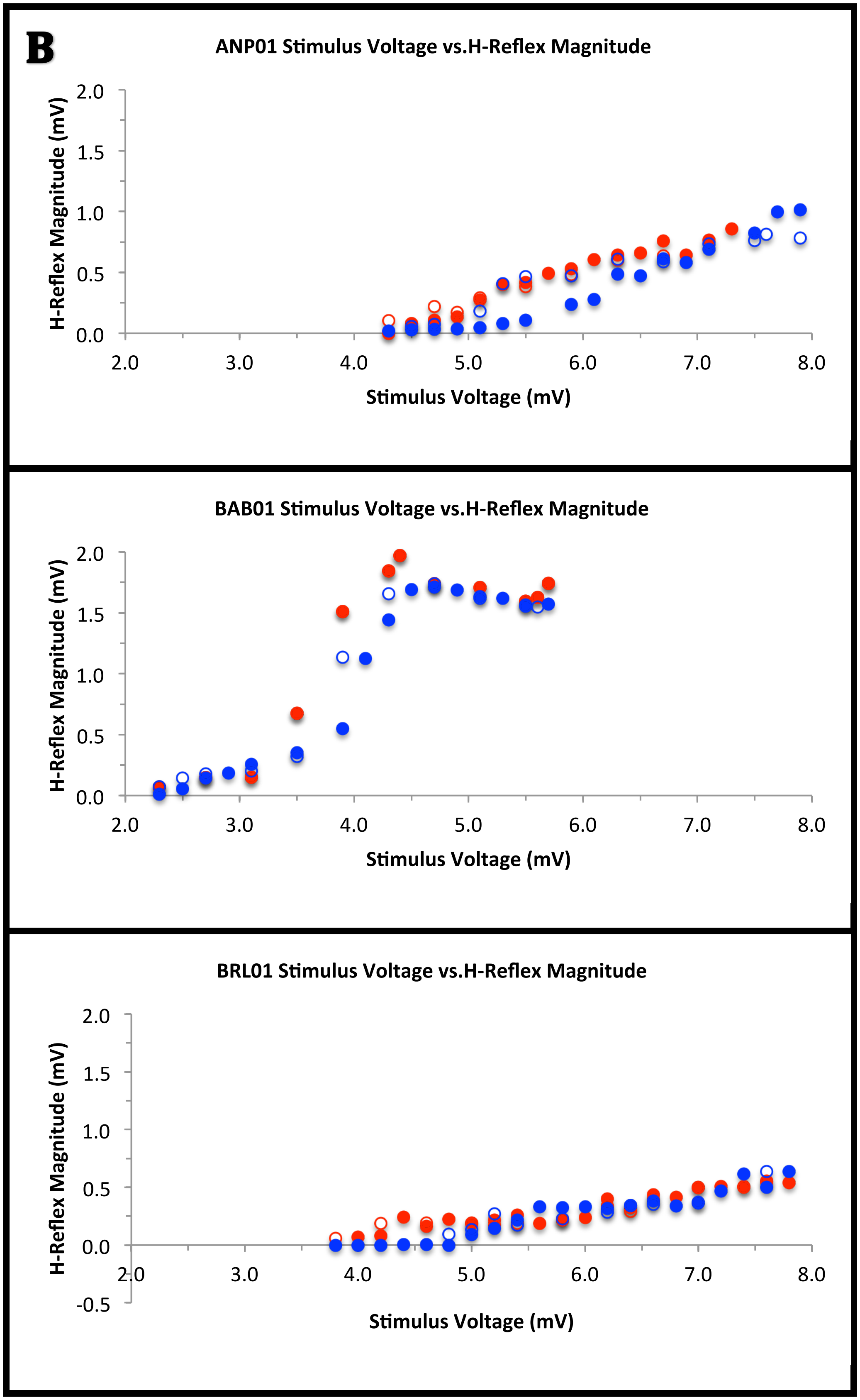
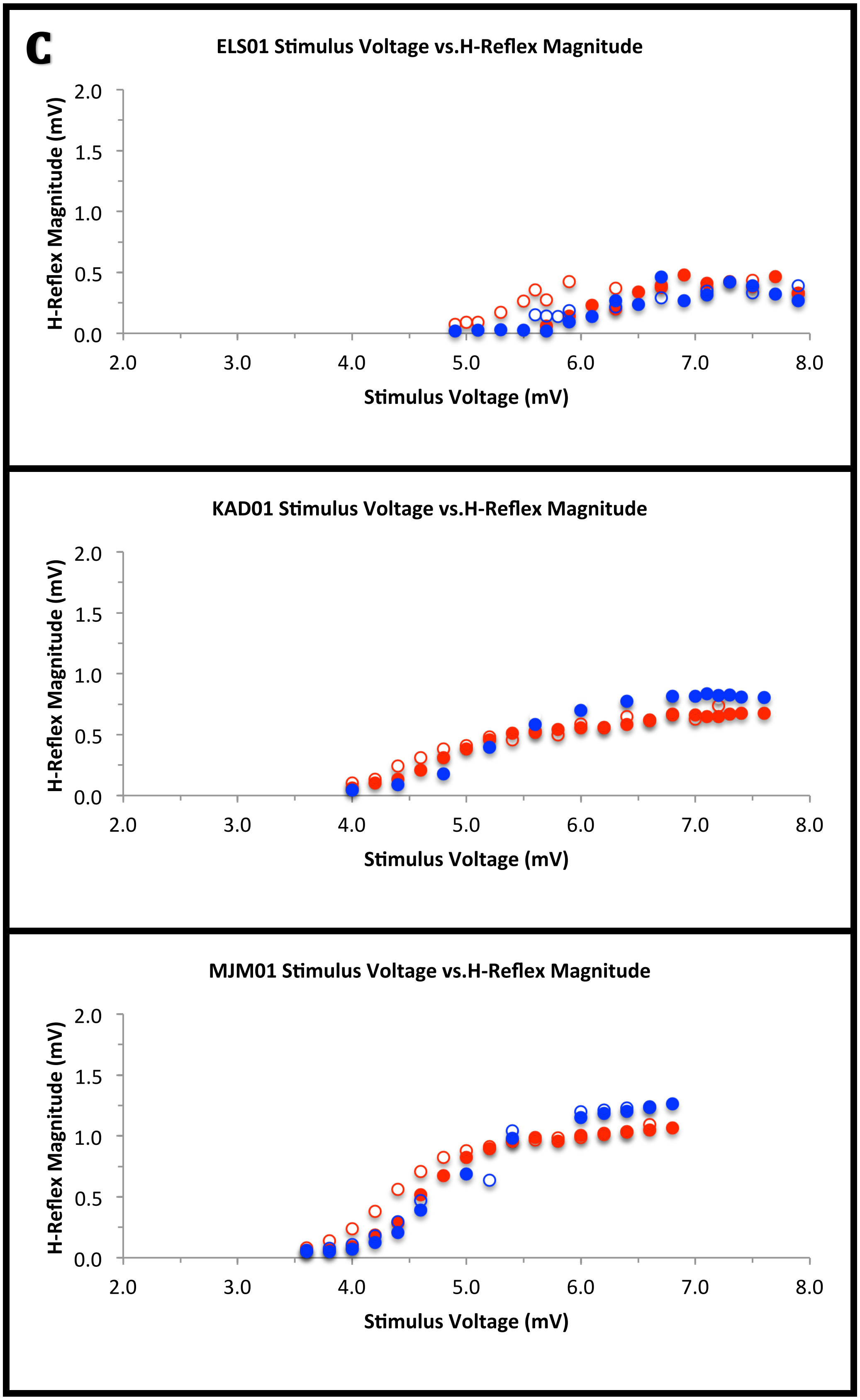
Appendix A

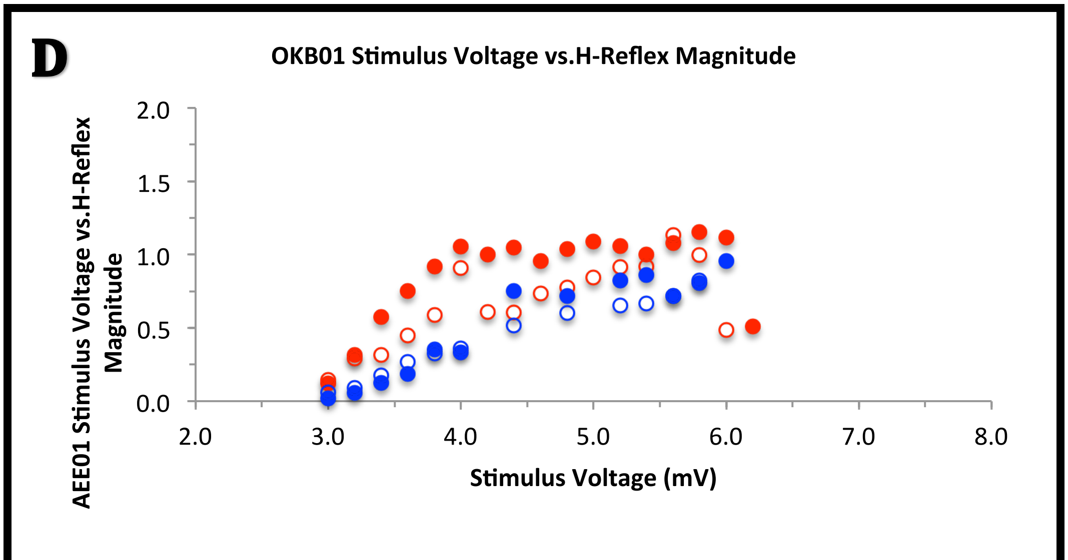
Supplementary figures: Figure 5, A through D. Figure 6, A through D.

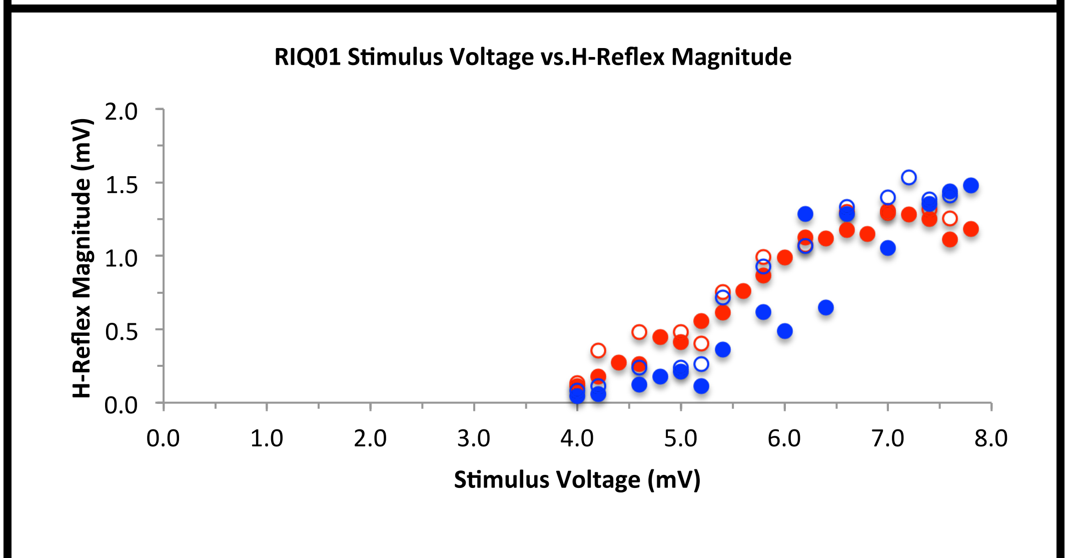

A
AEE01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
-0.5
1.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
ALL01 Stimulus Voltage vs. H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
ALP01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)


B
ANP01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
BAB01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
BRL01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
-0.5
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)



C
ELS01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
KAD01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)
MJM01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
Stimulus Voltage (mV)


D
OKB01 Stimulus Voltage vs.H-Reflex Magnitude
AEE01 Stimulus Voltage vs.H-Reflex Magnitude
Stimulus Voltage (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0


RIQ01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
Stimulus Voltage (mV)
2.0
1.5
1.0
0.5
0.0
0.0
1.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0

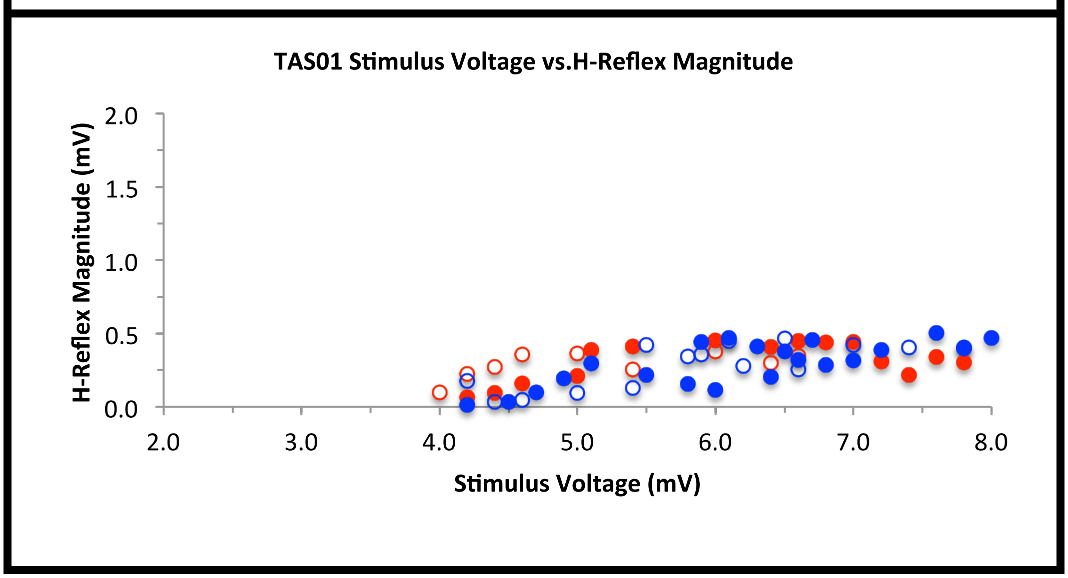
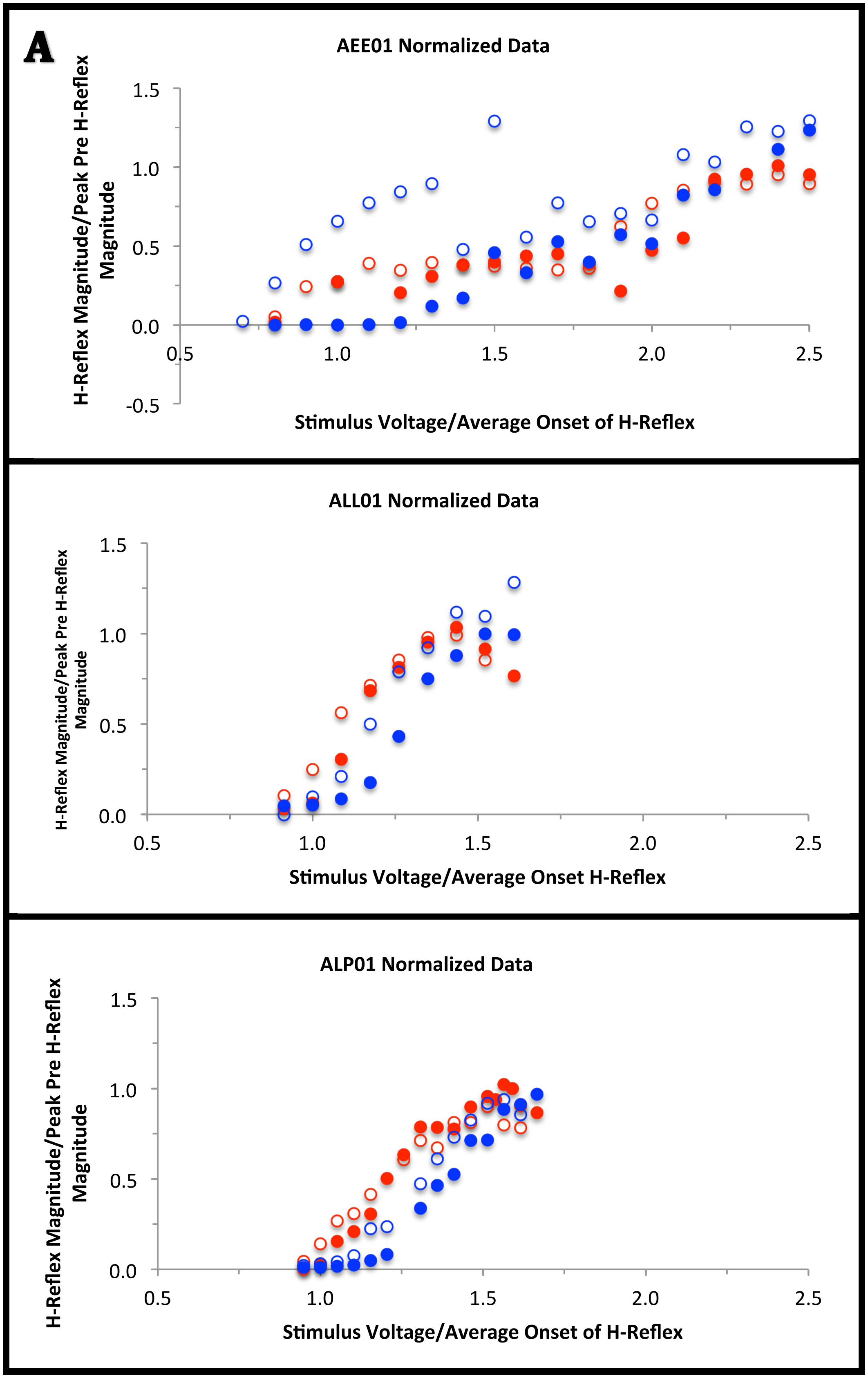
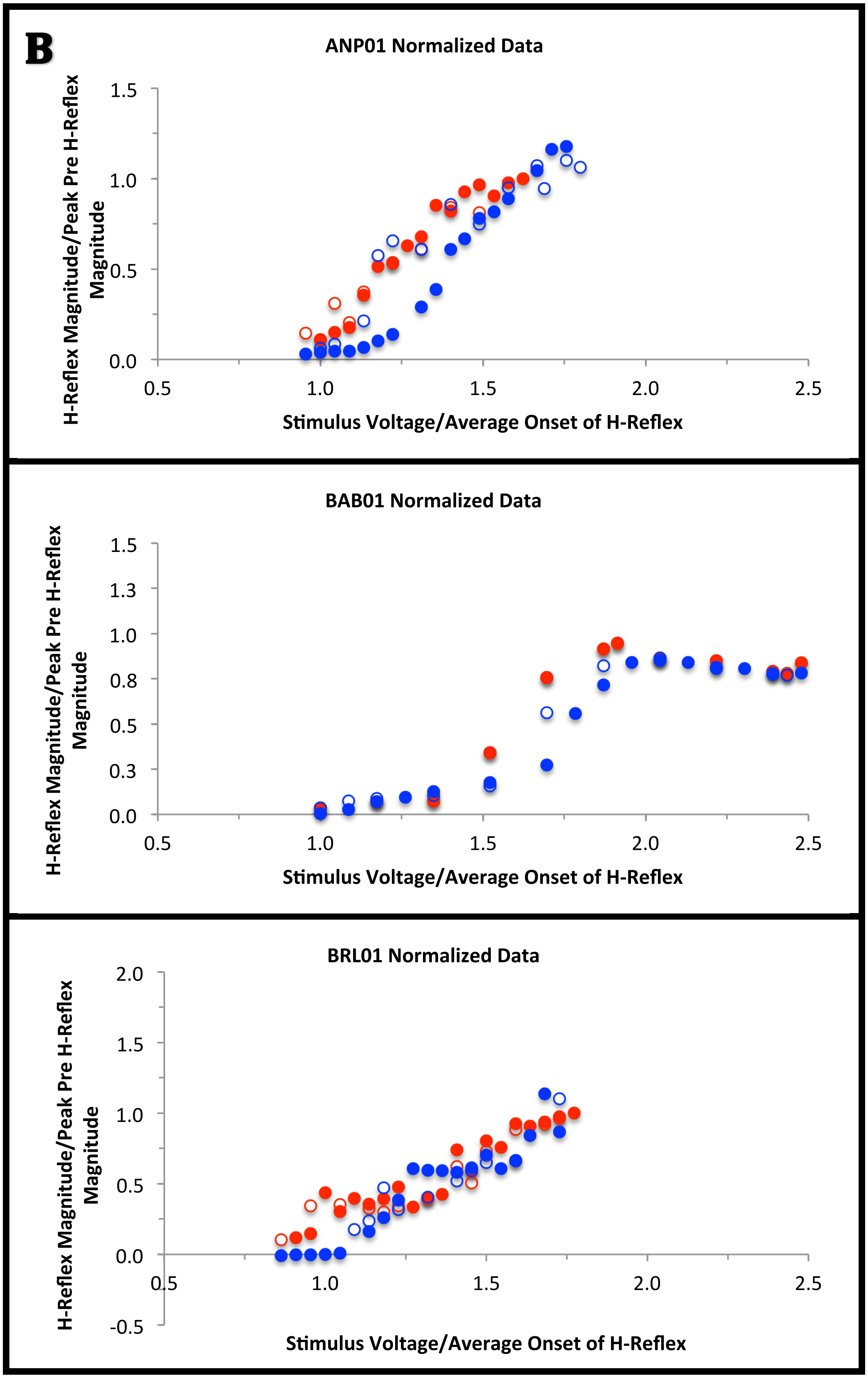
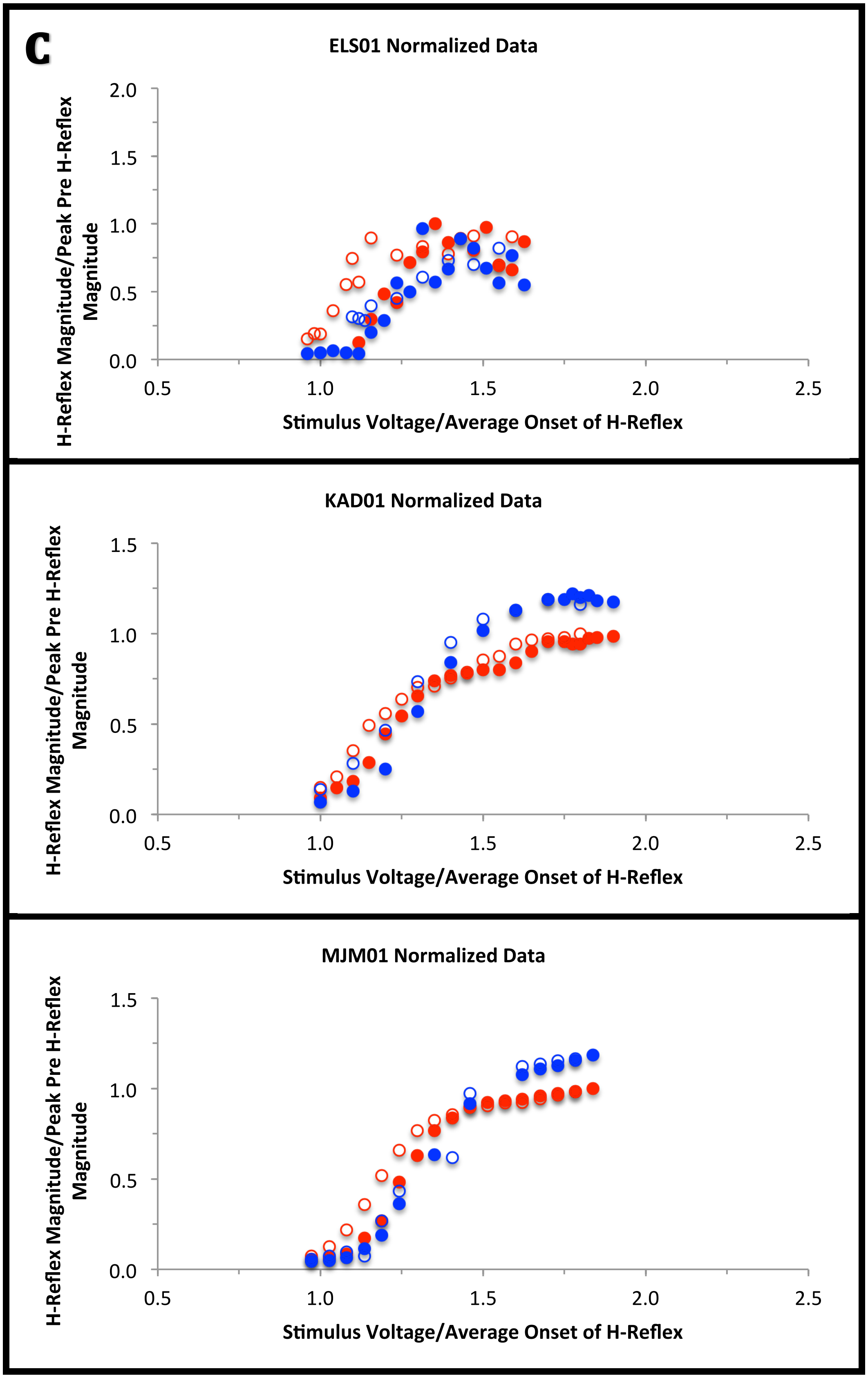

TAS01 Stimulus Voltage vs.H-Reflex Magnitude
H-Reflex Magnitude (mV)
Stimulus Voltage (mV)
2.0
1.5
1.0
0.5
0.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0

Figure 5: Sections A through D Showing the raw data, Stimulus Voltage vs. H-reflex magnitude, for each participant. The points represent the averages between the two trials. Filled in circles represent data collected going up to the max, circles with no fill represent data collected coming back down from the maximum. The red circles represent pre fatigue and blue represents post fatigue.


A
AEE01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
1.5
1.0
0.5
0.0
-0.5
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex
ALL01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
1.5
1.0
0.5
0.0
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset H-Reflex
ALP01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
1.5
1.0
0.5
0.0
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex


B
ANP01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
1.5
1.0
0.5
0.0
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex
BAB01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
1.5
1.3
1.0
0.8
0.5
0.3
0.0
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex
BRL01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
2.0
1.5
1.0
0.5
0.0
-0.5
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex


C
ELS01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
Stimulus Voltage/Average Onset of H-Reflex
0.0
0.5
1.0
1.5
2.0
2.5
KAD01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
Stimulus Voltage/Average Onset of H-Reflex
MJM01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
Stimulus Voltage/Average Onset of H-Reflex

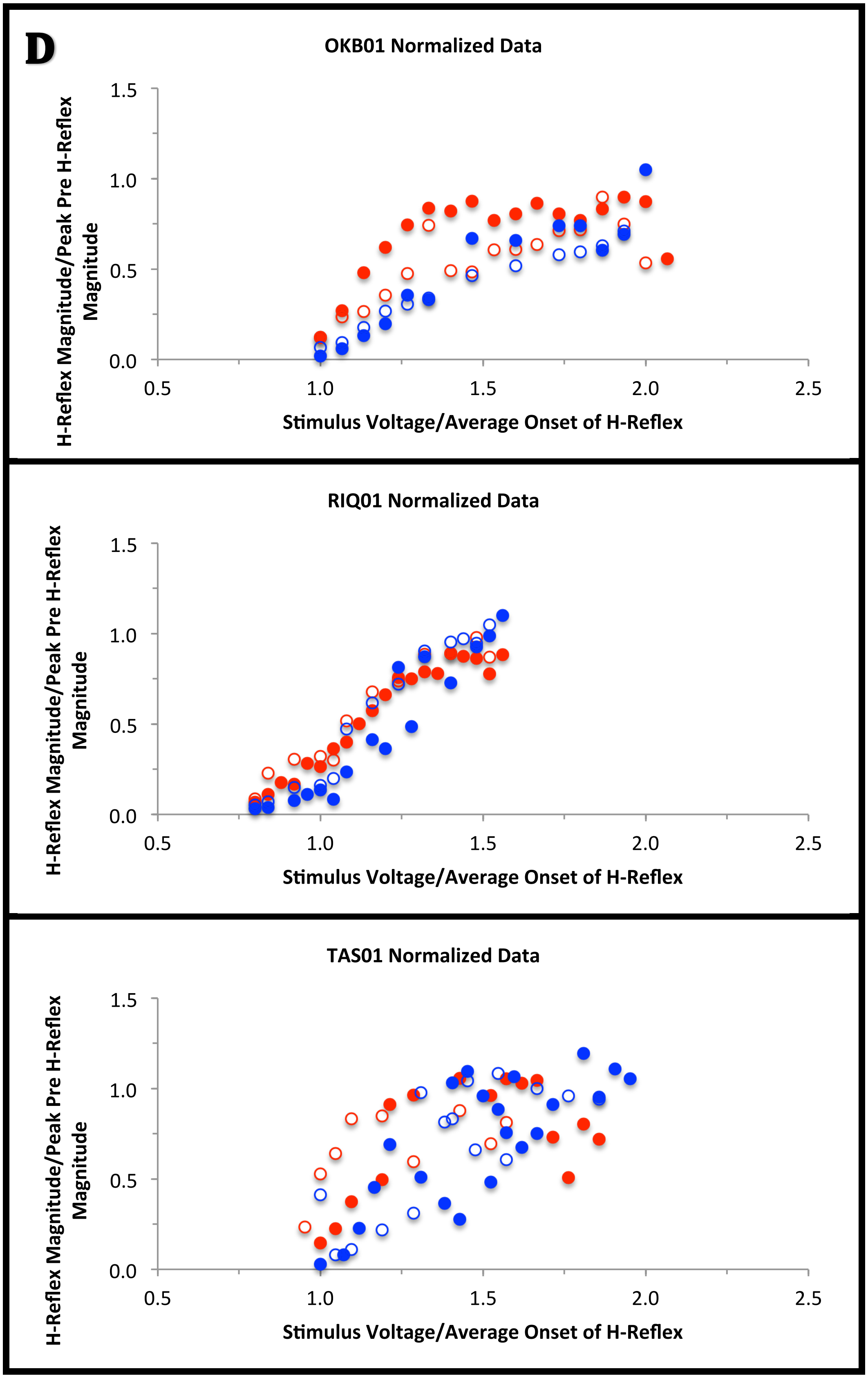

D
OKB01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
0.0
0.5
1.0
1.5
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex
RIQ01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
0.0
0.5
1.0
1.5
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex
TAS01 Normalized Data
H-Reflex Magnitude/Peak Pre H-Reflex Magnitude
0.0
0.5
1.0
1.5
0.5
1.0
1.5
2.0
2.5
Stimulus Voltage/Average Onset of H-Reflex

Figure 6: Sections A through D showing normalized data, stimulus voltage/average onset of H-reflex vs. H-reflex magnitude/peak pre H-reflex magnitude, for each participant. The points represent the averages between the two trials. Filled in circles represent data collected going up to the max, circles with no fill represent data collected coming back down from the maximum. The red circles represent pre fatigue and blue represents post fatigue.

Appendix B

IRB Form

Form 2: Non-Exempt Application Materials

University of Idaho procedures require that the Institutional Review Board (IRB) review and approve of projects involving humans. Official approval from the IRB must be given **before** the research can begin.

Forms should be emailed as attachments to irb@uidaho.edu in **Microsoft Word format**.

If you are a student, you should be listed as the student investigator and your faculty sponsor as the PI. You must submit your materials to your UI faculty sponsor/PI. After their review and approval, they will FORWARD your materials to the IRB for review.

If you are not a full-time faculty member or employee at the UI, you must contact a departmental faculty member, administrator or department chair. This person will become your faculty sponsor.

Once you have submitted the completed application, the Institutional Review Board will approve it. **You can begin the research ONLY AFTER receiving WRITTEN approval from the committee.**

Please allow at least **six weeks excluding holidays** for the initial review and approval process. [Note: The approval process takes longer when corrections are requested by committee members or when we have a large number of applications].

Note: All researchers participating in human subject's research are required to take the online course through the National Institutes of Health http://phrp.nihtraining.com/users/login.php

Copies of certificates of completion will be required before projects will be approved.

Please include your UI campus mail code address (83844 - _ _ _ _) on the summary form inside, and an **address** below.

Laura Jackson_______________________________________

Department of Neuroscience_________________________

875 Perimeter Drive MS 2401_____________________________

Moscow, ID 83844-2401__________________________________

Investigator e-mail: ljackson@uidaho.edu___________

Faculty Sponsor e-mail if applicable cpmcgowan@uidaho.edu_______

Form 2: University of Idaho Human Subject Review – Non-exempt Projects

This project qualifies for "Non-Exempt" status. Please complete the following application. In addition, the following information must be included:

1. An electronic copy of certification in PDF or Microsoft Word format that the online course sponsored by
   the National Institutes of Health has been completed by everyone listed on the project.

   NIH website: http://phrp.nihtraining.com/users/login.php

2. If applicable, an electronic copy of an Informed Consent Form that includes all components provided at: http://www.uidaho.edu/ora/committees/irb/irbforms

3. If applicable, a copy of the survey, questions intended to be asked, or if conducting qualitative research,
initial entry questions and items where the investigator might probe for additional information.

Principal Investigator: Craig McGowan______________________________ Academic Title: Ph.D_____________

Student Investigator: Laura Jackson____

Department/Division: Biological Sciences___________________ Campus Zip Code: 83844-3051____Phone: 208.885.6598_

Project Title: The Effects of Functional Fatigue on H-Reflex in Female Soccer Players to Predict ACL Injury

Proposal Number:
____

Previous IRB protocol Number: _______________________________

Anticipated Start Date: _July 2014______________________________

Anticipated End Date _May 2015_______________________________

Faculty Sponsor (if you are not principal investigator) Craig McGowan____

Is the project seeking funds? (Answer using a bold **"X"**) YES ____ NO _**X**__

If yes,

Granting Agency: ____

Grant Title: ____

Principal Investigator on Grant:
____

If a continuation, date of previous approval:
____

I. SUBJECTS/PARTICIPANTS

A. Approximate number: 25 Females

B. Age Range: 18-23__________________ (Note: Participants less than age 18 have additional requirements)

C. How will participants be selected or recruited?

Participants will be recruited from the University of Idaho NCAA Division 1 Women's Soccer team.
The athletes will be asked to volunteer to attend an information session with a brief description of our research study and IRB approval. If they choose to volunteer they will then attend a session whereby the participant will be shown what the procedure entails, practically, and consent forms signed if they wish to continue.

Example of flyer attached as appendix B

D. Are there participants who will be excluded? Why?

Criteria for exclusion will include: current musculoskeletal injury of lower extremities or back, lingering symptoms of musculoskeletal injury in lower extremities or back, current musculoskeletal pain in the lower extremities or back, chest pain with exertion, shortness of breath, known hypertension, history of heart attack, or pregnancy. These subjects will be excluded because of the potential for increased risk of cardiac problems or musculoskeletal injury during the testing procedure. All subjects will be examined by a Certified Athletic Trainer through the University of Idaho Athletics program. Due to NCAA division 1 rules for student athletes a physical checkup is required in July, therefore any participants disqualified for soccer activity due to lower extremity/back pain or dysfunction will be disqualified for the study also.

The subject screening form is attached as appendix C

E. Will participants be paid? If yes, how much, when, and how? Must they complete the project to be paid?

Participants will not be compensated for participating in this study.

F. Are any of the participants not competent to give consent (e.g., minors, prisoners, institutionalized)? If yes, how will consent be obtained? From whom? Are there procedures for gaining assent (if appropriate)?

No, the subjects will in no case be minors, prisoners or otherwise members of a vulnerable population.

If appropriate, how will "assent" be obtained? (Participants themselves, even though deemed not competent, must agree to the research.)

Not Applicable

G. Will this study be conducted in an Educational (School / Pre K - 12) setting and involve children or teachers actively teaching within the classroom as part of the study? If yes, **ATTACH** documentation from a Teacher and School Principal, Superintendent, or other administrator indicating approval. Also, **ATTACH** appropriate material regarding FERPA regulations (if applicable).

No, an educational setting will be needed for this study.

II. DESCRIPTION OF PROJECT. Type answers in the spaces provided. Although you may cut and paste materials from other documents, **Do Not** refer to attached grants, papers, dissertation proposals, etc. Be clear, brief and specific. **The IRB application must stand on its own.**

A. Describe the Purpose of the Research.

The purpose of this research is to investigate the effect of functional fatigue on the H-Reflex in female division 1 soccer players. Prior to muscular fatigue the H-Reflex of each participant will be measured. Through using the BioPac system, a skin electrode will be used to elicit the femoral nerve's H reflex. At least three measures for the H reflex will be recorded in order to calculate the mean during analysis. Further, electromyography (EMG) electrodes will be used to monitor muscle excitability in the vastus medialis muscle. Muscular fatigue in this muscle will be achieved through one bout of a Wingate test, which includes pedaling as hard as possible on a stationary bike against a resistance of 8.6% of body weight for 20 seconds. To calibrate these measurements, we will collect simple measurements of body weight for the Wingate Bike Test. Muscle activity will again be recorded through the EMG technique while the stimulating electrode will be again used to elicit the femoral nerve H-Reflex. The voltage at which the first H reflex was found will again be tested post fatigue to compare to pre Wingate results. Furthermore the voltage will be increased in order to try and find the new H reflex, repeated a minimum of three times to find a mean for analysis. Due to the hypothesis, it is predicted that in order to find the H reflex post fatigue the intensity of the stimulating electrode must be increased. All values of the H-Reflex, once found, will be repeated at least three times for statistical analysis of the mean.

If recovery of the H-Reflex is seen, back to baseline levels, the H reflex will be retested and post Wingate results retested at the discretion of the participant.

B. Describe the Research Design (Survey, Naturalistic Observation. 2 by 3 Factorial Design, Qualitative Design, etc).

The experimental design will involve repeated measures of our dependent variables prior to and following one bout of the 20 second Wingate tests. Data will be analyzed using a mixed factor repeated measures ANOVA to quantify the effects of fatigue on our dependent variables.

C. Describe the Procedures (What will the Participants do).

- In July 2014, prior to meeting in the first session, all candidates would have already been seen and cleared by a certified athletic trainer for full activity within the sport of Division 1 NCAA soccer due to requirements set by the organization.
- During the recruitment session participants that attend will have the procedure explained and if willing to volunteer will then schedule a meeting time in the lab (session 1). Further study purposes and practical procedures will be described to the participants. Then the informed consent form will be given to the participant to ensure understanding and aid in compliance. A further certified athletic trainer will conduct an FMS (functional movement screen) screen on the participant which is a ranking and grading system that documents movement patterns that are key to normal function. By screening these patterns, the FMS will identify functional limitations and asymmetries. These are issues that can reduce the effects of functional training and physical conditioning and distort body awareness.

Furthermore in session 1 will take part in lab protocol familiarization. Participants will be instructed in the completion of a Wingate test protocol. The participants will also have their H-Reflex elicited as an idea of electrode placement for the testing day. Participants will complete the Wingate and nerve stimulation until they are comfortable with the protocol.

Participants will be invited back for data collection within 24 hours to 1 week from the protocol familiarization meeting.

Session 2 will consist of the experimental paradigm. EMG electrodes will be placed on the skin of the legs over the specific muscles to be analyzed, which will allow for muscle activity to be measured. Furthermore 2 stimulating electrodes will be placed over the femoral nerve and the Gluteus Maximus in order to stimulate the femoral nerve. A ground electrode will also be placed just superior to the patella. Each participant will have their H-Reflex tested to find the maximum and steady readings for the pre fatiguing section of the experimental data, repeated at least three times for mean statistical analysis. They will then take part in one bout of the Wingate Bike test. Cycling bouts will include 20 seconds of stationary cycling at maximal effort with a resistance calculated at 8.6% of a subject's body weight. Immediately after every exercise bout, the participant will be asked to have their H-Reflex retested with the electrode at the exact same location as before the test (electrodes will be kept on during the cycling bout and also the area will be marked in case it falls off due to the physical activity).

If recovery of the H-Reflex is seen, back to baseline levels, the H reflex will be retested and post Wingate results retested at the discretion of the participant.

The certified athletic trainer that conducts the full body screening prior to day 1 will document no information, the only information that will be given is whether the participant should be included or excluded from the study. Listed below is the exclusion criteria for the study:

do not sign informed consent form,

do not meet age criteria,

have contraindications for maximal intensity exercise (based on ACSM guidelines),

have a history of back injury,

or if they have had lower extremity injury or surgery 6 months prior to data collection.

All participants will be screened by a Certified Athletic Trainer and will be excluded from this study if they have any lumbar spine or lower extremity pain or dysfunction.

The informed consent form is attached in appendix A
The Screening Form is attached in appendix C

D. If any deception (withholding of complete information) is required for the validity of this activity, explain why this is necessary and **attach** a debriefing statement.

No information will be withheld at any point in the experiment.

III. ASSESSMENT OF RISKS AND BENEFITS.

A. Describe the nature of any potential risks. These include stress, social, legal, discomfort, invasion of privacy, embarrassment, or side effects.

Drop landing from a 60-cm height, nerve excitability, cut turns or exercising on a bicycle can cause fatigue, minor discomfort, and potentially musculoskeletal injury. In extreme instances exercise can cause irregular heartbeat during exercise and in very rare instances the risk of heart attack or death can be somewhat increased during exercise.

B. Describe how each of the risks in part A will be minimized. Be detailed and complete.

The cycling and landings used in this study do not present any greater risk than playing sports in the community or at the Rec Center. By selecting subjects that participate daily in division 1 soccer and physical activity we will greatly decrease the possibility of fatigue, discomfort and musculoskeletal injury.

Cycling bouts of maximal effort will be short duration lasting 20 seconds. This will ensure safety to protect against overexertion during each cycling bout. It is unlikely that participants would voluntarily overexert themselves to the extent that would cause any physical harm especially with the short duration of the event. Participants with any history of cardiac or musculoskeletal problems will be excluded from participation in the study.

With every participant screened by a Certified Athletic Trainer, musculoskeletal pain or dysfunction will be identified and those compromised will be excluded from the study to ensure that no injuries are acquired through the demands of the procedures. The certified athletic trainer will document no information, the only information that will be given is with regard to whether the participant should be included or excluded from the study.

In regard to the rarely increased incidence of irregular heartbeat, heart attack or death, the level of exercise that will be used in this study is recommended and safe for most people. Specifically, the *American College of Sports Medicine Guidelines for Exercise Testing and Prescription* states that medical evaluation prior to and monitoring during a moderate intensity exercise program is not necessary for the apparently healthy individuals who are less than 40 years old and have no coronary risk factors[1]. By selecting for healthy subjects who regularly participate in physical activity this risk is even further decreased.

1. Mahler DH. *American College of Sports Medicine Guidelines for Exercise Testing and Prescription*. 5th ed. Baltimore, Md: Williams & Wilkins; 1995:373.

C. In the event that any of these potential risks occur, how will they be handled (e.g., compensation, counseling, etc.)?

In the event that mild injury occurs during the trials, on-site first aid will be provided at no charge. In the unlikely event that any more serious injury occurs, the participant will be responsible for seeking and paying medical expenses.

D. Will this study interfere with any subject's normal routine (e.g., school attendance, medical treatment, etc.)?

Participation in this study will require a maximum of 6 hours of each subject's time, including the recruiting meeting, session 1 and session 2. Scheduling will be very flexible, which will ensure that there will not be any significant interference with any of the participant's normal routine.
Participation will also be required in the summer when the normal routine of the athlete will be decreased in volume.

E. Describe the expected benefits to society and to the individual subjects.

The expected benefits to society include increased understanding of how repeated bouts of fatigue affect the body's ability to perform physical tasks and how the nervous system reflex is affected. Many ACL injuries occur in the late stages of soccer games when the body is in a fatigued state and to date the effects of fatigue and its contribution to injury are poorly understood. This research study would be unique in that few studies measure the effects of fatigue on the H-Reflex, especially in the femoral nerve.

There are no expected benefits to the individual participant.

F. Will blood be taken? (Answer using a Bold **"X")** YES____ NO__**X**__

Who will take the blood? ____

How often? ______________________________ How much? ________________________________

Describe the procedure for drawing the blood:

IV. CONFIDENTIALITY OF DATA

Using a bold "X" answer the following questions

A. Will data be anonymous (i.e., even the researcher will not be able to link the identity of the subjects/participants with responses)?

YES _____ (Go to Part C)

NO __**X**___ (If NO, complete item IV-B.

B. Will data be confidential? YES__**X**___NO_____

If **<u>YES</u>,**

Will the data be coded to a master list? YES__**X**__ NO____

Will the list be kept separate from the data? YES__**X**__ NO____

If **<u>NO</u>,**

Who else will have access to the data?

____

Why? ____

How will confidentiality be maximized?

____

C. How will the data will be stored? Locked laboratory_**X**__ Locked file cabinet ___

Restricted Computer_**X**___ Other (describe):

D. How will the data eventually be deleted? If not deleted, how will linkage to identities be broken?

Once the study has been published, the coded master list with participant names and participant number will be deleted from the laboratory computer, so that the data will only be identifiable by participant number thereafter.

V. ADDITIONAL IMPORTANT CONSIDERATIONS

Using a bold "X" answer the following questions

A. Will any investigational NEW drug (IND) be used? YES_____ NO__**X**___

B. Will any other drugs be used? YES_____ NO__**X**___

If **<u>YES</u>** to A or B, list for each drug:

1) the name of the drug;

2) the source of the drug;

3) the dosage;

4) any side effects or toxicity;

5) how it will be administered; and

6) by whom it will be administered.

ATTACH PDR OR EQUIVALENT MATERIAL IN AN APPENDIX TO THIS PROPOSAL

C. Will a new investigative device (IDE) be used? YES_____ NO___**X**__

IF **YES**, has the Idaho Research Foundation been notified? YES_____ NO_____

D. Will ethyl alcohol be ingested by the participants ? YES_____ NO__**X**___

If **YES**, fill out the Alcohol Human Subjects Form found on the IRB website

Refer to the guidelines for administration of ethyl alcohol in human experimentation available from the UI Research Office.

E. Will audio-visual tapes, audiotapes or photographs be taken? YES__**X**___ NO_____

If **YES**:

Where will the tapes be stored?

Digital video files from the data capture will be stored in the hard drive of the computer located within the locked laboratory. These files will be password-protected. Only the electrode placement will be photographed so that subject faces will not be visible. The digital videos will be used for referencing of electrode placement for possible future publications.

When will this material be destroyed?

This material will be destroyed once the research study has been published in a peer-reviewed journal.

F. Will a written consent form be obtained? YES__**X**__ NO_____

If **<u>YES</u>**: please attach consent form (refer to the Components of a Consent Form included in packet).

If **<u>NO</u>**: how will consent be obtained?

Why is this method being used?

VI. INTERNATIONAL PROJECTS

Using a bold "X" answer the following question

A. Will the project be conducted outside the United States YES _____ NO __**X**_____

If **<u>YES</u>**: Has an IRB been contacted in the country where the study will be conducted? YES ____ NO ____

If yes, provide documentation indicating approval.

If no, provide an explanation why an IRB has not been contacted and/or explain how you will comply with the Belmont Report, Declaration of Helsinki or similar document.

VII: OTHER AGENCIES

A. Some projects require additional approvals beyond IRB/IRB approval (e.g., Office of Management and Budget for surveys in federal parks, Native American Tribal Councils, U.S. Food and Drug Administration, etc). List additional agencies where project approval has been obtained. Attach appropriate documentation. If materials are under review at these agencies indicate the review is in progress.

1. ____
2. ____
3. ____

VIII: Sponsored Programs

If this project seeking funding YES ________ NO _____**X**______

Has Sponsored Programs been notified? YES ________ NO ___________

IX: ONLINE COURSE COMPLETION

List the names of all investigators and indicate the date(s) of completion for all investigators taking the

Protection of Human Subjects from the National Institutes of Health on line class.

http://cme.cancer.gov/clinicaltrials/learning/humanparticipant-protections.asp

FACULTY SPONSOR NOTE: A copy of the completion certificate or other verification must be included for ALL investigators including laboratory assistants, observation observers, etc.

Name of Investigator	Date of Course Completion	Certificate Number of Online Course
Laura Jackson	01/28/13	1086553
Craig McGowan	04/05/2011	630792
Jeff Seegmiller	11/06/07	Not provided
Katrina Taylor	11/11/2010	565714

Certificate of Completion

The National Institutes of Health (NIH) Office of Extramural Research certifies that **Laura Jackson** successfully completed the NIH Web-based training course "Protecting Human Research Participants".

Date of completion: 01/28/2013

Certification Number: 1096553

Certificate of Completion

The National Institutes of Health (NIH) Office of Extramural Research certifies that **Katrina Taylor** successfully completed the NIH Web-based training course "Protecting Human Research Participants".

Date of completion: 11/11/2010

Certification Number: 565714

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Human Participant Protections Education for Research Teams

Completion Certificate

This is to certify that

Jeff Seegmiller

has completed the **Human Participants Protection Education for Research Teams** online course, sponsored by the National Institutes of Health (NIH), on 11/06/2007.

This course included the following:

- key historical events and current issues that impact guidelines and legislation on human participant protection in research.
- ethical principles and guidelines that should assist in resolving the ethical issues inherent in the conduct of research with human participants.
- the use of key ethical principles and federal regulations to protect human participants at various stages in the research process.
- a description of guidelines for the protection of special populations in research.
- a definition of informed consent and components necessary for a valid consent.
- a description of the role of the IRB in the research process.
- the roles, responsibilities, and interactions of federal agencies, institutions, and researchers in conducting research with human participants.

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A Service of the National Cancer Institute

FIRSTGOV

Certificate of Completion

The National Institutes of Health (NIH) Office of Extramural Research certifies that **Craig McGowan** successfully completed the NIH Web-based training course "Protecting Human Research Participants".

Date of completion: 04/05/2011

Certification Number: 630792

If this project will be submitted or will receive external funding, print out the last page sign on the following signature line using a pen, provide the date of submission, and mail it to:

Institutional Review Board
University of Idaho
POB 443010
Moscow, Idaho 83844-3010

Currently, an electronic copy or electronic signature is not enough to comply with the Federal regulations/requirements for <u>funded</u> research

ADDITIONAL
INSTITUTIONAL REVIEW BOARD APPLICATION INFORMATION

1) The completed Institutional Review Board application form must be received by the Institutional Review Board (IRB) at least 6 weeks prior to the intended start date. Send the form and all attachments electronically to irb@uidaho.edu. DO NOT SEND HARD COPIES OF THE APPLICATION. THEY WILL BE RETURNED TO YOU WITHOUT REVIEW. HOWEVER, DO SEND A COPY OF THE SIGNATURE PAGE IF RESEARCH IS FUNDED.

2) For a project to obtain IRB approval, the IRB shall determine that all of the following requirements are satisfied:
   a. Risks to subjects are minimized:
      i. By using procedures which are consistent with sound research design and which do not unnecessarily expose subjects to risk, and
      ii. Whenever appropriate, by using procedures already performed on the subjects for diagnostic treatment purposes.

b. Risks to subjects are reasonable in relation to anticipated benefits, if any, to subjects and the importance of the knowledge that may be expected to result. In evaluating the risks and benefits, the IRB should consider only those risks and benefits that may result from the research (as distinguished from risks and benefits of therapies subjects would receive even in not participating in the research). The IRB should not consider possible long-range effects of applying knowledge gained in the research (for example, the possible effects of the research on public policy) as among those research risks that fall within the purview of its responsibility.

c. Selection of subjects is equitable. In making this assessment the IRB will take into account the purposes of the research and the setting in which the research will be conducted.

d. Informed consent will be sought from each prospective subject or the subject's legally authorized representative (See Components of a Consent Form).

e. Informed consent will be appropriately documented. This might include a written consent form approved by the IRB and signed by the subject), or it might be a tic box indicating the participant was verbally informed of the project.

f. Where appropriate, the research plan makes adequate provisions for monitoring the data collected to insure the safety of subjects. This may include follow-up procedures.

g. Where appropriate, there are adequate provisions to protect the privacy of subjects and to maintain the confidentiality of data.

3. Projects cannot begin **<u>until</u>** the IRB approval is obtained.

4. Once the IRB review is completed, an email will be sent to the investigator advising of the project approval or conditions that must be met to obtain approval.

GENERAL CONSIDERATION FOR THE ETHICAL TREATMENT OF HUMAN SUBJECTS

1. Subjects are entitled to dignified treatment during all phases of experimental procedures.

2. At no time are subjects to be coerced into participating in experimental procedures. Subjects may immediately terminate or withdraw from experimental procedures and earn the incentives promised them for their participation.

3. Subjects will be given sufficient information regarding the procedures to enable them in making an informed decision regarding their participation.

4. Confidentiality of subject data will be respected and preserved at all times. Experimenters will maintain control over access to subject records.

5. When appropriate, experimenters should inform subjects of the rationale of the study at some time during or following the conclusion of the procedures.

6. Experimenters should design their studies such that the costs to a subject are reasonably comparable to the rewards of participation. Any incentive promised for participation in experimental procedures will be given regardless of the quality of the subject's performance. Additional incentives may be given if they are greater in value to those that would be otherwise possible for participation.

7. Experimenters are responsible for the behavior of others (e.g., assistants, confederates, data encoders, etc.) that may influence the rights of the subjects. Assistants should be briefed by experimenters regarding the appropriate treatment of subjects.

8. No subjects will be exposed to procedures of a frivolous or clearly meaningless nature.

9. Subjects may be exposed to aversive or onerous treatments only if the potential benefits of the research to society well exceed the costs to the subject. Subjects in those procedures should be reminded of their right to terminate the procedures. Signed informed consent will be required of all subjects in such procedures.

10. The committee will retain the right to revoke its approval of, and terminate, any experiment in which accepted or defined ethical standards are not followed.

11. All non-exempt projects will have ongoing review.

Project Signature Page
(For Funded Research)

Title of Project____

____

____

Principal Investigator: The information provided above is accurate and the project will be conducted in accordance with applicable Federal, State, and University of Idaho regulations.

Signature: ____ Date: _______________

Institutional Review Board: This project has been properly filed as required by Federal, State and University of Idaho procedures.

Signature: ____ Date: _______________

Currently, an electronic copy or electronic signature is not enough to comply with the Federal regulations/requirements for <u>funded</u> research. For Funded Research, print this page, acquire all needed signatures and mail to:

IRB

Morrill Hall 209

PO Box 443010

Moscow, ID 83844-3010

Or Mail drop 3010

Received by ____

Date received ____

APPENDIX A:

Informed Consent Form

Consent Form

The University of Idaho Institutional Review Board has approved this project.

Title of Research:

The Effects of Repeated Bouts of Fatigue on Kinetics and Kinematics during Drop Landings in Recreational Athletes

IRB PROTOCOL NUMBER:_________________________

Researcher: Laura Jackson

Faculty Sponsor: Dr. Craig McGowan

The purpose of this study:

The purpose of this research is to investigate the effect of functional fatigue on the H-Reflex in NCAA Division I female soccer players.

Through bouts of fatigue it is hoped that the affect the body's ability to perform physical tasks and how the nervous system reflex is affected will be seen. Many ACL injuries occur in the late stages of soccer games when the body is in a fatigued state and to date the effects of fatigue and its contribution to injury are poorly understood. This research study is unique in that few studies measure the effects of fatigue on the H-Reflex in the femoral nerve.

<u>Description of the study:</u>

The purpose of this research is to investigate the effect of functional fatigue on the H-Reflex in female division 1 soccer players. Prior to muscular fatigue the H-Reflex of each participant will be measured. Through using the BioPac system, a skin electrode will be used to elicit the femoral nerve's H reflex. Further, electromyography (EMG) skin electrodes will be used to monitor muscle excitability in the vastus medialis muscle. At least three measures for the maximum H reflex will be recorded in order to calculate the mean during analysis.

Muscular fatigue in this muscle will be achieved through one bout of a Wingate test, which includes pedaling as hard as possible on a stationary bike against a resistance of 8.6% of body weight for 20 seconds. To calibrate these measurements, we will collect simple measurements of body weight for the Wingate Bike Test. Muscle activity will again be recorded through the EMG technique while the stimulating electrode will be again used to elicit the femoral nerve H-Reflex. The voltage at which the first H reflex was found will again be tested post fatigue to compare to pre Wingate results. Furthermore the voltage will be increased in order to try and find the new maximum H reflex, repeated a minimum of three times to find a mean for analysis. Due to the hypothesis, it is predicted that in order to find the H reflex post fatigue the intensity of the stimulating electrode must be increased. All values of the H-Reflex, once found, will be repeated at least three times for statistical analysis of the mean.

If recovery of the H-Reflex is seen, back to baseline levels, the H reflex will be retested and post Wingate results retested at the discretion of the participant.

Risks and Discomforts

Depending on individual's differences exercising on a bicycle might present a variation in responses. As with any other physical activity, there is a slight risk that mild musculoskeletal injuries may occur. As a safety mechanism that protect against overexertion is the short duration of the activity of the exercise bout. We will take measures to ensure that the risk of injury is extremely minimal and no greater than the risk associated with any other physical activity. However in the event that any injuries occur, immediate first aid will be provided at no charge (all researchers in the lab are first aid and CPR certified). In the unlikely event that any more serious injury occurs, the participant will be responsible for seeking and paying medical expenses.

Benefits

The expected benefits to society include increased understanding of how repeated bouts of fatigue affect the body's ability to perform physical tasks and how the nervous system reflex is affected. Many ACL injuries occur in the late stages of soccer games when the body is in a fatigued state and to date the effects of fatigue and its contribution to injury are poorly understood. This research study would be unique in that few studies measure the effects of fatigue on the H-Reflex, especially in the femoral nerve.

There are no expected benefits to the individual participant.

Confidentiality and Records

All personal information will be kept confidential and will be placed in secured files that will be accessed only by the researchers. You will be given an identity for our data purposes that do not have your name in it. This study will be published for scientific purposes.

Contact Information

Please do not hesitate to ask the investigator(s) if you have any questions during the orientation session or anytime throughout the study.

Investigator	Faculty Sponsor
Laura Jackson	Dr. Craig McGowan
University of Idaho	University of Idaho
Biological Sciences	WWAMI/Biological Sciences
Moscow, ID 83844-3051	Moscow, ID 83842
Ph. 914-410-7169	Ph. 208-885-6598

During the course of this study, you may stop at any time with no penalty.

If you do stop your participation in the study, there will be no penalties associated with your withdrawal. All you need to say is that I no longer wish to participate.
I have reviewed this consent form and understand and agree to its contents.

Participant Name ________________________________Date ____________

Witness name (if appropriate) ____________________________________

Date of Birth Line ____

Experimenter Name ____

Flyer

Example flyer to solicit participation posted in the Kibbie Dome on the University of Idaho campus.

Research Study Participants Needed

<u>Are you a division 1 soccer player at the University of Idaho? Want to know how fatigue following exercise influences your ability to protect your joints from ligament damage? Participation in this important research study will help us understand the effects of fatigue on the reflexes of the legs after maximal exercise.</u>

If you are between 18-23 years of age and would like participating in a study to measure the effects of fatigue following short bursts of cycling effort, please keep reading.

We are looking for female NCAA Division 1 Soccer players to participate in a research study that will measure the effects of fatigue on the reflexes of the quadriceps muscles. A series of tests will give us a glimpse of your neuromuscular ability to perform activities following physically fatiguing events.

All participants must be willing to:

- Come to the information session on____________________
- Come to our lab and learn and practice cycling bouts of maximal intensity and the procedure involved in stimulating the femoral nerve.
- Return to our lab for nerve stimulation pre and post maximal cycling bouts of exercise.

All laboratory tests will be absolutely no cost to you. If you are interested in participating, please call or e-mail: 914-410-7169 or ljackson@uidaho.edu

Thank you.

Screening Form

Subject Number: ____________________

Listed below are the exclusion criteria for the study:

- **do not sign informed consent form,**
- **do not meet age criteria,**
- **have contraindications for maximal intensity exercise (based on ACSM guidelines),**
- **have a history of back injury,**
- **or if they have had lower extremity injury or surgery 6 months prior to data collection.**
- **All participants will have been be screened by a Certified Athletic Trainer and will be excluded from this study if they have any lumbar spine or lower extremity pain or dysfunction.**

1. Please tell us whether or not you have had any surgeries in the last six month. If so, what kind of surgery?

2. Please tell us whether you have had any history of any orthopedic, pain or discomfort particularly in the lower extremities or the back in the last six months.

3. Please keep us informed if you noticed any pain or comfort while participating in our study.

Please mark YES, No or "I do not know" to the following:

1. Do you have any cardiorespiratory diseases/disorders, systematic chronic illness, injuries, additional orthopedic or musculoskeletal problems or any other health problems that causes you pain or Physical limitations while exercising (i.e, Asthma, diabetes, osteoporosis, high blood pressure, high cholesterol, arthritis, anorexia, bulimia, anemia, epilepsy, respiratory ailments, back problems, bursitis, neck, wrist, shoulders, bad knee, etc.)?

Yes ________________________ I do not know____________

2. Do you have any concerns about the safety of exercise?

Yes ________________________ I do not know____________

- If you have marked **YES** to any of the above, please elaborate and specify below, If you marked "I do not know", it is important that you know before you can participate in this study:

____

____

____

3. Are you pregnant now or have you given birth within the last 6 months?

Yes ______________________ I do not know____________

4. Do you take any medications, either with prescription or without prescription? Yes___ No____

a) What is the medication for?____

b) How does this medication affect your ability to exercise or achieve your fitness goals?

____