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HIP MUSCLE FUNCTION IN INDIVIDUALS WITH CHRONIC ANKLE
INSTABILITY

A Dissertation

Submitted to the John G. Rangos Sr. School of Health Sciences

Duquesne University

In partial fulfillment of the requirements for
the degree of Doctor of Philosophy

By

Lindsay A. Carroll

December 2021

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Lindsay A. Carroll

2021

HIP MUSCLE FUNCTION IN INDIVIDUALS WITH CHRONIC ANKLE
INSTABILITY

By

Lindsay A. Carroll

Approved November 5, 2021

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ABSTRACT

HIP MUSCLE FUNCTION IN INDIVIDUALS WITH CHRONIC ANKLE INSTABILITY

By

Lindsay A. Carroll

December 2021

Dissertation supervised by Benjamin Kivlan, PhD, PT

Purpose: Hip muscle function has been reported to be altered in individuals with chronic ankle instability (CAI). The purpose of this study was (1) to determine whether the single leg squat test (SLST) could be used to detect differences in proximal muscle activation between individuals with and without CAI and (2) to determine if there was a difference in maximum isometric body weight normalized torque output of the hip lateral rotators, extensors, and abductors between individuals with and without CAI.

Subjects: Forty-eight subjects (14 males, 34 females; median age = 22.00 years, interquartile range (IQR) = 1.00; median height = 1.68 m, IQR = 0.15; median weight = 68.04 kg, IQR = 19.84; median body mass index = 24.41 kg/m², IQR = 3.70) participated

in this study. Subjects were separated into CAI (n = 18), coper (n = 15), and control (n = 15) groups based on published criteria.

Main outcome measures: Gluteus maximus and gluteus medius muscle activation during the SLST was measured using percent maximum voluntary isometric contraction (% MVIC) activation. Strength of the hip lateral rotators, extensors, and abductors was quantified using maximum isometric body weight normalized torque.

Results: Separate one-way analyses of variance and a Kruskal-Wallis one-way analysis of variance by ranks were used to determine group differences on these measures. The CAI group demonstrated significantly more gluteus maximus muscle activation (mean activation = 36.03% MVIC, standard deviation (SD) = 10.85% MVIC) during the SLST than both the coper (mean activation = 18.30% MVIC, SD = 10.39; $p < .001$) and control (mean activation = 21.04% MVIC, SD = 8.14; $p < .001$) groups. The CAI group also demonstrated decreased strength of the hip lateral rotators (mean maximum isometric body weight normalized torque = 0.560 Nm/kg, SD = 0.13) when compared with the coper (mean maximum isometric body weight normalized torque = 0.667 Nm/kg, SD = .009, $p = .001$) and control (mean maximum isometric body weight normalized torque = 0.757 Nm/kg, SD = .009, $p < .001$) groups. The CAI group was also significantly weaker than the control group when maximum isometric body weight normalized torque of the hip extensors was compared (median maximum isometric body weight normalized torque CAI group = 1.10 Nm/Kg, IQR = 0.15, median maximum isometric body weight normalized torque control group = 1.65 Nm/Kg, IQR = 0.37; $p < .001$). There were not

significant differences between the groups on gluteus medius muscle activation during the SLST ($p = .155$) or hip abduction strength ($p = .02$). There were no significant differences between the copers and control groups for any of the main outcome measures.

Conclusions: Subjects with CAI performed the SLST with significantly more gluteus maximus activation than subjects without CAI. Subjects with CAI demonstrated significantly decreased strength of the hip lateral rotators and extensors than subjects without CAI.

Clinical Relevance: The results of the study suggest that the SLST has potential for use as a clinical measure of gluteus maximus muscle function in individuals with CAI. Further study is needed to determine whether the differences in activation that occurred are clinically detectable. This study's findings also support previous work showing that individuals with CAI have weakness of the hip lateral rotators and extensors when compared to individuals without CAI.

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Chapter 1: Introduction

1.1 Background

Lateral ankle sprains (LAS) are the most prevalent musculoskeletal injury sustained by active individuals,¹ occurring at a rate of 11.55 per 1000 exposures.² Acute and subacute complications of lateral ankle sprains include pain,³⁻⁵ instability,^{3,4,6} swelling,³⁻⁵ decreased strength,^{4,7} decreased range of motion,³⁻⁵ and decreased level of function.^{3,8,9} Sensorimotor deficits, such as impaired proprioception, postural control, decreased strength, and changes in both local and proximal muscle activity have also been reported.^{5,10-14} More than half of individuals that sustain a LAS report that they have not fully recovered 3 years after the initial injury.³ When impairments associated with a LAS persist for greater than one year following an index lateral ankle sprain (LAS), individuals are considered to have chronic ankle instability (CAI).^{4,15,16}

CAI occurs in 40-70% of individuals who sustain a LAS.^{4,16,17} It is characterized by recurrent sprains or feelings of instability about the ankle, decreased range of motion, decreased strength, and decreased function.^{4,15,16} CAI has also been related to decreased ability to participate in physical activity⁸ and lower reported health-related quality of life.¹⁸⁻²¹ Annual societal costs of LAS and CAI in the United States have been estimated to be \$6.2 billion,¹⁵ making effective management of LAS and CAI a significant concern.

Although physical therapy interventions have been shown to be effective at resolving acute deficits related to LAS and CAI,¹⁵ the high prevalence of persistent symptoms and disability^{3,22,23} following LAS is concerning. Prolonged deficits following LAS may occur because current physical therapy management techniques are not

adequately identifying and addressing the impairments associated with CAI. Despite the evidence showing that there are hip-centric impairments in this population,^{11, 13, 14, 24-39} the typical physical therapy examination for a patient with LAS or CAI may not include functional evaluation of the hip complex.^{5, 40} It may be beneficial for physical therapists to evaluate neuromuscular function of the hip, as the literature consistently identifies impairments in hip muscle strength and muscle activation patterns in individuals with CAI.^{11-14, 24-26, 34}

Moderate to large effect sizes¹² have been reported in studies examining eccentric isokinetic and isometric hip muscle performance²⁴⁻²⁷ in individuals with CAI. These findings are shown in Tables 1 and 2. Negahban et al.²⁷ found lower isokinetic eccentric hip flexor strength at 60 degrees/second in individuals with CAI when compared to healthy controls. Decreased isometric hip lateral rotation, extension, and abduction strength has also been identified in individuals with CAI.²⁴⁻²⁶ When the results of individual reports on isometric hip muscle performance were pooled, there were significant differences in triplanar muscle performance between individuals with and without CAI.¹²

Table 1. Proximal changes reported in individuals with CAI: muscle performance.

Author year	Subjects	Task, measurement	Significant results
Negahban 2013 ²⁷	40 subjects (20 with CAI, 20 controls)	<ul style="list-style-type: none"> Eccentric torque output (isokinetic dynamometer, 60° per second) 	<ul style="list-style-type: none"> CAI group had lower eccentric hip flexor torque at 60° per second bilaterally.
McCann 2017 ²⁵	105 subjects (30 with CAI, 29 copers, 26 healthy controls)	<ul style="list-style-type: none"> Isometric strength (hip abduction, lateral rotation, extension) 	<ul style="list-style-type: none"> CAI group had lower isometric hip abduction strength compared to copers. CAI group had lower lateral rotation strength compared to copers and controls.
McCann 2018 ²⁴	60 subjects (20 with CAI, 20 copers, 20 healthy controls)	<ul style="list-style-type: none"> Isometric strength (hip abduction, lateral rotation, extension) 	<ul style="list-style-type: none"> CAI group had less hip extension and lateral rotation strength than coper and control groups. No differences in abduction strength.
McCann 2019 ²⁶	76 subjects (26 with CAI, 25 copers, 25 healthy controls)	<ul style="list-style-type: none"> Single leg landings Isometric strength (hip abduction, lateral rotation, extension) 	<ul style="list-style-type: none"> CAI group had less lateral rotation strength than coper and control groups. CAI group may have less hip extension strength than controls (based on effect size, $d=0.63$) but this finding was not statistically significant. Hip strength not associated with kinematics at the hip during landing.

Table 2. Differences in hip muscle performance among individuals with CAI, copers, and controls.

Key:

^: maximum isometric body weight normalized torque output

‡: eccentric average peak torque to body weight ratio at 60°/second

*: statistically significant finding (relative to CAI group) at $p < 0.05$

‡: moderate to large effect size (pairwise comparisons versus CAI group)

		CAI	Copers	Controls
Lateral rotators^	McCann 2017	0.5 Nm/kg	0.6 Nm/kg*‡	0.7 Nm/kg*‡
	McCann 2018	0.47 Nm/kg	0.55 Nm/kg*	0.58 Nm/kg*
	McCann 2019	0.57 Nm/kg	0.66 Nm/kg *	0.70 Nm/kg *
Extensors^	McCann 2018	0.98 Nm/kg	1.30 Nm/kg*	1.38 Nm/kg*
	McCann 2019	1.28 Nm/kg	1.49 Nm/kg	1.53 Nm/kg‡
Abductors^	McCann 2017	1.4 Nm/kg	1.7 Nm/kg*‡	1.8 Nm/kg*‡
Flexors‡	Negahban 2013	1.41 N•m ⁻¹ •kg ⁻¹	Not studied	1.94 N•m ⁻¹ •kg ⁻¹ *

Differences in hip muscle activity during closed kinetic chain tasks have also been identified in individuals with CAI.^{11, 13, 14, 28-30, 32-34} These findings are displayed in Table 3. Differences in hip muscle activity have been associated with moderate (Cohen's $d = 0.4 - 0.79$) to large (Cohen's $d \geq 0.8$) effect sizes,¹² indicating that the magnitude of strength deficits reported in individuals with CAI was at least 0.4 standard deviations below that of the control group. Changes in the amount of gluteus medius muscle activation have been reported during treadmill walking^{11, 14, 28} and in the stance extremity during kicking.³⁰ Individuals with CAI have also been reported to perform the Star Excursion Balance Test (SEBT)³³ and drop landings³² with altered timing of gluteus medius³³ and tensor fascia lata³² when compared to individuals without CAI. Differences in the amount^{13, 33, 34} and timing^{29, 33} of gluteus maximus activation have also been reported during dynamic squatting tasks^{13, 29, 33} and a hop landing.³⁴ Though this evidence suggests that there may be hip-centric movement system dysfunctions occurring in

individuals with CAI, the broad range of tasks and laboratory-based measurement techniques utilized in this research make application of this evidence challenging for clinicians.

Table 3. Proximal changes reported in individuals with CAI: muscle activity.

Abbreviations: sEMG: surface electromyography, CAI: chronic ankle instability

Author year	Subjects	Task, measurement	Significant results
Fu 2007 ³²	<ul style="list-style-type: none"> 39 male basketball players (19 with bilateral multiple ankle sprains, 20 healthy controls) 	<ul style="list-style-type: none"> Landing from expected and unexpected drops sEMG of tensor fascia lata (TFL) 	<ul style="list-style-type: none"> Later activation of TFL after expected but not unexpected drops in the multiple ankle sprains group
Webster 2013 ²⁹	<ul style="list-style-type: none"> 18 subjects (9 with CAI, 9 healthy controls) 	<ul style="list-style-type: none"> 10 repetitions of a rotational lunge and a single leg rotational squat sEMG of gluteus medius and gluteus maximus 	<ul style="list-style-type: none"> CAI group has significantly lower gluteus maximus activation during the rotational squat
Rios 2015 ³⁰	<ul style="list-style-type: none"> 42 subjects (21 with CAI, 21 healthy controls) 	<ul style="list-style-type: none"> Single leg stance on compliant and non-compliant surface while kicking a ball sEMG of biceps femoris, rectus femoris, gluteus medius 	<ul style="list-style-type: none"> CAI group has increased proximal muscle activity relative to distal activity during compensatory adjustments following the kicking task
Koldenhoven 2016 ²⁸	<ul style="list-style-type: none"> 34 subjects (17 with CAI, 17 healthy controls) 	<ul style="list-style-type: none"> Treadmill walking sEMG gluteus medius 	<ul style="list-style-type: none"> CAI group had increased gluteus medius activity (root mean square area under curve) at pre-initial contact CAI group had higher sEMG amplitude of gluteus medius during the last 50% of stance and the first 25% of stance
Webster 2016 ³⁴	<ul style="list-style-type: none"> 32 subjects (16 with CAI, 16 healthy controls) 	<ul style="list-style-type: none"> Lateral hop landing with and without fatigue sEMG of gluteus medius and gluteus maximus 	<ul style="list-style-type: none"> CAI group had higher gluteus maximus activity just before landing

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Jaber 2018 ³³	<ul style="list-style-type: none"> 48 subjects (16 with CAI, 16 copers, 16 healthy controls) 	<ul style="list-style-type: none"> Star Excursion Balance Test (SEBT) sEMG of gluteus medius and gluteus maximus 	<ul style="list-style-type: none"> CAI group had less gluteus maximus activation during the posterolateral reach direction on the SEBT CAI group activated gluteus maximus later than control group during the SEBT in the anterior direction CAI group activated gluteus medius later than the coper group during the SEBT in the posteromedial direction
DeJong 2019 ¹⁴	<ul style="list-style-type: none"> 40 subjects (20 with CAI, 20 healthy controls) 	<ul style="list-style-type: none"> Gait Ultrasound imaging was utilized to obtain functional activation ratios of gluteus medius and gluteus maximus 	<ul style="list-style-type: none"> CAI group had decreased gluteus medius activity bilaterally during gait from 0-40% of the gait cycle
DeJong 2020 ¹¹	<ul style="list-style-type: none"> 28 females (14 with CAI, 14 copers) 	<ul style="list-style-type: none"> Treadmill walking at 3 different speeds Ultrasound imaging was utilized to obtain functional activation ratios of gluteus medius and gluteus maximus 	<ul style="list-style-type: none"> CAI group had decreased gluteus medius activity at all three gait speeds
DeJong 2020 ¹³	<ul style="list-style-type: none"> 40 subjects (20 with CAI, 20 healthy controls) 	<ul style="list-style-type: none"> Y-balance test (YBT) Ultrasound imaging was utilized to obtain functional activation ratios of gluteus medius and gluteus maximus at the maximum reach point of the YBT 	<ul style="list-style-type: none"> CAI group had greater gluteus maximus functional activation ratios than the control group.

Validated functional performance tests may allow clinicians to detect the neuromuscular impairments that have been identified in biomechanical studies. However, most of the functional performance tests that have been validated for use in a population with LAS and CAI have been shown to differentiate between individuals with and without ankle instability,⁴¹⁻⁴⁷ elicit complaints of instability,^{5, 48, 49} and detect differences in balance^{33, 43, 50-55} rather than to detect proximal muscle activation patterns. There is some evidence that the SEBT³³ and Y-balance test¹³ (YBT) can detect differences in hip muscle activity in individuals with CAI, but performance on these tests is also associated with other impairments, such as decreased ankle dorsiflexion range of motion,⁵³ impaired balance,^{53, 56} and decreased ankle strength,⁵³ making it difficult for clinicians to identify hip muscle activation impairments based on these tests. Additionally, performance on the SEBT and the YBT is measured based on reach distances and there is no evidence of a relationship between reach distance and hip muscle activity or performance.^{13, 33} Currently, there is not a functional performance test that has been validated to specifically detect hip muscle activation impairments in individuals with CAI.

The single leg squat test (SLST) is a functional performance test that may help physical therapists identify proximal muscle impairments in individuals with CAI, as movement quality on this test has been related to deficits in hip muscle performance and activation patterns.⁵⁷⁻⁶⁴ Poor performance on the SLST has been related to decreased hip muscle strength^{59-63, 65-67} and altered neuromuscular function of gluteus medius^{57, 64, 65, 68, 69} and gluteus maximus.^{58, 68} The SLST has been used to detect these neuromuscular deficiencies in healthy individuals^{57, 58, 60} and in individuals with lower extremity orthopedic dysfunctions such as patellofemoral pain syndrome^{65, 69, 70} and individuals who were status post anterior cruciate ligament

reconstruction.⁵⁹ Further, strength training of the hip has been shown to improve SLST performance in recreational runners⁶³ and in individuals with pre-arthritis hip pain.⁷¹ The ability of the SLST to serve as an indicator of closed kinetic chain hip muscle activity and performance makes it a candidate for use in individuals with CAI.

The reliability and responsiveness of the SLST also make it a candidate for clinical use with individuals with CAI. A 2019 systematic review of the reliability of visual assessment of the SLST reported moderate inter-rater reliability ($\kappa = 0.58$, 95%CI = 0.5 – 0.65) and substantial intra-rater reliability ($\kappa = .68$, 95% CI = 0.60-0.74).⁷² The SLST has also been shown to be capable of detecting functional improvement in individuals with pre-arthritis hip pain.⁷¹ Although the reliability and responsiveness of the SLST have not been studied in individuals with CAI, these findings in other populations suggest that the SLST may have potential as a valuable functional performance test for clinicians treating patients with CAI.

The validity of the SLST as an indicator of hip muscle performance and activation, its reliability, and its responsiveness make the SLST a functional performance test that may help clinicians assess dysfunctional hip muscle activity in individuals with CAI. Validation of this test for use in this population could help clinicians identify and manage neuromuscular dysfunction about the hip that may be limiting return to function in individuals with CAI. Therefore, the purpose of this study was (1) to determine whether the SLST could be used to detect differences in proximal muscle activation between individuals with and without CAI and (2) to determine if there was a difference in maximum isometric body weight normalized torque output of the hip lateral rotators, extensors, and abductors between individuals with and without CAI.

1.2 Operational definitions

1. Chronic ankle instability (CAI) group: This group consisted of individuals fitting the guidelines for research on CAI established by the International Ankle Consortium in 2014.¹⁶ Individuals in this group had: (1) a history of at least one significant ankle sprain that was associated with a loss of at least 1 day of physical activity and inflammatory symptoms (pain, swelling), the first of which occurred ≥ 12 months ago, and the most recent of which occurred ≥ 3 months ago; (2) a history of recurrent sprains, feelings of instability, and/or a history of the ankle “giving way”; (3) a score of > 11 on the Identification of Functional Ankle Instability (IdFAI) patient-reported outcome measure; (4) a score of $< 90\%$ on the Foot and Ankle Ability Measure activities of daily living (FAAM-ADL) scale and a score of $< 80\%$ on the Foot and Ankle Ability Measure sports (FAAM-sports) scale.
2. Individuals without ankle instability: This general term includes individuals in both the coper and control group.
3. Coper group: This group was comprised of individuals that met the criteria published by Wikstrom and Brown.⁷³ Copers were individuals that had sustained a significant ankle sprain¹⁶ at least 12 months prior to enrolling in the study but had not experienced recurrent sprains, episodes of giving way, or feelings of instability.⁷³ Copers had to have returned to at least moderate levels of weight bearing physical activity for ≥ 12 months following the initial sprain.⁷³ These individuals scored ≤ 10 on the IdFAI patient-reported outcome measure and reported little to no functional disability (FAAM-ADL $> 95\%$, FAAM-sports $> 95\%$).⁷³ This group was included in the study in order to improve the

understanding of the factors that may differentiate between individuals that do and do not develop CAI after sustaining an ankle sprain.^{73, 74}

4. Control group: This group consisted of healthy individuals who had never sustained a lateral ankle sprain. They denied episodes of their ankles giving way or feelings of instability about either ankle. These individuals will have a score of > 95% on the FAAM-ADL scale, > 95% on the FAAM-sports scale, and ≤ 10 on the IdFAI patient-reported outcome measure.
5. Significant ankle sprain: An ankle sprain that was associated with inflammatory symptoms, such as pain and swelling, and resulted in at least 1 day of interruption of desired physical activity.¹⁶
6. Index sprain: The first occurrence of a lateral ankle sprain.⁷⁵
7. Recurrent sprain: Two or more sprains in the same ankle.¹⁶
8. Giving way: Regular, uncontrolled, excessive inversion of the rearfoot that does not result in a lateral ankle sprain.¹⁶
9. Feeling of ankle joint instability: The subjective report of instability or the fear that a lateral ankle sprain may occur during activities of daily living or sports.¹⁶
10. Single leg squat test (SLST): This is a functional performance test that was performed as described by McGovern et al.⁷⁶ In this protocol, a “T” (6” horizontal, 10” vertical) was taped on the floor using 1.5” wide athletic tape. The subject stands on the vertical tape on their unshod test extremity and flexed the other knee to 90°. They were instructed to squat until they could no longer see the horizontal tape in front of their toes and then return to the starting position.

11. Surface electromyography (sEMG): This technology utilizes adhesive electrodes that are applied to the skin to measure the timing and amount of muscle activity.
12. Muscle function: This general term refers to the qualities of muscle performance, timing of muscle activity, and amount of muscle activation.
13. Muscle performance: This term refers to qualities of muscular strength, endurance, and power.
14. Muscle strength: The ability of a muscle to generate force.
15. Maximal voluntary isometric contraction (MVIC): This is an open-chain, maximal-effort, isometric contraction of selected muscle groups. Output will be measured in Newtons using a handheld dynamometer.
16. Maximal voluntary isometric contraction activation (MVIC activation): The amount of muscle activation that is measured during the MVIC. The highest consecutive 0.5 seconds of muscle activation that occurs during any of the 3 MVIC trials represents 100% MVIC⁶⁴ and is used to normalize the amount of muscle activity occurring during the SLST.
17. Percent maximal voluntary isometric contraction (% MVIC): This represents the amount of muscle activation that occurs relative to the maximal amount of muscle activation that was measured during the MVIC activation test. This is a normalized value that determines the maximal amount of muscle activation by measuring the amount of muscle activity that occurs during a maximal voluntary isometric contraction.

1.3 Limitations and assumptions

1. Instrumentation was applied in the same manner to all subjects and testing conditions:
 - a. sEMG electrodes
 - b. Inertial measurement units (IMUs)
 - c. Stabilization and dynamometer placement for MVIC tests
2. Electrical noise, skin impedance, and room temperature were consistent across all subjects during testing.
3. The amount of muscle activity that was measured during MVIC testing is representative of the amount of the maximum amount of muscle activation possible at that muscle's recording site.
4. The amount of force that was measured during MVIC testing was representative of the maximal amount of force the muscle was capable of generating.
5. Subjects gave consistent effort during testing.
6. sEMG detected the amount of muscle activity accurately and consistently.
7. Observed differences in sEMG during the SLST between the three groups (CAI, coper, control) were representative of the proximal neuromuscular functional status of these groups.
8. Observed differences in force output between the three groups (CAI, coper, control) were representative of the proximal neuromuscular functional status of these groups.
9. The order of testing could not be randomized: MVIC testing had to occur prior to SLST testing for each subject in order to calculate the percent of MVIC activation occurring during the SLST.

10. Any differences that were observed between the groups were representative of muscle function at the time of testing. Temporal relationships of the development of CAI and differences in muscle function cannot be interpreted from this study.

1.4 Delimitations

1. Forty-eight subjects (18 with CAI, 15 copers, and 15 controls) were recruited from Duquesne University, Shenandoah University, and the local communities. An *a priori* power analysis was performed using power = 0.8, $\alpha = 0.01$, the anticipated use of 5 one-way analyses of variance (ANOVAs), and an effect size of 0.6. The effect size was determined based on Webster et al.'s²⁹ 2013 publication reporting an effect size of Cohen's $d = 0.71$. It was expected that the SLST would have required less gluteus maximus activation than the rotational squatting exercises used by Webster²⁹ and would thus be associated with a lower effect size, so $d = 0.6$ was used for the power analysis. The power analysis suggested a sample size of 45 subjects. Three additional subjects were included in the CAI group in case of the event of lost data. Alpha was adjusted to 0.01 from 0.05 to account for multiple comparisons on the same dataset. Alpha for post hoc testing was also adjusted to account for multiple comparisons and was set at 0.0033. This number of subjects was determined to be attainable given the high prevalence of individuals with a history of ankle sprains in the community^{1, 2} and Koshino et al's⁷⁷ reported prevalence of college-age athletes that met the research inclusion criteria for CAI.

2. Subjects were grouped based on the inclusion/exclusion criteria described by the International Ankle Consortium for selecting research subjects with CAI¹⁶ and Wikstrom and Brown's⁷⁴ guidelines for copers.
3. Subjects were between the ages of 18 and 45 years.
4. Subjects followed the evidence-based SLST protocol⁷⁶ so that ankle dorsiflexion range of motion and foot posture were unlikely to be different between those that pass and fail the SLST.⁷⁸
5. Only the amount of muscle activation (% MVIC) and maximal isometric output of selected proximal contractile structures were measured. This included the amount of muscle activity (% MVIC) of the gluteus maximus and gluteus medius muscles and the maximum isometric body weight normalized torque output of the hip lateral rotators, extensors, and abductors.

1.5 Purpose statement

The purpose of this study was (1) to determine whether the SLST could be used to detect differences in proximal neuromuscular function (measured by determining the amount of muscle activity occurring during the SLST) between individuals with CAI and without CAI (copers and controls); and (2) to determine whether there is a difference in maximum isometric body weight normalized torque output of the hip lateral rotators, extensors, and abductors between individuals with CAI, copers, and controls.

1.6 Independent variable

The independent variable was instability status. This variable had three levels: CAI, coper, control.

1.7 Dependent variables

1. Amount of muscle activity during the SLST, as measured by sEMG.
 - a. Percent of MVIC activation of gluteus maximus occurring during the SLST
 - b. Percent of MVIC activation of gluteus medius occurring during the SLST
2. Maximum isometric body weight normalized torque produced by the hip lateral rotators.
3. Maximum isometric body weight normalized torque produced by the hip extensors.
4. Maximum isometric body weight normalized torque produced by the hip abductors.

1.8 Research questions

1. Is there a difference in the percent of MVIC activation of gluteus maximus utilized during the SLST by individuals with CAI and without CAI?
2. Is there a difference in the percent of MVIC activation of gluteus medius utilized during the SLST by individuals with CAI and without CAI?
3. Is there a difference in the amount of maximum isometric body weight normalized torque generated during a maximum voluntary isometric contraction of the hip lateral rotators between individuals with and without CAI?

4. Is there a difference in the amount of maximum isometric body weight normalized torque generated during a maximum voluntary isometric contraction of the hip extensors between individuals with and without CAI?
5. Is there a difference in the amount of maximum isometric body weight normalized torque generated during a maximum voluntary isometric contraction of the hip abductors between individuals with and without CAI?

1.9 Hypotheses

The primary hypothesis of this study was that individuals with CAI would demonstrate a different amount of muscle activity of the gluteus maximus and/or gluteus medius muscles during the SLST when compared to individuals without CAI. The secondary hypothesis was that individuals with CAI would produce less maximum isometric body weight normalized torque of the hip lateral rotators, extensors, and abductors than subjects without CAI.

Hypotheses by research question

1. There will be a difference in the amount of gluteus maximus activation utilized by individuals with CAI and without CAI during the SLST.
2. There will be a difference in the amount of gluteus medius activation utilized by individuals with CAI and without CAI during the SLST.
3. There will be a difference in maximum isometric body weight normalized torque of the hip lateral rotators between individuals with CAI and without CAI.

4. There will be a difference in maximum isometric body weight normalized torque of the hip extensors between individuals with CAI and without CAI.
5. There will be a difference in maximum isometric body weight normalized torque of the hip abductors between individuals with CAI and without CAI.

Chapter 2: Literature review

2.1 Introduction

Chronic ankle instability (CAI) is a multi-factorial,^{4, 79, 80} costly¹⁵ problem that has been related to long-term disability^{3, 22, 23} and decreased health-related quality of life.¹⁸ Impairments that are commonly associated with CAI include the feeling of instability or giving way,^{40, 41, 47, 48} decreased balance,^{33, 40, 50-53, 81, 82} decreased range of motion,^{40, 53, 83} impaired joint mobility,^{40, 79, 84} decreased muscle performance,^{53, 85} and altered muscle activation patterns.^{13, 33, 79, 85} Deficits in hip strength^{12, 24-27} and activation^{11-14, 28, 29, 32, 34} have recently been identified as impairments of interest in this population but these have not yet been studied or captured in functional performance tests. The single leg squat test (SLST) is a functional performance test that shows promise to detect these impairments,^{57, 60, 66-68, 86, 87} but this has not yet been investigated in the CAI population.

The purpose of this review is: (1) to describe the neuromuscular impairments occurring at the hip that have been reported in individuals with CAI, (2) to demonstrate that the clinical functional performance tests that are validated for use in those with CAI population do not detect these impairments, and (3) to propose the SLST as a functional performance test that may be useful for detecting hip muscle in a population with CAI. This review will synthesize peer-reviewed literature reporting neuromuscular deficits in individuals with CAI, including evidence describing deficits in muscular performance (strength and endurance) and activation (amount of activity, as measured by surface electromyography [sEMG] or musculoskeletal ultrasound) occurring in muscles with actions at the hip. Next, this review will describe the impairment-

based validity of functional performance tests for use in individuals with CAI. Finally, this review will describe the validity of the SLST for detecting hip-centric neuromuscular deficits and will briefly report the reliability of visual grading schemes used in clinical settings.

2.2 Chronic ankle instability

Outcomes following a lateral ankle sprain occur along a spectrum of patient presentations ranging from complete return to prior level of function without symptoms (coper) to an unfavorable diagnosis of chronic ankle instability.⁴ The ability of an individual to function at their prior level without symptoms distinguishes a coper from an individual with CAI⁴. A coper may or may not have impairments similar to those of an individual with CAI, but a coper's impairments do not impact their function.⁴ Clinical diagnosis of chronic ankle instability is not impairment-based, but occurs when an individual has decreased function and symptoms related to instability related to an index sprain that occurred at least 1 year prior.^{4, 16}

For research purposes, individuals with CAI are individuals who, \geq 1-year following an index ankle sprain,: (1) score > 11 on the Identification of Functional Ankle Instability (IdFAI) outcome measure or score ≤ 24 on the Cumberland Ankle Instability Tool (CAIT), and (2) score $< 90\%$ on the Foot and Ankle Ability Measure Activities of Daily Living scale (FAAM-ADL) and $< 80\%$ on the Foot and Ankle Ability Measure Sport scale (FAAM-sport).^{4, 16} An individual is classified as a coper if, at ≥ 1 year following the initial ankle sprain, they (1) score ≥ 28 on the CAIT or ≤ 10 on the IdFAI, (2) score $\geq 95\%$ on both the FAAM-ADL and FAAM-sports, and (3) have not experienced episodic giving way of the ankle or recurrent sprains.^{4, 74} Both subjects with CAI and copers should have a history of a sprain that occurred at least 1 year prior and was

associated with pain and/or swelling and resulted in the loss of at least 1 day of desired physical activity.¹⁶ Table 4 shows the criteria for classification as an individual with CAI or a lateral ankle sprain copper.

Table 4. Criteria for subjects to be classified as an individual with CAI or copper for research purposes.

**All three criteria must be met in order for a subject to be classified as an individual with CAI*

Criteria	Chronic Ankle Instability	Copper
History of sprain*	≥ 1 year prior	≥ 1 year prior
Instability score*	IdFAI > 11 OR CAIT < 24	IdFAI ≤ 10 OR CAIT < 24
Functional score*	FAAM- ADL < 90% AND FAAM- sports < 80%	FAAM- ADL > 95% AND FAAM- sports > 95%
Episodic giving way	Yes	No

The model describing CAI is multi-factorial and involves pathomechanical, sensory-perceptual, and motor-behavioral impairments imposed on the biopsychosocial model.⁴ There are well-studied movement system impairments and targeted intervention strategies^{4, 5, 40, 55, 88, 89} associated with CAI, but, despite this, disability, health-related quality of life, and impaired function persist in this population.¹⁸⁻²¹ Physical impairments that may be clinically identified in individuals with CAI include local deficits such as decreased foot and ankle range of motion and joint mobility, decreased local muscle performance, changes in neuromuscular control about the ankle, altered proprioception, decreased balance.^{4, 5, 80} New evidence, however, shows that impairments and alterations in movement behavior in individuals with CAI may occur

proximally, about the hip joint, as well as in the contralateral extremity.^{12, 79, 90} These changes can include characteristics related to hip muscle performance,^{12, 24-27} and neuromuscular recruitment patterns.^{11-14, 28, 29, 32-34, 91} It is possible that these proximal impairments are contributing to the inability of individuals with CAI to return to their prior level of function. The following sections will explore the neuromuscular changes that have been reported to occur in the gluteus maximus, gluteus medius, and hip lateral rotator muscles in individuals with CAI, including changes in muscle force output and neuromuscular recruitment patterns during functional tasks.

2.2.1 Evidence for neuromuscular dysfunction occurring at the hip: muscle performance

A recent meta-analysis¹² pooled results from three studies²⁴⁻²⁶ and reported that, compared to individuals without CAI, subjects with CAI had significantly impaired triplanar isometric hip strength ($p < .001$, effect size range: 0.52-0.93).¹² Decreased isometric strength of the hip lateral rotators was found in all three studies when individuals with CAI were compared with copers and controls ($p = .03$;²⁴ $p = .01$;²⁵ $p = .04$;²⁶ $p < .01$ ²⁶). Two of these studies showed that individuals with CAI had decreased hip extension strength when output was compared to both copers and controls.^{24, 26} These differences were statistically significant in one of the studies ($p = .02$)²⁴ and were associated with a moderate effect size in the other (Cohen's $d = 0.63$).²⁶ Isometric strength of the hip abductors was also found to be significantly diminished in individuals with CAI when compared with lateral ankle sprain copers ($p = .03$)²⁵. Although the differences in normalized torque measurements were small (Table 5), decreased hip strength was found to significantly influence functional movement.²⁵ Decreased strength of the lateral rotators

and abductors was associated with worse performance on the Star Excursion Balance Test (SEBT) ($R^2 = .25$, $p = .01$),²⁵ suggesting that there may be a relationship between isometric hip strength and movement system dysfunction.

Table 5. Differences in hip muscle performance among individuals with CAI, copers, and controls.

Key:

^: maximum isometric body weight normalized torque

‡: eccentric average peak torque to body weight ratio at 60°/second

*: statistically significant finding (relative to CAI group) at $p < 0.05$

‡: moderate to large effect size (pairwise comparisons versus CAI group)

		CAI	Copers	Controls
Lateral rotators [^]	McCann 2017	0.5 Nm/kg	0.6 Nm/kg*‡	0.7 Nm/kg*‡
	McCann 2018	0.47 Nm/kg	0.55 Nm/kg*	0.58 Nm/kg*
	McCann 2019	0.57 Nm/kg	0.66 Nm/kg *	0.70 Nm/kg *
Extensors [^]	McCann 2018	0.98 Nm/kg	1.30 Nm/kg*	1.38 Nm/kg*
	McCann 2019	1.28 Nm/kg	1.49 Nm/kg	1.53 Nm/kg‡
Abductors [^]	McCann 2017	1.4 Nm/kg	1.7 Nm/kg*‡	1.8 Nm/kg*‡
Flexors [‡]	Negahban 2013	1.41 N•m ⁻¹ •kg ⁻¹	Not studied	1.94 N•m ⁻¹ •kg ⁻¹ *

Further evidence of decreased hip muscle performance among individuals with CAI has been shown in studies examining eccentric torque²⁷ and the impact of strength training on function.⁹² Negahban et al.²⁷ found that individuals with CAI had decreased eccentric torque output of the hip flexors at 60°/second when average peak torque to body weight ratio was compared with healthy controls that had never experienced an ankle sprain ($p = 0.03$; see Table 5). Evidence of meaningful impairments in hip muscle performance was also demonstrated by Smith et al.,⁹² who studied the effects of a hip muscle strengthening program in individuals with

CAI. This study showed that individuals with CAI that participated in the training program had improvements in strength of the hip abductors ($p < .001$) and hip lateral rotators ($p < .001$), and score improvements on the Foot and Ankle Ability Measure – sports subscale (FAAM-sports) ($p < .001$) when compared to group that did not participate in the training program.⁹² Thus, an improvement in hip strength was associated with a significant improvement in FAAM-sports score in individuals with CAI that underwent a hip strengthening program.⁹² The findings of these investigations suggest that hip muscle performance may be an impairment that is associated with the limitations in function that define this population.^{27, 92}

2.2.2 Evidence for neuromuscular dysfunction occurring at the hip: muscle activation

Individuals with CAI have been reported to demonstrate differences in hip muscle activation patterns during closed chain tasks.^{11, 13, 14, 28, 29, 32-34, 91} Variations in muscle activation patterns occurring in individuals with CAI that will be explored include differences in the amount of muscle activity of gluteus maximus and gluteus medius that have been measured using electromyography and musculoskeletal ultrasound.^{11, 13, 14, 28, 29, 33, 34, 91} When electromyography is used to measure muscle activity, the amount of muscle activity utilized by a subject during a task is represented by the percent of muscle activity occurring during the activity (% MVIC) relative to the maximum amount of muscle activation possible.⁹³ Maximal muscle activation is established by measuring the amount muscle activity occurring during a maximal voluntary isometric contraction (MVIC) of a muscle.⁹³ Researchers that measure muscle activity using electromyography may also use root mean square (RMS) area or area under the curve to report amounts of muscle activity occurring over a defined time period.²⁸

When musculoskeletal ultrasound imaging is used to determine the amount of muscle activity that occurs, the amount of activity is represented by functional activation ratios (FARs).^{13, 91} FARs are the ratio of muscle thickness measured during an activity to muscle thickness measured at rest and these are used as a measure of how much muscle activity is occurring during the activity.^{13, 91} Several authors have found differences in the amount of gluteus maximus and gluteus medius muscle activity occurring during closed chain tasks in individuals with CAI.^{11, 13, 14, 28, 29, 32-34, 91}

Differences in the amount of gluteus maximus activity in individuals with CAI have been reported during functional activities are described in Table 6.^{13, 29, 33, 34} Webster et al.²⁹ found that individuals with CAI used only 51.1% of their maximum gluteus maximus activity during a rotational squat. This was significantly ($p = 0.041$) lower than the amount of activity (78.6% MVIC) measured in healthy controls during this same exercise and was associated with a moderate effect size ($d = 0.71$).²⁹ Decreased gluteus maximus activity has also been reported in individuals with CAI during the Star Excursion Balance Test (SEBT).³³ Individuals with CAI performed the posterolateral reach of the SEBT with less gluteus maximus activity (25.6% MVIC) than copers and healthy controls (37.5% and 40.2%, respectively).³³ Other investigations have found that subjects with CAI may perform closed chain tasks with increased amounts of gluteus maximus activity when compared to copers and controls.^{13, 34} In 2016, Webster et al.³⁴ measured gluteus maximus activity during landing from a lateral hop. In this study, individuals with CAI had increased activation of gluteus maximus just before landing from a lateral hop (45.5%) when compared to that of healthy control subjects (36.8%).³⁴ This difference was statistically significant ($p = 0.049$) and was associated with a moderate effect size ($p = 0.71$).³⁴

Greater activity of the gluteus maximus was also reported in individuals with CAI (versus healthy subjects) when musculoskeletal ultrasound imaging was used to measure muscle activation during the anterior reach of the YBT (mean difference of functional activation ratios = 0.08, Cohen $d = 0.57$, $p = .05$).¹³ This was associated with a decrease in reach distance in the anterior direction.¹³

Table 6. Changes in gluteus maximus activity in individuals with CAI. All findings were statistically significant.

Key:

**: measured using surface electromyography; results reported as percent of maximal activation*

^: measured using musculoskeletal ultrasound; results reported as functional activation ratios (ratio of muscle thickness at rest versus muscle thickness during activity)

Task	Amount of activity: CAI group	Amount of activity: control group
Rotational squat*	51.1%	78.6%
Landing from lateral hop*	45.5%	36.8%
Star Excursion Balance Test-posterolateral*	25.6%	40.2%
Y-Balance Test-anterior [^]	1.06	1.08

Individuals with CAI have also been found to utilize different amounts of gluteus medius activation during functional tasks.^{11, 14, 28} In 2016, Koldenhoven et al.²⁸ found that, when compared with healthy controls, individuals with CAI had greater activation of gluteus medius during gait. This increase in activation occurred during the 100ms pre-initial contact (RMS area CAI group = 8.9, RMS area control group = 1.3; $p < .003$), during the final 50% of stance phase (RMS area CAI group = 3.6, RMS area healthy group = 2.4; $p < .045$), and during the first 25%

of swing phase (RMS area CAI group = 3.6, RMS area healthy group = 2.1; $p < .045$).²⁸ Two additional studies examined gluteus medius activity during gait using musculoskeletal ultrasound imaging and functional activation ratios.^{11, 14} One 2019 study showed that individuals with CAI demonstrated decreased gluteus medius activation during the first 40% of the gait cycle when compared to healthy controls (mean difference: 0.16-0.17; $p < .05$).¹⁴ These findings were supported in a similar study that was published in 2020.¹¹ This study reported that, when compared with lateral ankle sprain copers, individuals with CAI ambulated with less gluteus medius activation throughout the entire gait cycle (mean difference = 0.10 – 0.18; $p < .001$).¹¹ This difference was significant at all three gait speeds that were investigated.¹¹

2.2.3 Summary

These reports offer insight into the differences in proximal neuromuscular function in individuals with CAI. Key findings include:

- impaired hip lateral rotator, abductor, and extensor muscle strength,^{12, 24-27}
- differences in gluteus maximus activation patterns during closed kinetic chain tasks (rotational squat,²⁹ landing from a lateral hop,³⁴ anterior direction of the SEBT³³/YBT,¹³ and during the posterolateral reach direction of the SEBT³³); and
- differences in gluteus medius activity during gait.^{11, 14, 28}

Given that current management approaches allow persistent decreased function and health-related quality of life among individuals with CAI, it may be important for clinicians to assess and manage gluteal muscle strength and activity impairments in this population. Without

laboratory equipment to measure muscle activation patterns in the clinic, assessment of muscle activation patterns may best be performed using functional performance tests.

2.3 Functional performance tests and impairments-based validation in individuals with CAI

Functional performance tests can be used to aid in the clinical diagnosis of movement system dysfunction and identification of physical impairments in patients undergoing physical therapy.⁹⁴ It is important that functional performance tests have psychometric evidence to support their use so that clinicians can properly interpret test results and apply appropriate interventions to their patients. Psychometric evidence may include evidence for validity, reliability, and responsiveness. Validity is the ability of a test to measure what it is believed to measure and reliability is a measure of consistency or reproducibility.⁹⁵ Reliability can relate to the consistency of multiple raters to perform or interpret a test (inter-rater reliability) or to the ability of one rater to reproduce results multiple times (intra-rater reliability).⁹⁵ Responsiveness is the ability of a test to detect change.⁹⁵ Although many functional performance tests have been validated for use with patients with CAI, there is not currently a test that can be used clinically to identify altered gluteal muscle activation patterns in this population. The following section will summarize the ability of functional performance tests to identify movement system impairments in individuals with CAI.

2.3.1 Tests for instability

Instability, or the perception of the ankle giving way, is a common symptom identified among individuals with CAI and is often used to distinguish between individuals with and without CAI.^{4, 40, 41} Hop tests^{5, 41, 47-49, 96} (**Table 7**) and portions of the Star Excursion Balance Test (SEBT)^{25, 97-99} are functional performance tests that may be used to identify impaired stability. The following section describes the ability of these functional performance tests to detect and elicit instability among individuals with CAI.

Table 7. Ability of hop tests to identify impaired stability.

*: differentiates between groups if the involved limb gives way

^: differentiates between groups without the involved limb giving way

Tests that may differentiate between individuals with and without instability	Tests that may differentiate between the involved and uninvolved limbs in individuals with instability	Decreased performance may be related to presence of worse instability	Tests that may <u>not</u> be able to differentiate between individuals with/without instability
<ul style="list-style-type: none"> • 6-meter crossover hop* • multiple hop test • side hop* • square hop* 	<ul style="list-style-type: none"> • 6-meter crossover hop*^ • figure-of-8 hop*^ • lateral hop ^ • side hop*^ • square hop* 	<ul style="list-style-type: none"> • figure-of-8* • side hop* 	<ul style="list-style-type: none"> • 6-meter crossover hop • figure-of-8 • lateral hop • side hop • single limb hop for distance • triple crossover hop

A 2019 systematic review with meta-analysis by Rosen et al.⁹⁶ concluded that the multiple hop test (mean effect size: 1.399; $p < .001$), side hop test (mean effect size: -2.314 ; $p = .001$), and timed-hopping tests (mean effect size: -1.056 ; $p = .009$) were best able to differentiate

between individuals with ankle instability and those without because of their large pooled effect sizes. In this review, timed-hopping tests were grouped together for analysis.⁹⁶ Standardized, timed hop tests that have been shown to detect the impairment of instability include the 6-meter crossover hop test (mean difference: 0.93 seconds; 95% CI: 0.31 to 1.55 seconds; $p = .01$),⁴¹ the side hop test (mean difference: 1.61 seconds; 95% CI: 0.51 to 2.72 seconds; $p = .01$),⁴¹ and the square hop test (mean difference: 3.28 seconds; 95% CI: 0.51 to 6.04 seconds; $p = .02$).⁴¹ Other reports show less ability of hop tests to identify individuals with instability.^{41, 47, 48} Performance on the crossover hop,⁴⁸ the figure-of-8 hop,^{41, 48} the lateral hop,⁴⁸ the side hop,⁴⁸ single limb hop for distance,⁴⁷ and the triple crossover hop,⁴⁸ tests was reported not to be related to instability ($p > .05$). Timed hopping tests may be best for identifying the impairment of instability in individuals with suspected CAI.

The validity of hop tests in detecting instability has also been studied in light of subjects' reports of giving way during testing. The 6-meter crossover hop (mean difference, 0.96 seconds; 95% CI: 0.32 to 1.60 seconds; $p = .01$), the side hop (mean difference: 2.00 seconds; 95% CI: 0.70 to 3.31 seconds; $p = .01$), and the square hop tests (mean difference, 3.78 seconds; 95% CI: 0.76 to 6.80 seconds; $p = .02$) were all able to differentiate between subjects with instability who did and did not experience giving way during testing.⁴¹ Performance on the figure-of-8 hop test was not different between subjects with instability who did and did not report giving way during testing ($p > .05$), however.⁴¹ Performance on the 6-meter crossover hop, side hop, and the square hop test may be best for identifying instability that results in giving way during testing.

Researchers have also tested the ability of hop tests to differentiate between the involved and uninvolved limbs of individuals with ankle instability. Among individuals with CAI, the 6-

meter crossover hop ($p = .003$), the figure-of-8 hop ($p = .008$), and the lateral hop ($p = .007$), and the side hop ($p = .002$) tests have been shown to elicit a greater perception of instability when the unstable, involved limb is tested (versus the stable, uninvolved limb).⁴⁸ Another study had similar findings and reported performance deficits on the unstable limb (versus the stable limb) of subjects that had instability and reports of giving way during hop testing.⁴¹ These differences were found on the 6-meter crossover hop (mean difference: 0.42 seconds; 95% CI: 0.16 to 0.68 seconds; $p = .01$), the figure-of-8 hop (mean difference, 0.37 seconds; 95% CI: 0.01 to 0.75 seconds; $p = .05$), the side hop (mean difference, 0.57 seconds; 95% CI: 0.08 to 1.06 seconds; $p = .02$), and the square hop test (mean difference: 2.22 seconds; 95% CI: 0.34 to 4.09 seconds; $p = .02$).⁴¹ These tests may be clinically useful for detecting the impairment of instability and comparing to the uninvolved limb.

Degree of instability has been investigated using functional performance tests.^{47, 100} One study examined the relationship between performance on hop tests and the degree of instability.⁴⁷ Among individuals with unstable ankles that reported symptoms of instability during the figure-of-8 hop and side hop tests, there was a positive relationship between degree of instability and performance deficits (figure-of-8: $r = .31$, $p \leq .02$; side hop: $r = .35$, $p \leq .01$).⁴⁷ This relationship did not exist among healthy subjects.⁴⁷ Lopez-Valenciano et al.¹⁰⁰ found that subjects with worse ankle instability had worse composite YBT scores (severe instability group reach distance: 83.99% of limb length, mild instability group reach distance: 90.55% of limb length, $p = .048$). Performance on the figure-of-8 hop test, the side hop test, and the YBT may be clinically useful for judging the degree of ankle instability.

The Star Excursion Balance Test (SEBT) has also been investigated for its ability to identify individuals with impaired stability.¹⁰¹ A 2019 meta-analysis¹⁰¹ reported that the anteromedial (SEBT-AM), medial (SEBT-M), and posteromedial (SEBT-PM) reach directions had the best ability to identify individuals with unstable ankles, though the calculated effect sizes were small to moderate (SEBT-AM pooled effect size = 0.326, $p = .022$; SEBT-M pooled effect size: 0.369, $p = .006$; SEBT-PM pooled effect size: 0.406, $p = .001$). The majority of publications suggest the SEBT may not be able to differentiate between individuals with and without instability.^{25, 46, 55, 99, 102-104} However, other reports contradict this finding.^{25, 97-99, 102} When performance on the anterior and medial reach directions were studied, some investigators found no differences between stable and unstable groups (anterior reach direction^{46, 55, 102} and medial reach direction^{46, 55, 103}) while others reported that there were performance differences between the groups (anterior reach direction^{25, 97-99} and medial reach direction^{97, 98}). There is also conflicting evidence regarding the posteromedial reach direction. Most reports indicate that reach distances are not different between individuals with and without instability,^{25, 46, 55, 99, 103, 104} but two studies indicate otherwise.^{98, 102} Finally, the posterolateral^{25, 46, 55, 99, 102} and anteromedial^{46, 55, 103} reach directions were both found to be unable to differentiate between individuals with and without instability, while the posterior reach direction was able to do so.^{97, 98} Because of the conflicting evidence regarding the ability of the SEBT to detect impaired stability, it may be difficult for clinicians to rely on the SEBT to detect this physical impairment.

2.3.2 Tests for balance

Decreased balance is a commonly identified impairment in patients with ankle instability.^{5, 40, 79} Tests such as static balancing, the Star Excursion Balance Test (SEBT), the figure-of-8 hop, and the side hop test have been investigated for their validity in detecting balance impairments in this population.^{33, 50, 51, 81} Instrumented and clinical measures of balance, such as velocity of center of pressure,^{50, 81, 82} postural sway (average distance from the mean center of pressure^{50, 51}; time-to-boundary⁵⁰), number of counted errors,^{50, 52} time-in-balance⁵⁰, number of foot lifts,⁵⁰ and reach distance⁵⁰ have been used to validate functional performance tests for detecting impaired balance in this population.

Static balance tests, including bilateral, tandem, and unilateral standing balance on firm and compliant surfaces, have been studied to determine their abilities to detect impaired balance in individuals with ankle instability. While there are reports of differences in instrumented measures of eyes closed bilateral static standing balance between individuals with ankle instability and healthy controls ($p = .02$),⁸¹ it is unclear whether these differences are clinically detectable.^{52, 81} Impaired static bilateral balance may be clinically detectable during the tandem stance/foam surface portion of the Balance Error Scoring System (BESS) ($p < .001$), but bilateral standing on firm or foam surfaces was not found to detect differences in balance between individuals with ankle instability and healthy subjects ($p > .05$).⁵² Differences in the ability to maintain static unilateral balance under eyes open ($p < .01$) and eyes closed ($p < .05$) conditions have been reported among individuals with ankle instability when their performance was compared to healthy controls.⁵¹ These differences existed only when individuals with mechanical joint laxity (ligamentous laxity, measured using imaging) were compared to healthy controls.⁵¹ Differences in the ability to maintain unilateral static balance in the eyes open or eyes closed

conditions did not exist when individuals with functional (self-reported) instability were compared to healthy subjects.⁵¹ When the presence of mechanical versus functional instability was not reported, static unilateral standing balance testing alone did not detect impairments in balance in subjects with ankle instability.⁸² The BESS assesses performance on a combination of static balance tests and has been shown to detect balance impairments that occur in individuals with ankle instability ($p < .001$).⁵² Some authors have determined performance benchmarks for static balance measures that can be used to identify balance impairments in this population. The inability of an individual with ankle instability to maintain static unilateral standing balance for ≥ 25.89 seconds ($p = .006$), the occurrence of ≥ 14 total errors on the Balance Error Scoring System, or ≥ 5 foot lifts during the Foot Lift test has been reported indicate the presence of impaired balance in individuals with CAI.⁵⁰ Static balance may be clinically assessed using the BESS, as it combines several balance tests and has been validated in this population.

Dynamic balance tests such as the SEBT/YBT, the figure-of-8 hop test, the side hop test, and the multiple hop test have also been studied to determine their validity in detecting balance impairments in individuals with ankle instability. Worse performance on the SEBT has been related to impairments in balance among subjects with ankle instability.^{33, 50, 53} Linens et al.⁵⁰ determined that individuals with CAI who were unable to reach more than 91% of their limb length during the posteromedial direction of the SEBT ($p = .04$) had impaired postural stability and would benefit from balance training.⁵⁰ This group also identified cut-off times for the figure-of-8 and side hop tests that may be used to determine whether balance deficits exist.⁵⁰ Subjects with CAI that required of ≥ 17.36 seconds ($p = .03$) to complete the figure-of-8 hop test or ≥ 12.88 seconds ($p = .006$) to complete the side hop test were likely to have impaired balance.⁵⁰

The multiple hop test has also been shown to detect balance impairments that exist in individuals with ankle instability versus healthy control subjects: subjects with ankle instability perform the test with more balance errors than healthy control subjects (mean errors healthy control group: 4.2 +/- 2.2; mean errors instability group: 12.0 +/- 5.8; $p < .001$).⁸¹ Weight shifting has also been investigated for its potential to detect dynamic balance deficits but its validity was not supported.⁸¹ Dynamic balance may be clinically assessed using the SEBT, the figure-of-8 hop test, the side hop test, or by counting errors during the multiple hop test.

2.3.3 Tests for range of motion

Few functional performance tests have been investigated for their ability to detect range of motion (ROM) impairments in individuals with ankle instability. The tests that have been studied for this purpose include the SEBT and the step-down test. Gabriner et al.⁵³ investigated the contributors to performance on the SEBT and found that 16% of the variance in performance on the SEBT- anterior reach could be explained by a combination of weight bearing dorsiflexion ROM and impaired plantar cutaneous sensation.⁵³ This was considered to be a clinically-relevant model (effect size ≥ 0.15), suggesting that poor clinical performance on this portion of the SEBT may be influenced by decreased dorsiflexion ROM.⁵³ Decreased weight bearing dorsiflexion ROM has also been reported to be associated with poor movement quality during the lateral step down test.⁸³ Individuals with CAI that performed the lateral step-down test with poor movement quality had significantly less dorsiflexion ROM than those that performed the test with good movement quality (dorsiflexion ROM in good movement quality group: 42.3°, dorsiflexion ROM in poor movement quality group: 36° ; $p = .01$).⁸³ There was also a negative correlation (r

= -.39; $p = .002$) between dorsiflexion ROM and lateral step down test movement quality scores.⁸³ Some authors have recommended against the use of the step down test in individuals with ankle instability, as the decrease in dorsiflexion ROM that may be present in this population impacts performance on this test.¹⁰⁵

2.3.4 Tests for muscle activation patterns and muscle performance

Muscle activation patterns and the relationship between muscle performance and functional performance tests in both local (ankle-foot complex) muscles and in proximal muscles have also been studied in this population. Surface electromyography, musculoskeletal ultrasound, and instrumented strength testing have been used to study the validity of the SEBT, the Y-Balance Test, and the lateral hop tests for detecting impairments in muscle function. Although some of these tests have been reported to detect impairments in muscle activation and/or performance in individuals with CAI, these impairments are not always associated with clinically measurable performance deficits, limiting their clinical utility.

Impairments in ankle-foot muscle activation have been identified during the SEBT^{33, 53} and the lateral hop test.⁸⁵ Decreased activity of the tibialis anterior was found during the anterior reach direction of the SEBT when individuals with ankle instability were compared to both ankle sprain copers and healthy control subjects (CAI: 33.1% versus copers: 44.8% versus controls: 51.7%; $p < .01$).³³ This finding was associated with significantly reduced anterior reach distances in the CAI group, which may allow for clinical application of these findings (CAI group: reach distance = 82.1%; copers group reach distance = 89.1% of limb length, $p = .021$, effect size = 0.30; control group: reach distance = 90.1% of limb length, $p = .009$, effect size = 0.35).³³

Another study showed that impaired eversion strength may also contribute to performance on the SEBT.⁵³ Ankle eversion strength, combined with instrumented measures of medial-lateral postural stability, was found to explain a clinically meaningful (effect size ≥ 0.15) portion of the variance associated with performance on both the SEBT posteromedial and posterolateral reach directions.⁵³ Local muscle activity was also studied during the lateral hop test.⁸⁵ Increases in tibialis anterior and soleus muscle activation were reported during the lateral hop test when muscle activity in individuals with ankle instability was compared to healthy control subjects ($p < .05$).⁸⁵ Though these findings were posited to be associated with an increased risk of injury, these changes were not described in relation to clinically-observable movement patterns or performance deficits, limiting clinical application of these findings.⁸⁵ Fibularis longus activity was also studied during the lateral hop test but researchers did not find a difference muscle activity between subjects with ankle instability and healthy controls.⁸⁵ Impairments in tibialis anterior muscle activation patterns and eversion strength may be clinically detected using the SEBT, but alterations in local ankle-foot muscle activation patterns identified during the lateral hop test may not be clinically evident.

Differences in proximal muscle activity have been reported in individuals with ankle instability during the SEBT,³³ the Y-Balance test,¹³ and the lateral hop test.⁸⁵ Altered gluteal muscle activation patterns were identified during the posterolateral reach of the SEBT in a 2018 study.³³ This study found that individuals with CAI utilized less gluteus maximus activity than both copers and controls (CAI group: 25.6%, copers: 37.5%, controls: 40.2%; $p = 0.011$), but this difference in muscle activation was not associated with a change in reach distance ($p = .304$), limiting clinical utility.³³ Worse performance on the anterior reach of the Y-Balance test was

found to be associated with greater functional activation ratios of the gluteus maximus in individuals with ankle instability (versus healthy control subjects; mean difference of functional activation ratios = 0.08, Cohen $d = 0.57$, $p = .05$).¹³ Reach distances were also significantly different between the groups (CAI group reach distance = 65.85% of limb length, control group reach distance = 70.22% of limb length; $p \leq .05$). Impairments in rectus femoris activation have been identified during the lateral hop test: individuals with ankle instability performed the lateral hop test with greater muscle activity of the rectus femoris just before and just after initial contact ($p < .05$).⁸⁵ These findings were not associated with clinically-observable measures, again limiting the ability of clinicians to use the lateral hop test to identify muscle activation impairments. While performance on the SEBT-PL and the lateral hop test may not be able to detect impairments in proximal muscle activation patterns, poor performance on the anterior reach direction of the Y-Balance test may indicate an impairment in gluteus maximus activation patterns.

2.3.5 Summary

Impairments that may be clinically detectable via functional performance tests include impaired stability, impaired balance, decreased dorsiflexion ROM, impaired ankle muscle function, and impaired proximal muscle function (Table 8). Of the functional performance tests that have been validated for detecting proximal muscle function, only the SEBT-PL and Y-Balance Test may detect impaired gluteal muscle function. Impairments in gluteus maximus muscle activity were identified during both of these tests, but clinical application of this evidence may be difficult. The alteration in gluteus maximus activity that was identified during the SEBT-

PL was not related to test performance (reach distance), limiting the ability of clinicians to identify gluteal muscle impairments based on this test. The difference in the activation of gluteus maximus that was identified during the anterior reach direction of the Y-Balance Test was associated with decreased reach distances, allowing for clinical identification of this impairment. However, impaired reach distances on similar tests (such as the SEBT-A) have also been associated with decreased dorsiflexion ROM and decreased local muscle function. This limits the ability of clinicians to use these tests to identify gluteal muscle activation impairments, as poor performance on these tests has been reported to be related to multiple impairments. To date, there are no functional performance tests that have been validated to detect impairments in gluteal muscle function in this population without other impairments confounding results.

Table 8. Impairment-based validity of functional performance tests in CAI.

*: *findings not related to clinically measurable performance*

Stability impairments	Balance impairments	ROM impairments: decreased dorsiflexion ROM	Local muscle impairments	Proximal muscle impairments
<ul style="list-style-type: none"> • timed-hopping tests • SEBT-AM • SEBT-PM • SEBT-M 	<ul style="list-style-type: none"> • BESS • SEBT • figure-of-8 hop test • side hop test • multiple hop test 	<ul style="list-style-type: none"> • SEBT-A • step-down test 	<ul style="list-style-type: none"> • SEBT-A • SEBT-PM • SEBT-PL • lateral hop 	<ul style="list-style-type: none"> • Y-Balance Test- anterior • SEBT-PL* • lateral hop test*

2.4 Single leg squat test

The single leg squat test (SLST) is a functional performance test that has been used by clinicians and researchers to assess neuromuscular performance in patients with lower extremity musculoskeletal dysfunction.^{59, 71, 106-112} Visually-assessed performance on the SLST has been associated with differences in strength measures of the hip extensors,^{60, 86} lateral rotators,^{60, 66, 86} and abductors^{60, 67} and different activation patterns of the hip abductors⁶⁸ and adductors.^{68, 87} Because the SLST has been reported to be capable of detecting the hip muscle impairments that have been identified in individuals with CAI, the SLST may have potential for clinical use in identifying these impairments in this population. The following sections will review the literature describing the ability of the SLST to detect the proximal muscle impairments that have been identified in individuals with CAI and the test's reliability.

2.4.1 Validity for detecting neuromuscular dysfunction at the hip and relationship to deficits identified in CAI

Muscle performance

The impairments in hip muscle performance that have been reported in individuals with CAI are similar to those that the SLST has been reported to detect. These impairments include decreased muscle performance of the hip extensors, abductors, and lateral rotators.^{12, 24-27} Tri-planar hip muscle weakness has also been associated with worse performance on the SLST, both in healthy individuals and in individuals with orthopedic dysfunction.^{59-63, 66, 67} Specifically, strength deficits of the hip abductors,^{57, 59-62, 67} extensors,^{60, 66} and lateral rotators⁶³ have been identified in individuals that perform poorly on the SLST. Because of the potential for the SLST to detect the hip muscle performance impairments that may be present in individuals with CAI,

the SLST may be useful in this population. The following section will further explore the relationship between hip muscle performance deficits and SLST movement assessment.

Quality of performance on the SLST, measured using kinematics and clinical visual assessment, has been shown to be associated with hip abductor muscle performance. Increased frontal plane movement during the SLST is a common finding^{113, 114} and is used as a marker of poor movement patterns in both kinematic reports^{58, 115-117} visual rating scales.^{57, 58, 76, 118, 119} Two studies showed significant correlations between frontal plane movement and hip abductor strength in healthy individuals.^{60, 67} Stickler et al.⁶⁰ reported that, as hip abductor strength decreased, there was an increase in frontal plane movement during the SLST ($r = .466$, $p = .002$), indicating that hip abductor strength accounted for 22% of the variance in frontal plane movement in healthy females ($p = .002$).⁶⁰ Claiborne et al.⁶⁷ had similar findings in a group of male and female subjects: isokinetic peak force of the hip abductors, along with the knee flexors and extensors, was found to predict frontal plane knee motion.⁶⁷ There was also a significant negative correlation between hip abductor strength and frontal plane movement ($r = -.365$, $p < .05$).⁶⁷ Correlations between eccentric hip abduction torque and excessive frontal plane movement at the femur and knee in healthy males and females has also been reported (correlation with femur movement: $r = -.55$, $p = < .001$; correlation with knee movement: $r = .49$; $p = .004$), with increased medially-directed movement occurring with decreased abductor torque.⁶² Hip abductor strength has also been reported as a factor when overall SLST performance is visually assessed using a scale that includes tri-planar assessment of movement quality.⁵⁷ Crossley et al.⁵⁷ reported that healthy subjects whose movement quality during the

SLST was rated as “poor” had significantly less hip abductor strength than subjects whose movement quality was visually rated as “good” ($p = .016$).

The relationship between performance on the SLST and hip abductor strength has been reported in subjects seeking care for orthopedic dysfunction, as well.^{59, 61} Among individuals with hip-related groin pain, hip abductor strength was also found to be an important factor in SLST performance: subjects with less hip abduction strength performed the test with less depth than subjects with stronger hip abductors ($p \leq .01$).⁶¹ Findings were similar among individuals with patellofemoral pain syndrome (PFPS). Individuals with (PFPS) had both significantly decreased eccentric torque of the hip abductors (PFPS group torque: $0.67 \text{ Nm/kg}\cdot\text{m}$, healthy group torque: $0.81 \text{ Nm/kg}\cdot\text{m}$, $p < .05$) and increased frontal plane movement at the hip ($p < .0001$) and knee ($p < .0001$) during the SLST.⁶⁵ Lastly, among subjects who were 6 months s/p anterior cruciate ligament reconstruction, those who had less hip abductor strength performed worse on the SLST than those with better hip abductor strength ($p = .024$).⁵⁹ Isometric and eccentric measures of hip abductor torque have been related to worse performance on the SLST in healthy subjects and among individuals with musculoskeletal dysfunction.

Impaired strength of the hip extensors and lateral rotators has also been related to worse performance on the SLST. Hip extensor weakness was correlated with increased total frontal plane movement ($r = .395$, $p = .012$) and pelvic angle ($r = 0.550$, $p < 0.001$) in a 2014 study of healthy females.⁶⁰ This same study found a similar relationship between hip lateral rotator strength and frontal plane movement: as strength decreased, there was increased medially-directed frontal plane movement ($r = .464$, $p = .003$).⁶⁰ Willson et al.’s⁶⁶ 2006 study found a similar correlation between decreased lateral rotator strength and increased frontal plane

movement in a group of healthy male and female athletes ($r = .40$, $p = .004$). Further evidence of the relationship between hip strength and SLST performance was shown in a 2011 intervention study.⁶³ When hip lateral rotator and abductor strength was increased through a 6-week training program, subjects demonstrated improvement on the SLST, performing the test with less frontal plane movement at the knee and hip.⁶³ Their improvements in both SLST performance and hip strength were significantly different versus the control group that did not participate in the resistance training program.⁶³ Strength of the hip extensors and lateral rotators may be related to SLST performance.

Muscle activity

The impairments in muscle activation patterns that have been reported in individuals with CAI may be detectable with the SLST. Activation patterns of the gluteus maximus^{13, 29, 33} and gluteus medius^{11, 14, 28} have been reported to be altered during closed chain tasks among individuals with ankle instability. Similarly, quality of SLST performance has been related to altered activation patterns of gluteus maximus^{58, 68} and gluteus medius.^{57, 64, 68, 69} It is possible that the SLST may be able to detect these impairments in individuals with CAI. The following section will further explore the relationship between activation patterns of the gluteus maximus and gluteus medius and performance on the SLST.

Poor performance on the SLST has been associated with differences in activation patterns of the gluteus maximus.^{58, 68} One 2014 study of healthy females showed that subjects whose overall SLST performance was rated as “poor” utilized less gluteus maximus muscle activity than healthy females whose performance was visually rated as “good”.⁵⁸ Another group

supported these findings when they reported that altered gluteus maximus recruitment accounted for 35% of the variance in frontal plane movement in healthy females (partial $r = 0.35$, 95% CI = 0.05 to 0.59, $p = 0.04$).⁵⁸

Altered activation patterns of gluteus medius have also been associated with poor performance on the SLST.^{57, 64, 68} Crossley et al⁵⁷ found that healthy individuals that performed the SLST poorly activated anterior ($p = .007$) and posterior portions of gluteus medius ($p = .045$) significantly later than those that performed the SLST with a “good” rating. Another study found that excessive frontal plane movement at the hip and knee was correlated with increased gluteus medius activity in healthy subjects ($r = 0.62$, $p = .005$).⁶⁴ These findings were supported by a 2012 study of females with and without PFPS.⁶⁵ Nakagawa et al.⁶⁵ reported that there was a relationship between decreased gluteus medius activity and increased frontal plane movement in females with patellofemoral pain syndrome ($p = .017$).⁶⁵ Mauntel et al.⁶⁸ also studied the relationship between frontal plane movement in healthy individuals, but this study examined ratios of hip muscle activity. This group reported that individuals who performed the SLST with excessive frontal plane motion were found to have lower ratios of gluteus medius to hip adductor ($p = .028$) and gluteus maximus to hip adductor activity ($p = .007$).⁶⁸ Altered activation patterns of both gluteus maximus and gluteus medius have been reported when healthy and injured subjects perform the SLST poorly.

2.4.2 Reliability

In addition to evidence of validity, evidence of reliability of functional performance tests should be reported so that clinicians can interpret test results. Although the reliability of visual

assessment of performance of the SLST in individual with CAI has not yet been reported in the literature, there is evidence for both inter- and intra-rater reliability in other populations.

Several studies have investigated the inter-rater reliability for visual rating scales for the SLST.^{57, 61, 120-132} A 2019 systematic review with meta-analysis by Ressman et al.⁷² included nearly all of these studies in its investigation of reliability of the single leg squat test. This systematic review reported large ranges of inter-rater reliability in the literature (kappa/intraclass correlation coefficients (ICC) = 0.00–0.95). When authors pooled values reported for kappa, ICC, and first order of agreement coefficient, they found that inter-rater agreement for visual assessment of the SLST reached moderate levels (pooled agreement = 0.58, 95% CI 0.50 – 0.65) despite varying SLST protocols and grading systems used across studies.⁷² Another study that was not included in this systematic review supported the finding that evaluation of SLST performance may be associated with at least moderate levels of inter-rater reliability.¹³² McGovern et al.¹³² reported moderate to excellent inter-rater reliability (kappa = 0.933) when expert raters used a 6-point grading scale to grade SLST as “pass” or “fail” in individuals with non-arthritic hip pain. In this study, the 6 categories of assessment were associated with moderate to excellent inter-rater reliability (kappa = 0.603-0.801).¹³² The SLST has been associated with moderate inter-rater reliability.

Similar to inter-rater reliability, there is a broad range of values reported for the intra-rater reliability of visual assessment of the SLST (kappa = 0.13–1.00, ICC = 0.49 - 0.81).^{57, 61, 72, 120, 123, 127, 129, 130, 133} When Ressman et al.⁷² pooled reported values for intra-rater reliability, agreement was substantial at 0.68 (95% CI 0.60 to 0.74).⁷² Visual assessment of the single leg

squat test has been associated with acceptable inter-rater and intra-rater reliability, making it a candidate for future clinical application in individuals with CAI.

2.5 Conclusion

It is possible that the impairments in hip muscle performance and activation patterns that have been identified in laboratory studies are contributing to the inability of individuals with CAI to return to their prior level of function. Physical therapists currently do not have a clinical test to identify deficits in hip muscle activation in individuals with CAI, but functional performance tests may be capable of detecting these impairments. Hop tests, balance tests, and functional reach tests have been validated for use in this population, but none of these tests have been shown to identify hip muscle activation impairments without other deficits, such as ROM or balance, impacting results. The SLST is an easy-to-perform clinical test that clinicians and researchers use to identify hip muscle strength and gluteal muscle activation impairments in patients with lower extremity orthopedic dysfunction. The impairments that the SLST may identify in other patient populations are similar to those that have been reported to exist in individuals with CAI, making the SLST a candidate for use in a population with CAI. Additionally, a 2021 study showed that performance on the SLST was not related to ankle dorsiflexion ROM or static single leg balance, suggesting that the test may be most useful for detecting proximal impairments.⁷⁸ The SLST may be able to detect the proximal muscle activation impairments that have been reported in individuals with CAI but this must be further explored in a laboratory investigation before the test should be clinically applied to this patient population.

Chapter 3: Methods

3.1 Experimental design

A case-control study design was used to compare muscle strength and activation patterns between individuals with chronic ankle instability (CAI), copers, and healthy individuals (controls) during the single leg squat test (SLST). The independent variable was instability status. The dependent variables were (1) % MVIC activation of the gluteus maximus muscle, (2) % MVIC activation of the gluteus medius muscle, (3) maximum isometric body weight normalized torque output of the hip lateral rotators, (4) maximum isometric body weight normalized torque output of the extensors, and (5) maximum isometric body weight normalized torque output of the abductors. The primary investigator (LAC) gathered all data and performed the analysis.

3.2 Subjects

Sample size was determined using a power analysis with power = 80%, $\alpha = 0.01$, and an effect size of 0.6. This effect size was based on the results of Webster et al.'s²⁹ 2013 publication reporting an effect size of Cohen's $d = 0.71$, indicating a large effect of instability status on gluteus maximus muscle function during rotational squatting exercises. It was expected that the SLST would have required less gluteus maximus activation than the rotational squatting exercises used by Webster²⁹ and would thus be associated with a lower effect size, so $d = 0.6$ was used for the power analysis. The power analysis indicated that the study required a sample size of 45 subjects. Three additional subjects were placed in the CAI group in case of a loss of data.

Alpha was corrected to .01 from .05 to adjust for multiple comparisons on the same data.

Subjects were recruited from the Duquesne University and Shenandoah University communities.

A total of 48 subjects were included in the study.

Inclusion and exclusion criteria for all subjects were based on the 2014 guidelines from the International Ankle Consortium.¹⁶ To be considered for the study, subjects had to be: (1) between the ages of 18 and 45 years, (2) capable of performing the SLST, and (3) able to read and communicate in English. Exclusion criteria for all subjects included¹⁶: (1) history of musculoskeletal surgery in either lower extremity, (2) history of fracture in either lower extremity that required realignment, (3) injury to the musculoskeletal structures of either lower extremity in the previous 3 months that resulted in limited physical activity for 1 or more days, (4) the presence of any active medical diagnosis that limits or prohibits participation in physical activity. If volunteers satisfied the general inclusion criteria, group assignment was then determined.

To be included in the CAI group, subjects had to have at least 1 significant lateral ankle sprain that occurred ≥ 12 months prior to enrollment in the study.¹⁶ A significant ankle sprain was defined as one that was associated with pain and/or swelling and resulted in the loss of at least 1 day of desired physical activity.¹⁶ Subjects in the CAI group also had a history of recurrent sprains, giving way, or the perception of instability of the ankle, and had to score > 11 on the Identification of Functional Ankle Instability (IdFAI) outcome measure.¹⁶ These subjects also had to demonstrate a loss of function by scoring $< 90\%$ on the Foot and Ankle Ability Measure- activities of daily living subscale (FAAM-ADL) and $< 80\%$ on the Foot and Ankle Ability Measure- sports subscale (FAAM-sports).¹⁶ Grouping criteria are depicted in Table 4.

To be included in the copers group, subjects had to have at least 1 significant lateral ankle sprain¹⁶ that occurred ≥ 12 months prior to enrollment in the study.⁷⁴ Copers did not have recurrent sprains, giving way, or the perception of instability⁷⁴ and scored ≤ 10 on the IdFAI. Members of the copers group had returned to at least moderate levels of weight bearing physical activity for ≥ 12 months following the index sprain and reported little to no functional disability⁷⁴ (FAAM-ADL and FAAM-sports $\geq 95\%$).

Healthy control subjects were individuals who had never had a significant ankle sprain,¹⁶ did not experience giving way or instability of the ankle and reported little to no functional disability (FAAM-ADL and FAAM-sports $\geq 95\%$).

3.3 Instrumentation

Muscle activity was measured with the Noraxon Desktop DTS surface electromyography system (Noraxon USA Inc, Scottsdale, AZ) (Figure 1). The myoMUSCLE module of the myoRESEARCH software platform, version 3.16 (Noraxon USA Inc., Scottsdale, AZ), was used for data acquisition and processing. Electrodes were disposable, self-adhesive HEX dual snap, pre-gelled, silver chloride electrodes with 2.0cm of fixed inter-electrode distance (Noraxon USA Inc., Scottsdale, AZ) (Figure 2). Kinematic data were gathered using inertial measurement units (Opal wearable sensors, APDM Inc., Portland, Oregon) (Figure 3), a sync box (Figure 4) (APDM Inc., Portland Oregon), and Moveo Explorer software (APDM Inc., Portland, Oregon). Maximum force output was measured using a MicroFET handheld dynamometer (Hoggan Scientific, Salt Lake City, UT) (Figure 5). Body weight and height were measured using a Health o meter Professional scale (Pelstar LLC/Health o meter, McCook, IL) (Figure 6).

Figure 1. The Noraxon Desktop DTS (Noraxon USA, Scottsdale, AZ) surface electromyography (sEMG) system was used to gather muscle activity data.

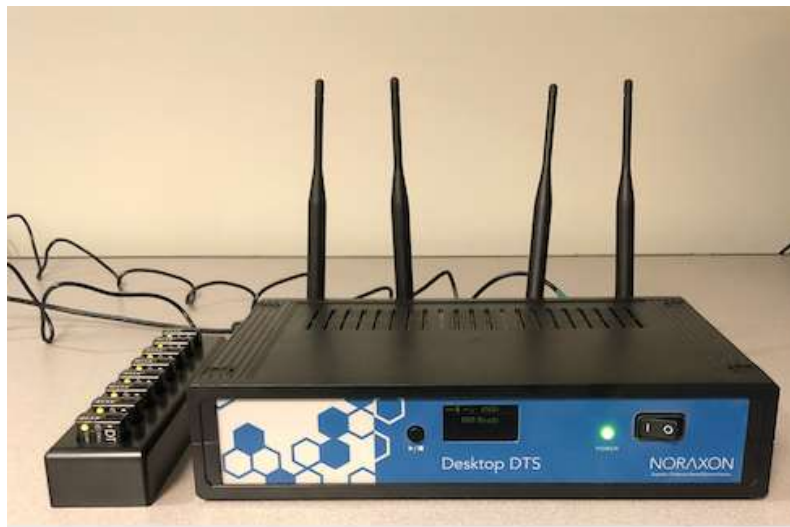


Figure 2. Electrodes were disposable, self-adhesive HEX dual snap, pre-gelled, silver chloride electrodes with 2.0cm of fixed inter-electrode distance (Noraxon USA Inc., Scottsdale, AZ).



Figure 3. Wearable inertial measurement unit (Opal wearable sensors, APDM Inc., Portland, Oregon).



Figure 4. A sync box (APDM Inc., Portland Oregon) was used to synchronize sEMG and kinematic data to determine muscle activity during the SLST.



Figure 5. The Hoggan Scientific MicroFET 2 handheld dynamometer (Hoggan Scientific, Salt Lake City, UT) was used to measure maximum force output of the hip lateral rotators, extensors, and abductors.



Figure 6. Body weight and height were measured using a Health o meter Professional scale (Pelstar LLC/Health o meter, McCook, IL).



3.4 Procedures

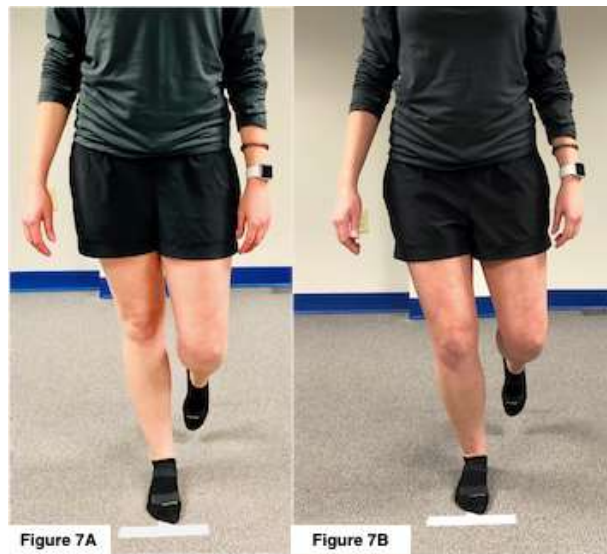
All procedures, save for the order of strength testing, were identical for each subject. Following informed consent, subjects completed the IdFAI, FAAM-ADL, and FAAM-sports to determine group assignment. The IdFAI is a valid measure of ankle instability that is associated with an overall test-retest reliability of 0.92.¹³⁴ The FAAM-ADL is a valid measure of general physical function for individuals with foot and ankle dysfunction and has been associated with test-retest reliability of 0.89.¹³⁵ It is responsive to change (minimal detectable change of +/- 5.7 points) and its minimal clinically important difference is 8 points.¹³⁵ The FAAM-sports subscale is a validated measure of sports function in individuals with foot and ankle diagnoses.¹³⁵ It has been associated with test-retest reliability of 0.87, a minimal detectable change of +/- 12.3 points, and minimal clinically important difference of 9 points.¹³⁵ Both the FAAM-ADL and FAAM-sports subscales have been validated for use in individuals with CAI.¹³⁶ If subjects did not meet criteria for group assignment based on their scores on the IdFAI, the FAAM-ADL, and/or the FAAM-sports, they did not participate in the study. Following group assignment, subjects reported their age and sex, and completed the Tegner Activity Scale. The Tegner Activity Scale is a reliable (test-retest reliability: intraclass correlation coefficient [ICC] = 0.8), responsive (minimal detectable change = 1) measure of participation in physical activity.¹³⁷

After subjects completed the above self-report measures, anthropometric data was gathered. Height and body weight were measured using a Health o meter Professional scale (Pelstar LLC/Health o meter, McCook, IL). Subjects then performed a 5-minute warm up on a stationary bicycle (Monark Ergomedic 828 E, Monark Exercise, Vansbro, Sweden) at a self-selected pace.⁶⁸ After the warm up, dominant extremity was determined using a previously

published protocol in which the dominant extremity was that which the subject chose for a unilateral drop jump landing in at least 3 of 5 trials.¹³⁸ Femur length (distance from the center of the greater trochanter to the most distal aspect of the lateral femoral epicondyle) and tibia length (distance from the medial knee joint line to the most distal aspect of the medial malleolus) were measured in centimeters.²⁵

Next, subjects were instructed in the evidence-based SLST protocol.⁷⁶ In this protocol, a “T” (6” horizontal, 10” vertical) was taped on the floor using 1.5” wide athletic tape. The subject stood on the vertical tape on their barefoot test extremity and flexed the other knee to 90° (Figure 7A). They were then instructed to squat until they could no longer see the horizontal tape in front of their toes (Figure 7B) and then return to the starting position. A smartphone metronome application (The Metronome by Soundbrenner for iOS) set to 60 beats per minute was used to maintain a rate of 1 squat every 2 seconds. Subjects were permitted to practice the SLST protocol until they felt comfortable with the instructions.

Figure 7 A-B. Performance of the single leg squat test (SLST). A. Starting position. B. Squat position.



Testing for strength and maximal muscle activity was then performed. Isometric strength of the hip lateral rotators, extensors, and abductors was performed in a randomized order for each subject. The order of strength testing was determined using a list randomizer (random.org, Randomness and Integrity Services Ltd, Dublin, Ireland). All strength testing trials lasted 5 seconds and consisted of 3 test trials. For each trial, subjects were instructed to increase the intensity of their muscle contraction during the first 3 seconds of the test and then give maximum effort for the 4th and 5th seconds.²⁵ Subjects were given a 30-second rest interval after each trial.²⁵ To test hip lateral rotator strength, subjects were in short sitting at the edge of a plinth with a stabilizing strap placed across their thighs and a towel roll between their thighs.²⁵ The dynamometer was placed 2 inches (5.08cm) proximal to the most distal aspect of the medial malleolus, as seen in Figure 8.²⁵ Because maximum voluntary isometric contraction (MVIC) activation data was gathered during strength testing of the hip extensors and abductors, the

surface electromyography (sEMG) electrodes were placed on the skin prior to strength testing of these muscle groups. The skin overlying gluteus maximus and gluteus medius was cleaned with isopropyl alcohol and electrodes were applied per SENIAM guidelines.⁹³ For the gluteus maximus, the electrode was placed halfway between the second sacral vertebrae and the greater trochanter.²⁹ The electrode was oriented parallel to the line between the posterior superior iliac spine and the middle of the posterior aspect of the thigh.²⁹ For gluteus medius, the electrode was placed overlying the muscle belly, between the iliac crest and the greater trochanter, about 1 inch below the iliac crest.¹³⁹ This electrode was oriented parallel to the muscle fibers.¹³⁹ Proper labeling of the electrode channels was confirmed by tapping the electrodes and observing increased activity on the correct channel on the computer monitor. Strength and MVIC activation of the hip extensors was measured in prone with the knee flexed to 90 degrees. The dynamometer was placed 2 inches (5.08cm) proximal to the distal aspect of the lateral femoral epicondyle and stabilizing straps were placed across the posterior superior iliac spine and the dynamometer, as in Figure 9.²⁵ Electrode placement over gluteus maximus was confirmed by observing real-time muscle activity on the computer screen during MVIC testing.⁶⁸ For strength and MVIC activation testing of the hip abductors, the patient was positioned in sidelying (test side up) with pillows between their lower extremities to maintain neutral hip abduction and adduction. The dynamometer was placed 2 inches (5.08cm) from the distal aspect of the lateral femoral epicondyle and stabilization straps were placed over the dynamometer and the iliac crest, as seen in Figure 10.²⁵ Electrode placement over the gluteus medius was confirmed by observing real-time muscle activity on the computer screen during MVIC testing.⁶⁸ The reliability of using MVIC activation to measure the amount of gluteal muscle activity has been reported to be

excellent (gluteus maximus reliability: ICC = 0.94 – 0.98,¹⁴⁰ gluteus medius reliability: ICC = 0.93¹³⁹). After strength and MVIC activation testing, 4 inertial measurement units (Opal wearable sensors, APDM Inc., Portland Oregon) were applied to the subject's waist, thigh, leg, and foot (Figure 11). Subjects then performed the SLST while muscle activity data for the gluteus maximus and gluteus medius were recorded. After subjects performed 5 repetitions of the SLST, data collection was complete.

Figure 8. Testing position for the hip lateral rotators.

Figure 9. Testing position for the hip extensors.

Figure 10. Testing position for the hip abductors.



Figure 11. Placement of inertial measurement units on subject.



3.5 Data reduction and analysis

Processing of the surface electromyographic data (both MVIC and SLST trial) was performed using the myoMUSCLE module of the myoRESEARCH software platform, version 3.16 (Noraxon USA Inc., Scottsdale, AZ). Lowpass (450 Hz⁶⁴) and highpass filters (20 Hz⁶⁴) were applied and the signal was smoothed using a root mean square (RMS) algorithm with a 125ms time constant.⁶⁴ The data were extracted by normalizing the signal amplitude to MVIC activation. MVIC activation was established by using the highest consecutive half-second period that occurred over the 3 MVIC trials.⁶⁴

Maximum isometric body weight normalized torque (Nm/kg) was calculated for the hip lateral rotators, extensors, and abductors using the maximum force output measured over the 3

MVIC trials (Newtons), multiplied by the moment arm (meters), and divided by body mass (kilograms). The moment arm for the hip extensors and abductors was femur length (converted from centimeters to meters) minus .0508m and the moment arm for the lateral rotators was tibia length (converted from centimeters to meters) minus .0508m.²⁵

Muscle activity data for the gluteus maximus and gluteus medius were analyzed for the second, third, and fourth squat repetitions. The timing of each squat was defined using kinematic data from the inertial measurement units (IMUs). The beginning of the second squat was defined as the time point when the knee on the stance extremity reached maximal extension after the first squat. The end of the second was defined as the point when the knee again reached maximal extension after the second squat was completed. Continuing this pattern, maximal knee extension was used as a marker for the start of one repetition and the end of the previous repetition. The amount of muscle activity occurring during each squat was represented using percent MVIC (% MVIC). % MVIC represents the amount of muscle activation occurring during the SLST, relative to the maximal amount of muscle activity that was measured during MVIC activation testing. The mean % MVIC that was measured during the second, third, and fourth repetitions of the SLST was reported and used for analysis.⁶⁸

3.6 Statistical analysis

Data analysis was completed using SPSS version 26 (IBM Corporation, Armonk, NY). Data were examined for outliers and missing data. Quantitative variables were summarized using means and standard deviations or medians and interquartile ranges and qualitative variables were summarized using frequency counts. Normality of continuous variables was assessed using the

Kolmogorov-Smirnov test. Levene's test was used to determine homogeneity of variance of continuous variables. Statistical assumptions were met for the variables gluteus maximus % MVIC, gluteus medius % MVIC, lateral rotation strength, and abduction strength. The assumption of homogeneity of variance was violated for extension strength and the demographic variables IdFAI, FAAM-ADL, and FAAM-sports. The assumption of normality was violated for all demographic variables (age, BMI, Tegner Activity Scale score, IdFAI score, FAAM-ADL score, FAAM-sports score). Research questions 1, 2, 3, and 5 were analyzed using separate one-way ANOVAs and Tukey's HSD tests were used for post-hoc analyses. Research question 4 and quantitative demographic variables (age, Tegner Activity Scale score, IdFAI score, FAAM-ADL score, and FAAM-sports score) were analyzed using a Kruskal-Wallis one-way analysis of variance (ANOVA) by ranks. Post-hoc testing was performed using 3 separate Mann-Whitney U tests. Chi square tests were performed to analyze the distribution of dominant extremity, test extremity, and sex between the groups. Because 5 separate research questions were asked and 5 statistical tests were performed on the same data set, the significance level value for the 5 research questions was corrected from 0.05 to 0.01 ($0.05 / 5 = 0.01$) *a priori*. The significance level for the post-hoc tests was also corrected to account for multiple comparisons and was set at 0.0033 ($0.01 / 3 = 0.0033$).

Chapter 4: Results

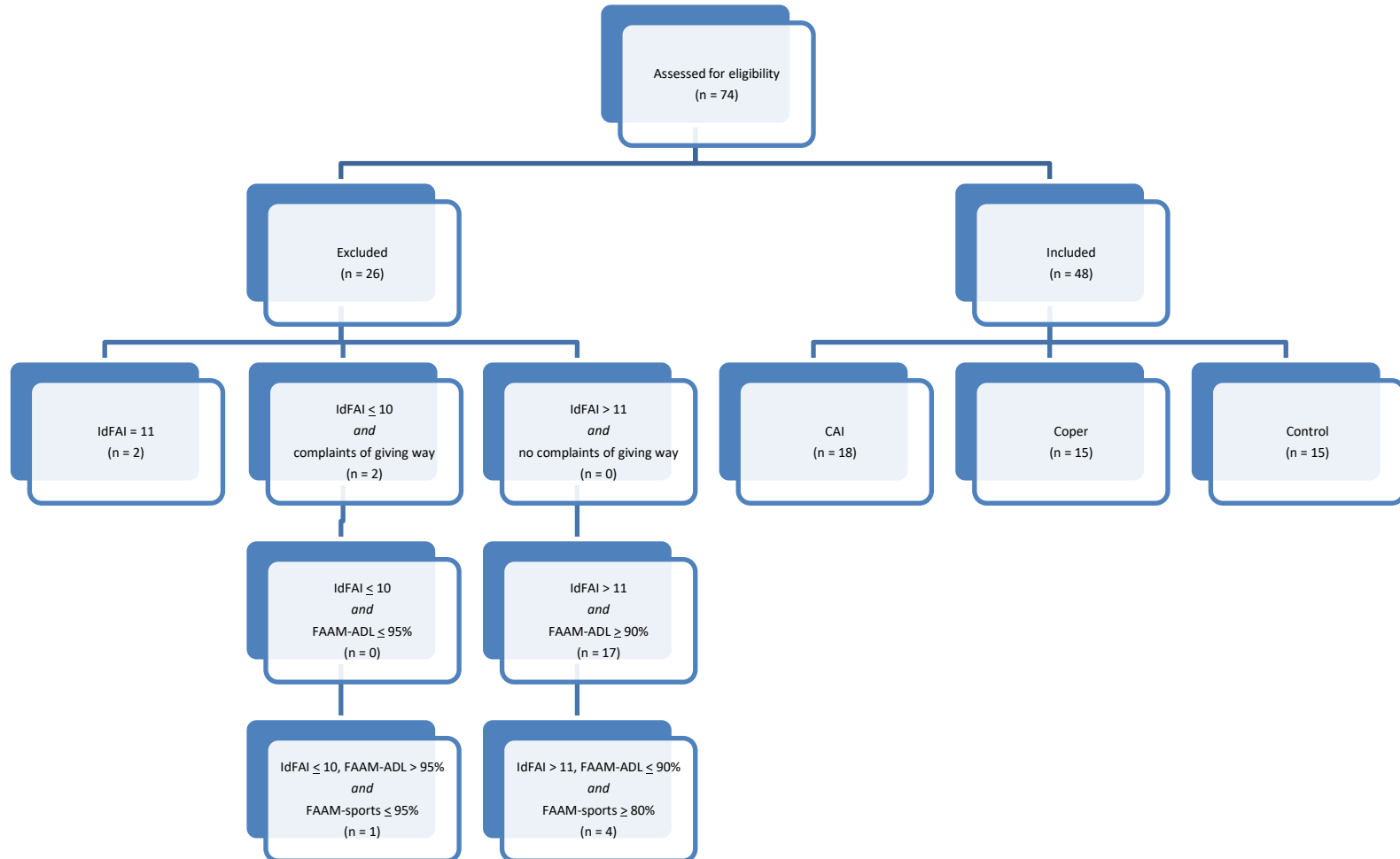
4.1 Subjects

Seventy-four individuals met the general inclusion criteria for the study. Of those, 48 participants were enrolled in the study. Eighteen subjects were included in the CAI group, 15 in the coper group, and 15 in the control group. The 26 individuals that met the general inclusion criteria but were not enrolled in the study were not enrolled because they did not meet the criteria for assignment to the chronic ankle instability (CAI), coper, or control group (Table 9). Of these, there were 21 individuals that scored > 11 on the Identification of Functional Ankle Instability tool (IdFAI) but did not score low enough on the Foot and Ankle Ability Measure- activities of daily living subscale (FAAM-ADL) or the Foot and Ankle Ability Measure- sports subscale (FAAM-sports) to be included in the CAI group. Seventeen of these individuals were excluded because they scored $> 90\%$ on the FAAM-ADL and 4 were excluded because they scored $> 80\%$ on the FAAM-sports. Two individuals were unable to be included in the study because they scored ≤ 10 on the IdFAI but reported episodic giving way of the ankle. One individual was excluded because they scored ≤ 10 on the IdFAI, $> 95\%$ on the FAAM-ADL, but $< 95\%$ on the FAAM-sports. Two individuals were excluded because they scored 11 on the IdFAI. A flow diagram of the subjects enrolled in the study is shown in Figure 13.

Table 9. Criteria for assignment to chronic ankle instability (CAI), coper, and control groups.

Criteria	CAI	Coper	Control
History of sprain*	≥ 1 year prior	≥ 1 year prior	Never
Reports of episodic giving way	Yes	No	No
Instability score*	IdFAI > 11	IdFAI ≤ 10	IdFAI ≤ 10
Functional score*	FAAM- ADL < 90% AND FAAM- sports < 80%	FAAM- ADL > 95% AND FAAM- sports > 95%	FAAM- ADL > 95% AND FAAM- sports > 95%

Figure 12. Flow diagram of subjects enrolled in the study



Data were collected on 48 subjects. Table 10 shows median age, height, weight, body mass index (BMI), Tegner Activity Scale score, IdFAI score, FAAM-ADL score, and FAAM-sports score for the entire sample and for the CAI, coper, and control groups. These variables were analyzed using separate Kruskal-Wallis one-way analyses of variance by ranks due to violations of the assumption of normality for all variables and violations of the assumption of homogeneity of variance for the variables IdFAI, FAAM-ADL, and FAAM-sports. There were no differences between the groups for the variables age ($p = .833$), height ($p = .921$), weight ($p = .824$), BMI ($p = .865$), or Tegner Activity Scale score ($p = .186$) between the groups. There was a significant difference between the groups on the variables of IdFAI ($p < .001$), FAAM-ADL ($p < .001$), and FAAM-sports ($p < .001$). Post-hoc comparisons using independent Mann-Whitney U tests showed that there were statistically significant differences between all 3 groups at $\alpha = .0033$ (CAI vs coper: $p < .001$, CAI vs control: $p < .001$, coper vs control: $p < .001$) on the IdFAI variable. On the variables of FAAM-ADL and FAAM-sports, the CAI group was significantly different from the coper ($p < .001$) and control groups ($p < .001$), but there was no significant difference between the coper and control groups (FAAM-ADL: $p = .345$, FAAM-sports: $p = .217$). Table 11 shows the distribution of sexes, dominant extremity, and test extremity for the groups. Twenty-nine percent of the sample was male, and males made up 50% of the CAI group, 29% of the coper group, and 29% of the control group. Seventy-one percent of the sample was right-limb dominant (56% of the CAI group, 87% of the coper group, 73% of the control group). The dominant limb was tested in 58% of the total sample, 56% of the CAI group, 87% of the coper group, and 73% of the control group. A Chi square statistic showed that there was no

difference between the groups on the variables of sex ($p = .886$), dominant extremity ($p = .142$), or test extremity (dominant versus non-dominant, $p = .172$).

Table 10. Median and interquartile range for age, height, weight, BMI, Tegner Activity Scale score, IdFAI, FAAM-ADL, FAAM-sports scores.

Data were statistically compared using a Kruskal-Wallis one-way analysis of variance by ranks.

Key:

*: $p < .05$ when compared to coper group

^: $p < .05$ when compared to control group

	CAI (n = 18)	Coper (n = 15)	Control (n = 15)	Total (n = 48)
Age (years)	22.00 (1.25)	22.00 (1.00)	22.00 (1.00)	22.00 (1.00)
Height (m)	1.689 (.20)	1.676 (0.15)	1.676 (0.13)	1.68 (.15)
Weight (kg)	63.05 (22.68)	70.31 (20.41)	68.76 (18.14)	68.04 (19.84)
BMI (kg/m²)	23.79 (2.85)	23.73 (3.66)	24.45 (4.93)	24.41 (3.70)
Tegner Activity Scale score (0-10 points)	6.00 (1.25)	5.00 (4.00)	5.00 (1.00)	5.00 (1.00)
IdFAI score (0 – 37 points)	20.00 (6.25)*^	5.00 (2.00)^	1.00 (2.00)*	5.00 (15.75)
FAAM-ADL score (0 – 100%)	88.10 (2.68)*^	100.00 (1.19)	100.00 (0.00)	100.00 (11.61)
FAAM-sports score (0 – 100%)*	75.00 (10.71)*^	100.00 (3.57)	100.00 (0.00)	100.00 (39.29)

Table 11. Sex, dominant extremity, and test extremity for CAI, coper, and control groups.

There were no statistically significant differences between the groups on these variables ($p > .05$).

	CAI (n = 18)	Coper (n = 15)	Control (n = 15)	Total (n = 48)
Sex (male/female)	6/12	4/11	4/11	14/34
Dominant extremity (right/left)	10/8	13/2	11/4	34/14
Test extremity (dominant/non-dominant)	13/5	9/6	6/9	28/20

4.2 Statistical results

All statistical assumptions were met for all research questions except for research question 4. For research questions 1, 2, 3, and 5, data were analyzed using separate one-way analyses of variance. Because the assumption for homogeneity of variance was not met for research question 4, a Kruskal-Wallis one-way analysis of variance by ranks was used to analyze this data. Table 12 shows the mean and standard deviation for the muscle activation variables. Results of strength testing are shown in Table 13.

Table 12. Mean and standard deviation (SD) for percent maximum voluntary isometric contraction (% MVIC) of gluteus maximus and gluteus medius utilized during the single leg squat test for each group.

Key

**: indicates statistically significant difference versus coper group at $p = .01$*

^: indicates statistically significant difference versus control group at $p = .01$

	CAI (n = 18)		Coper (n = 15)		Control (n = 15)	
	Mean	SD	Mean	SD	Mean	SD
Gluteus maximus (% MVIC)	36.03*^	10.85	18.30	10.39	21.04	8.14
Gluteus medius (% MVIC)	31.29	10.34	24.76	11.16	26.49	7.97

Table 13. Mean and standard deviation (SD) for maximum isometric body weight normalized torque output for each group.

Key

†: Indicates that a non-parametric test was performed to analyze this variable. Median and interquartile range are reported in place of mean and SD (respectively).

**: indicates statistically significant difference versus coper group at $p = .0033$*

^: indicates statistically significant difference versus control group at $p = .0033$

	CAI (n = 18)		Coper (n = 15)		Control (n = 15)	
	Mean	SD	Mean	SD	Mean	SD
Hip lateral rotators (Nm/kg)	0.58*^	0.13	0.67	0.09	0.76	0.09
Hip extensors (Nm/kg)†	1.10^	0.15	1.38	0.41	1.65	0.37
Hip abductors (Nm/kg)	1.43	0.22	1.47	0.18	1.65	0.27

Question 1: Is there a difference in the percent of maximum voluntary isometric contraction (% MVIC) of gluteus maximus utilized during the SLST by individuals with CAI and without CAI?

Hypothesis: There will be a difference in the amount of gluteus maximus activation utilized by individuals with CAI and without CAI during the SLST.

There was a statistically significant difference in % MVIC activation of the gluteus maximus between the groups ($p < .001$). Effect size was large with a partial $\eta^2 = .229$. Post hoc testing using a Tukey's HSD and $\alpha = .0033$ showed that there were significant differences between the CAI group and the coper group ($p < .001$) and between the CAI group and the control group ($p < .001$). The CAI group utilized a significantly higher % MVIC activation (36.03%) for gluteus maximus during the SLST than both the coper (18.30%) and control (21.04%) groups. There was no difference between the coper and control group ($p = .733$) on this variable.

Question 2: Is there a difference in the % MVIC activation of gluteus medius utilized during the SLST by individuals with CAI and without CAI?

Hypothesis: There will be a difference in the amount of gluteus medius activation utilized by individuals with CAI and without CAI during the SLST.

There was not a statistically significant difference in % MVIC activation of the gluteus medius between the groups ($p = .155$).

Question 3: Is there a difference in the amount of body weight normalized torque generated during a maximum voluntary isometric contraction of the hip lateral rotators between individuals with and without CAI?

Hypothesis: There will be a difference in the amount of body weight normalized torque generated by the hip lateral rotators during a maximum isometric contraction between individuals with CAI and without CAI.

There was a significant difference between the groups when maximum isometric body weight normalized torque output of the hip lateral rotators was compared ($p < .001$). Effect size was large with a partial $\eta^2 = .549$. Post-hoc analysis was completed with a Tukey's HSD and $\alpha = .0033$, which revealed that there were differences between the CAI and coper groups ($p = .001$), between the CAI and control group ($p < .001$). There was not a significant difference between the coper and control group ($p = .006$). The CAI group was significantly weaker than both the coper and control groups (maximum isometric bodyweight normalized torque CAI group = 0.582 Nm/Kg, coper group = 0.677 Nm/kg, control group = 0.757 Nm/kg).

Question 4: Is there a difference in the amount of body weight normalized torque generated during a maximum voluntary isometric contraction of the hip extensors between individuals with and without CAI?

Hypothesis: There will be a difference in the amount of body weight normalized torque generated by the hip extensors during a maximum isometric contraction between individuals with CAI and without CAI.

There was a significant difference between the groups when maximum isometric body weight normalized torque of the hip extensors was compared ($p = .001$). A post hoc analysis was performed using separate Mann-Whitney U tests and $\alpha = .0033$. This analysis showed that there were significant differences between the CAI group and the control group ($p < .001$), but no significant difference between the CAI group and the coper group ($p = .005$) or between the coper and control group ($p = .202$). The CAI group was significantly weaker than the control group (bodyweight normalized torque CAI group = 1.10 Nm/Kg, bodyweight normalized torque control group = 1.65 Nm/kg).

Question 5: Is there a difference in the amount of body weight normalized torque generated during a maximum voluntary isometric contraction of the hip abductors between individuals with and without CAI?

Hypothesis: There will be a difference in isometric, body weight normalized torque of the hip abductors between individuals with CAI and without CAI.

There was not a statistically significant difference between the groups when maximum isometric body weight normalized torque of the hip abductors was compared using $\alpha = .01$ ($p = .02$).

Reliability

Reliability for force output data was calculated using intraclass correlation coefficient model 3, 3 (ICC(3, 3)). Results showed excellent reliability for lateral rotation (ICC (3,3) = 0.985, standard error of measurement (SEM) = 0.016), extension (ICC (3,3) = 0.966, SEM =

0068), and abduction (ICC (3,3) = 0.962, SEM = 0.047) testing. Within-session reliability for surface electromyography data was calculated using a Pearson correlation coefficient. The correlation between the mean % MVIC during the second and third squat repetitions and the mean % MVIC for the fourth and fifth squat repetitions were strong for both the gluteus maximus ($r = .941, p < .001$) and gluteus medius ($r = .968, p < .001$).

Chapter 5: Discussion and conclusions

5.1 Introduction

The purpose of this study was (1) to determine whether the single leg squat test (SLST) could be used to detect differences in proximal muscle activation between individuals with and without CAI and (2) to determine if there was a difference in maximum isometric strength of the hip lateral rotators, extensors, and abductors between individuals with and without CAI. The group with CAI was hypothesized to demonstrate a different amount of muscle activation of the gluteus maximus and gluteus medius muscles during the SLST when compared to individuals without ankle instability. The CAI group was also expected to demonstrate decreased strength of the hip lateral rotators, extensors, and abductors compared to the groups without ankle instability (copers and controls). The results of the study supported the hypotheses that the CAI group would demonstrate a different amount of percent of maximal voluntary isometric contraction (% MVIC) activation of gluteus maximus during the SLST and the hypotheses that the CAI group would demonstrate decreased hip lateral rotator and extensor strength. The results of the study did not support the hypothesis that the CAI group would demonstrate a different amount of % MVIC activation of gluteus medius or the hypothesis that the CAI group would demonstrate decreased strength of the hip abductors.

This discussion will provide further analysis of the results of the study. Specifically, this section will compare the results of this study to previously published work and will discuss a potential mechanism that may explain these findings. The clinical significance of these results, as

well as limitations of this study, will also be detailed. Finally, this section will suggest future work that may be pursued given the results of this study.

5.2 Key findings

The main finding of this study was that the group with CAI demonstrated differences in proximal neuromuscular function when compared with the groups without CAI (copers and controls). Specifically, the 3 key findings of this study included: (1) increased % MVIC activation of gluteus maximus during the SLST, (2) decreased maximum isometric body weight normalized torque of the hip lateral rotators, and (3) decreased maximum isometric body weight normalized torque of the hip extensors. The following sections will discuss the results of the study in order of research question, with an emphasis on these 3 key findings.

5.2.1 Key findings: muscle activation

Gluteus maximus

The first of 3 key findings of this study was that there were differences in gluteus maximus activation between individuals with and without CAI. The CAI group demonstrated significantly more gluteus maximus muscle activation (36.03% MVIC) during the SLST than both the coper (18.30% MVIC; $p < .001$) and control (21.04% MVIC; $p < .001$) groups. The difference between groups on this variable was associated with a large¹⁴¹ effect size (partial $\eta^2 = .229$), indicating that instability status had a large effect on gluteus maximus muscle activation during the SLST.

The finding that individuals with CAI utilized increased gluteus maximus muscle activation during closed kinetic chain tasks has been shown previously in the literature during a variety of tasks.^{13, 34, 142, 143} In 2016, Webster et al.³⁴ compared gluteus maximus activity in individuals with CAI and healthy controls during a lateral hop. Results showed that the CAI group performed the lateral hop with an increase in gluteus maximus activation (CAI group: 45.55%, control group: 36.81%) just before landing from the lateral hop. Increased gluteus maximus activation has also been shown to occur during the Y-balance test (YBT).¹³ DeJong and colleagues¹³ measured gluteus maximus activation during the YBT using musculoskeletal ultrasound imaging. Musculoskeletal ultrasound quantifies muscle activation based on a ratio of thickness of the muscle at rest to thickness of the muscle during a contraction. This group found that, during the anterior reach of the YBT, subjects with CAI demonstrated significantly more gluteus maximus thickening (versus healthy controls), indicating an increase in muscle activation versus the control subjects.¹³ An increase in gluteus maximus muscle activation was also shown in 2021 by Lin et al.¹⁴² who measured gluteus maximus activation in individuals with CAI and healthy controls during treadmill walking. This study found that the CAI group responded to perturbations during treadmill walking with an increase in activation versus healthy controls.¹⁴² Increased gluteus maximus activation has also been reported during a jumping and cutting maneuver.¹⁴³ Kim et al.¹⁴³ reported an increase in gluteus maximus muscle activation in a group with CAI (versus healthy controls) when they performed a single leg jump landing immediately followed by a lateral cutting maneuver. This increase in activation occurred just before the initial unilateral landing on the involved limb.¹⁴³ Each of these studies examined activation of gluteus maximus on the unstable limb and found an increase in gluteus maximus muscle activation in the

CAI group when compared to a healthy control group. There are also published reports showing that individuals with CAI utilized less gluteus maximus muscle activation when compared to subjects without CAI.^{29, 33} Decreased gluteus maximus activity (versus healthy controls) has been reported when individuals with CAI perform a rotational squat²⁹ and during the posterolateral reach direction of the Star Excursion Balance Test (SEBT).³³ The reports of significant differences in gluteus maximus muscle activation in the CAI groups suggest that there are proximal neuromuscular changes occurring in individuals with CAI. The finding that some tasks are associated with an increase in activation in the CAI groups and some tasks are associated with a decrease in muscle activation in the CAI group may suggest that the direction of change in gluteus maximus muscle activation is task specific.

This study was unique in that it included a copers group. The presence of the copers group in this study may offer unique insight into factors that differentiate copers and individuals that develop long-term functional limitations and instability after a lateral ankle sprain. In this study, the copers group performed the SLST with gluteus maximus activation that was significantly less than that used by the CAI group but was not significantly different ($p = .733$) than the control group. This result suggests that there may be a relationship between gluteus maximus muscle activation during the SLST and functional status, as the copers and control groups had significantly higher Foot and Ankle Ability Measures- activities of daily living subscale (FAAM-ADL) and Foot and Ankle Ability Measures- sports subscale (FAAM-sports) scores. Unfortunately, the ability to corroborate these findings with previously published work is limited, as few authors have studied the amount of gluteus maximus muscle activation between all 3 of these groups. This hypothesis may be supported by Jaber et al.'s³³ work. Jaber and

colleagues³³ included individuals with CAI, copers, and controls in their study of muscle activation during the Star Excursion Balance Test (SEBT). This group reported that the copers group's gluteus maximus activation was significantly different from that of the CAI group but was not different from the control group. Though this is not conclusive evidence that proximal neuromuscular status is related to function, both our findings and Jaber et al.'s³³ results may support this suggestion. Additional research into the differences between individuals with CAI, copers, and healthy controls may help to clarify the relationship between functional status after a lateral ankle sprain and proximal muscle activation.

The reasons for findings of increased gluteus maximus activation in individuals with CAI are unclear and cannot be determined based on the results of this study. Authors have speculated that differences in muscle activation in individuals with CAI may be attributable to altered movement strategies utilized by this group during closed kinetic chain tasks.^{13, 103, 144, 145} Changes in movement patterns may result in altered demand on gluteus maximus, possibly increasing the amount of muscle activation required for a given task. For example, individuals with CAI have been reported to perform unilateral squatting tasks with increased trunk flexion.^{103, 145} Increased trunk flexion may shift the body's center of mass anteriorly, increasing the torque demand on the hip extensors, such as the gluteus maximus. Increased torque demand on the gluteus maximus could contribute to higher muscle activation measured by sEMG, as muscle force production and sEMG activation are believed to be grossly linearly related.¹⁴⁶ Kinematics and kinetics were not examined in this study, however, so this hypothesis cannot be directly supported with the results of this study. Another proposed rationale for this increase in gluteus maximus activity during closed kinetic chain tasks is that this may be a compensatory "bracing" of the proximal muscles

and transition to a more hip-focused balance strategy in response to distal instability.^{13, 30, 34, 147} Rios et al.³⁰ proposed this after finding that individuals with CAI utilized a higher ratio of proximal to distal muscle activation (versus healthy controls) on the unstable stance extremity during a ball kicking task. Though this may be a potential mechanism contributing to the findings of this study, this hypothesis also cannot be directly supported based on this study's results.

Gluteus medius

This study found that there was no difference between the groups when gluteus medius muscle activation during the SLST was analyzed ($p = .155$). Although there was no statistical difference between the groups on this variable, the CAI group did demonstrate higher mean gluteus medius activation than the coper or control groups (CAI group: 31.29% MVIC, coper group: 24.76% MVIC, control group: 26.49% MVIC). This pattern of the CAI group utilizing more hip muscle activation than the coper or control groups was consistent with the results that were observed when gluteus maximus muscle activation was analyzed.

The lack of significant findings on this variable could be due to the SLST protocol utilized in the study. It is possible that the demand placed on the gluteus medius during the SLST was insufficient to allow us to identify differences in activation between the groups. Though it is unknown what level of muscle activation is needed for researchers to identify differences between groups with and without CAI, it is possible that a higher level of activation than was demanded by the SLST would be needed. An examination of previous work may provide some insight. A single leg squat protocol that was similar to the one used in this analysis was studied

by Ayotte et al.¹⁴⁸ and was found to elicit 36% MVIC from gluteus medius in a group of healthy controls. It is possible that this moderate¹⁴⁹ level of activation contributed to a ceiling effect on our test: because of relatively low demands on the gluteus medius, impaired gluteus medius activation may not have been detected with this test. This notion of a possible ceiling effect for the SLST in detecting gluteus medius impairments may be supported by further examining our results in light of the report by Ayotte and colleagues.¹⁴⁸ Ayotte et al.¹⁴⁸ found that the single leg squat produced 57% MVIC from gluteus maximus and our results identified a significant difference between the groups when gluteus maximus muscle activation was compared. This may suggest that a task that requires a higher level of muscle activation may be better able to detect muscle activation impairments. Tasks that have been shown to be associated with a higher level of muscle activation demand, such as a full-depth single leg squat¹⁵⁰ or a unilateral stance task with movement of the contralateral limb,¹⁵¹ may be needed in order to identify gluteus medius activation differences in this population.

Other factors that may have contributed to these results include sEMG cross-talk and statistical power. Given the position of the gluteus medius deep to the gluteus maximus posteriorly and deep to the tensor fascia lata anteriorly, it has been suggested that cross-talk from gluteus maximus¹⁴ and tensor fascia lata^{152, 153} may limit the validity of sEMG measurements of gluteus medius muscle activation. It is possible that this may have contributed to the results of this study, though efforts to minimize cross-talk were taken. These efforts included use of appropriately-sized electrodes and careful placement of electrodes per SENIAM guidelines.⁹³ Statistical power also could have impacted the lack of significant finding on this variable, as this study was not powered *a priori* to answer this research question. A post-hoc analysis of effect

size and statistical power indicated that instability status was associated with a medium¹⁴¹ effect (partial $\eta^2 = .079$) on gluteus medius mean activation and that statistical power was low for this research question (power = 0.185). This may indicate that the study was underpowered for this research question and that there may have been a Type II error.

The findings that there were no differences in the amount of gluteus medius activation during a closed kinetic chain task are similar to findings reported in 2013²⁹ and 2016.³⁴ In 2013, Webster et al.²⁹ found that, when compared to healthy controls, individuals with CAI did not utilize different amounts of gluteus medius activation during a single leg rotational squat or during a rotational lunge. Three years later, Webster et al.³⁴ reported that there was no difference in gluteus medius activation between healthy individuals and individuals with CAI when a lateral hop was performed. Activation during the lateral hop was analyzed both before and after subjects participated in a fatigue protocol and no difference was found under either condition.³⁴ Researchers that identified a difference in the amount of gluteus medius activation examined activation during treadmill walking.^{11, 14, 28} It is possible that the reported changes in gluteus medius activation are task-specific, are related to timing of activation^{33, 142} rather than amount of activation, or that these findings are not consistent in individuals with CAI.

5.2.2 Key findings: muscle strength

Lateral rotators

The second key finding of this study was that individuals with CAI produced less maximum isometric body weight normalized torque of the hip lateral rotators than both copers and controls ($p < .001$). This measure served as an indicator of strength and supports the

hypothesis that the CAI group would have weaker hip lateral rotators than individuals without CAI. Mean maximum isometric body weight normalized torque production for the CAI group's hip lateral rotators was found to be 0.560 Nm/kg, while the copers group produced 0.667 Nm/kg, and the control group produced 0.757 Nm/kg. There was a significant difference on this variable when the CAI and copers groups ($p = .001$) and the CAI and control ($p < .001$) groups were compared. There was not a significant difference between the copers and control groups ($p = .006$) on the variable, but the difference between the groups on this measure (difference = 0.09 Nm/kg) did exceed the minimal detectable change (MDC) for this measure (MDC = 0.044 Nm/kg), suggesting that there may be a clinical difference between the copers and control groups. There was a large¹⁴¹ effect size (partial $\eta^2 = .549$) associated with the difference between groups on this variable, indicating a large effect of ankle instability status on hip lateral rotator strength.

Decreased strength of the hip lateral rotators in individuals with CAI has been previously reported in the literature in 3 separate studies by McCann et al.²⁴⁻²⁶ In 2017, McCann et al.²⁵ reported that individuals with CAI had significantly lower hip lateral rotation strength when compared to lateral ankle sprain copers and controls. This finding was supported in 2018²⁴ and 2019²⁶ when McCann and colleagues again measured maximum isometric body weight normalized torque production and found a difference between individuals with CAI, copers, and controls. Though there have been few studies examining hip lateral rotation strength in individuals with and without CAI, all that were identified in this search and those reported in a 2020 systematic review¹² supported the finding that there is a difference between groups on this variable. The presence of decreased strength of the hip lateral rotators in individuals with and without CAI appears to be consistently reported in the literature.

The muscle weakness observed in the current study for the CAI group may be due to disuse. It has been reported that individuals with CAI are less physically active than individuals without CAI.⁸ This decrease in activity could be related to the reports that individuals with CAI demonstrate decreased strength, but this does not explain the pattern of strength deficits seen in the current study. Our results indicate that the CAI group had less strength of the hip lateral rotators and extensors. It is possible individuals with CAI avoid performing tasks that place demand on the hip lateral rotators and extensors, thus contributing to weakness from relative disuse. Neumann¹⁵⁴ suggested that the hip lateral rotators are biomechanically responsible for performing tasks that require rotation of the trunk and pelvis over a fixed limb that is in single limb stance. Functionally, this group of muscles would be most active when an individual plants their limb and cuts laterally during running.¹⁵⁴ Perhaps individuals with CAI avoid performing lateral maneuvers and this activity avoidance contributes to decreased strength of these muscles. Though it is unknown if individuals with CAI avoid these movements to an extent that would contribute to decreased strength, nearly all of the individuals with CAI that participated in our study reported difficulty performing lateral cutting on the FAAM-sports. Thirteen subjects reported either extreme or moderate difficulty with this task, 4 subjects reported slight difficulty, and only 1 subject reported no difficulty when performing cutting and lateral maneuvers. Conversely, all but one subject in the coper and control groups reported no difficulty with this task. These differences in activity-specific impairments may be related to strength differences in the groups, but more study is needed to make a conclusive statement.

Central nervous system (CNS) changes following the index sprain could also be impacting the CAI group's ability to activate the hip lateral rotators and may explain the

observed strength changes. Individuals with CAI have been shown to require more stimulation of the motor cortex to contract peripheral musculature.¹⁵⁵ This may contribute to decreased muscle strength, as individuals with CAI may not regularly engage these muscles in the way that the copers and control groups do. This remains conjecture, however, as this conclusion cannot be drawn from the present study and this relationship has not been specifically studied in the hip lateral rotators in individuals with CAI. The proposed mechanism behind CNS changes following peripheral joint injury will be further explored in section 5.3.

The current study's finding that individuals with CAI had decreased strength of the hip lateral rotators may be related to this study's other key findings. Our key findings were that individuals with CAI performed the SLST with more gluteus maximus activation and that individuals with CAI had strength deficits of the hip lateral rotators and extensors. Gluteus maximus is a hip lateral rotator and extensor¹⁵⁶ and therefore may be involved in all 3 of these key findings. Clinically, it is possible that the CAI group's decreased strength of the hip extensors and lateral rotators may be related to the increase in gluteus maximus muscle activation during the SLST, as individuals with weakness may have needed to increase their muscle activation to perform the SLST. This hypothesized relationship cannot be directly supported with the results of this study, however, as the relationship between maximum isometric strength and % MVIC activation during a submaximal activity was not studied. Understanding the potential relationship between maximum strength and muscle activation during a submaximal task may require examining parameters other than normalized activation, such as latency, fatigability, or non-normalized signals.

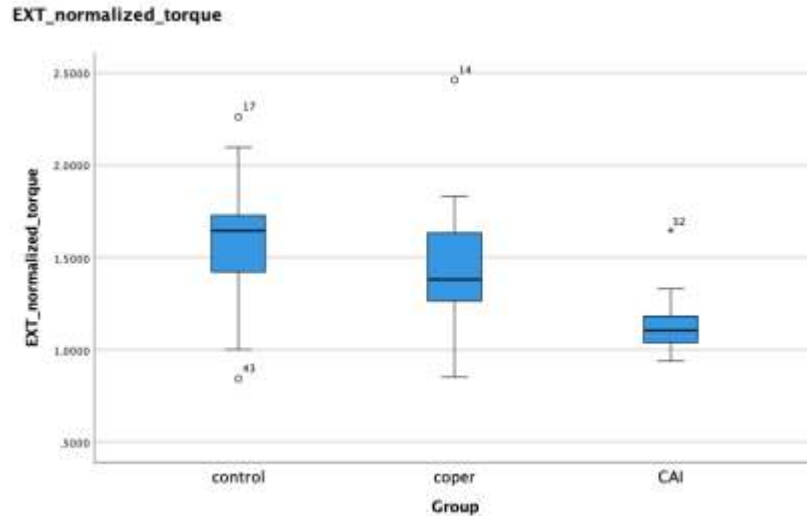
Extensors

The third key finding of this study was that the CAI group demonstrated less maximum isometric body weight normalized torque of the hip extensors ($p = .001$). This supported the hypothesis that the CAI group would demonstrate decreased strength of the hip extensors when compared to individuals without CAI. The CAI group was significantly weaker than the control group (maximum isometric body weight normalized torque CAI group = 1.10 Nm/Kg, control group = 1.65 Nm/kg; $p < .001$) but was not significantly weaker than the copers group (maximum isometric body weight normalized torque copers group = 1.38 Nm/kg; $p = .005$). There were no differences in hip extensor strength between the copers and control groups ($p = .202$). Similar to the results for the hip lateral rotators, the control group had the highest maximum isometric body weight normalized torque, the copers group had the next-highest output, and the CAI group had the lowest torque output. The difference between groups on this variable was associated with a large¹⁴¹ effect size (partial $\eta^2 = .270$), indicating a large effect of instability status on hip extensor strength.

This variable was analyzed using a Kruskal-Wallis one-way analysis of variance (ANOVA) by ranks because the statistical assumption of homogeneity of variance was violated. This statistical assumption relates to the spread of scores for the dependent variable at each level of the independent variable of instability status. For this research question, homogeneity of variance was examined by comparing the spread of the scores for hip extension strength between the CAI, copers, and control groups using Levene's test and visual analysis of box plots (Figure 13). Levene's test ($p = .034$) resulted in a failure to reject the statistical null hypothesis, indicating a difference between the groups on variance of hip extension strength scores. A visual

examination of box plots for the groups may offer insight into spread of the data for this variable. Box plots show the distribution of data into quartiles. The blue-shaded box in each plot shows the middle half of measurements (interquartile range) and the horizontal line inside the box indicates the median score.¹⁵⁷ The vertical lines extending from the top and bottom of the boxes indicate where the upper and lower quarter of observations fell.¹⁵⁷ Box height can be used as an indicator of variance,¹⁵⁷ thus comparing box height between the groups shows the variance for each groups on this variable. Figure 13 shows that the box height for the CAI group is less than that for the copers and control groups, indicating that the CAI group had less variance on the hip extension strength variable. This discrepancy in variance could be attributed to the more stringent requirements for inclusion in the CAI group versus the copers and control groups. The similarities of the CAI group on instability and functional status may have contributed to this group's lower variance on the measure of hip extensor strength while the more broad group assignment criteria for the copers and control groups may have been associated with more variance on this measure. This remains conjecture, however, as the study design does not allow us to directly determine this. To account for heterogeneity of variance observed for this variable, the Kruskal-Wallis one-way ANOVA by ranks was performed and it did show that there was a statistically significant difference between the groups on this variable ($p = .001$).

Figure 13. Boxplots showing variance of hip extensor body weight normalized torque for the control, copers, and CAI groups.



The post-hoc analysis utilized a conservative $\alpha = .0033$ to correct for multiple comparisons. This resulted in failure to reject the statistical null hypothesis that there was not a difference between the CAI and copers group on the variable of hip extension strength at $p = .005$. Despite the statistical conclusion that there was not a difference between the groups on this variable, data trends ($p = .005$) suggested that a clinically meaningful difference may exist between the CAI and copers groups. Additionally, the difference between the CAI group and copers group's body weight normalized torque outputs (difference = 0.28 Nm/kg) exceeded the MDC for this variable (MDC = 0.188), further suggesting that there may be a clinical difference in hip extension strength between the CAI and control groups. This is supported by a 2018 publication by McCann et al.²⁴ who reported that individuals with CAI demonstrated less isometric maximum body weight normalized torque output than lateral ankle sprain copers.

Other published work indicates that there may not be a difference between individuals with CAI and copers on this variable, however. Kosik and colleagues¹⁵⁸ and a 2019 investigation by McCann et al.²⁶ both reported that there was not a significant difference between CAI and coper groups in hip extension strength. Each of these reports did demonstrate a pattern of findings that was similar to this study's findings that the control group was the strongest, the CAI group was the weakest, and the coper group fell between the other groups.

Results from previous publications are mixed when hip extension strength is analyzed in individuals with and without CAI. McCann and colleagues^{24, 26} twice reported a that there was a difference in maximum isometric bodyweight normalized torque when individuals with and without CAI were compared. Kosik et al.¹⁵⁸ also found a significant difference on hip extensor strength between individuals with and without CAI. This study included both young (aged 18 – 40 years) and middle-aged (aged 41 – 71 years) and found that, regardless of age, hip extension strength was decreased in the group with CAI when compared to the group without CAI. Further evidence that hip extension strength may be related to ankle instability was shown in a 2017 prospective study of risk factors for lateral ankle sprain in youth soccer players.¹⁵⁹ This study reported that decreased hip extension strength was an independent risk factor for ankle sprains in this population.¹⁵⁹ Although this does not offer evidence that directly supports our results, it may support the notion that there is a relationship between hip extension strength and ankle instability. Conflicting reports have also been published. A 2021 report by Bain and colleagues¹⁶⁰ studied isometric hip extension strength in individuals with and without CAI. This group reported that there was no difference between the groups on this measure.¹⁶⁰ One key distinction between our study and Bain et al.'s¹⁶⁰ study was that our group required that subjects

demonstrate functional impairment (FAAM-ADL < 90%, FAAM-sports < 80%) to be included in the CAI group, while Bain and colleagues did not do this. Bain's¹⁶⁰ CAI group did have lower median FAAM-ADL scores than the non-CAI groups (median FAAM-ADL score CAI group = 95.2, median FAAM-ADL score copers/control groups = 100.00), but these differences did not exceed the published MCID for the FAAM-ADL of 8 points.¹³⁵ This may indicate that Bain et al.'s¹⁶⁰ CAI and non-CAI groups may have been functionally similar. These results suggest that the presence of foot and ankle-related functional limitations may be involved in the relationship between hip extension strength and ankle instability.

The reason for this study's results that the CAI group had weaker hip extensors than the copers and controls may be related to the proposed mechanism explaining weakness of the hip lateral rotators. This study's hip extensor strength test was identical to Kendall's¹⁶¹ gluteus maximus strength test, suggesting that our strength results were reflective of gluteus maximus torque output. Gluteus maximus is both a hip lateral rotator and extensor¹⁵⁶ so our results that both the hip lateral rotators and extensors were weaker in the CAI group may be related and attributed to similar etiology.

Abductors

The final research hypothesis in this investigation was that there would be a difference between individuals with and without CAI when hip abductor strength was evaluated. This hypothesis was not supported by the study. The CAI group's mean maximum isometric body weight normalized torque was lower than both the copers/control groups (body weight normalized torque CAI group = 1.43 Nm/Kg, copers/control group = 1.47 Nm/kg, control group = 1.65). This

difference was not statistically significant at $\alpha = .01$ and $p = .02$. The difference between maximum isometric body weight normalized torque of the hip abductors between the CAI and control groups did exceed the MDC for this variable (MDC = 0.13 Nm/kg, difference between CAI and control group = 0.22), suggesting that a clinical difference in hip abductor strength may have existed. Effect size was large¹⁴¹ (partial $\eta^2 = .159$) for these variables, indicating that, despite the failure to reject the null hypothesis for this research question, there was a large effect of instability status on hip abductor strength. Observed power was 0.500. It was determined that 81 subjects would have been required to reach power = 0.80 given the observed effect size, $\alpha = .01$ (G*Power 3.1¹⁶²).

These findings are similar to previously-published reports by Bain et al.¹⁶⁰ and McCann et al.²⁴ Both groups found that there was no difference in maximum isometric body weight normalized torque between individuals with and without CAI.^{24, 160} McCann and colleagues²⁴ analysis did not report effect size but did report $p = .07$ for this research question. This was just outside of their cut off for significance ($\alpha = .05$), which was similar to our findings ($p = .02$, $\alpha = .01$). Though these findings were not statistically significant, these results do not preclude the potential presence of a clinical difference in hip abduction strength between these groups. Evidence supporting that there is a difference in hip abduction strength between individuals with and without CAI was published in a 2017 report by McCann et al.²⁵ and a 2007 report by Hubbard et al.¹⁶³ McCann et al.²⁵ showed that there was a difference in maximum isometric bodyweight normalized torque of the hip abductors when this variable was compared between individuals with and without CAI²⁵. This difference existed between the CAI and coper groups and between the CAI and control groups.²⁵ Hubbard et al.¹⁶³ reported that there was a difference

in hip abduction peak torque between individuals with and without CAI. Given the body of evidence and this study's result on this variable, it remains unclear whether a meaningful difference exists between these groups on hip abduction strength.

5.2.3 Key findings: summary

The key findings of this study were that (1) the CAI group performed the SLST with more activation of the gluteus maximus than the groups without CAI, (2) the CAI group had less strength of the hip lateral rotators than both the coper and control group, and (3) the CAI group had less strength of the hip extensors than the control group. These findings may be evidence of a central neuromuscular change in the CAI group.^{164, 165} A potential mechanism for these changes will be explored in the following section.

5.3 Potential mechanism for neuromuscular changes in CAI

Several authors have proposed that the proximal changes that have been reported in individuals with CAI may be due to changes in the neuromuscular system occurring after the index sprain.^{11-14, 24-26, 29, 34, 164-171} Muscle activation and strength differences, such as those that were observed in our study, are considered to be evidence of this change in CNS output.¹⁶⁴ The mechanism behind this change is believed to begin with damage to peripheral mechanoreceptors which alters afferent input sent to the central nervous system (CNS).¹⁶⁴ Changes in afferent input are suspected to contribute to CNS reorganization and changes in CNS output, such as altered muscle activation and decreased strength.¹⁶⁴ Evidence of components of this pathway as it has

been reported in individuals with ankle instability^{155, 172-174} and anterior cruciate ligament (ACL) injuries¹⁷⁵ will be described in the following section.

The mechanism behind neuroplasticity following peripheral joint injury is suspected to begin with physical or chemical damage to mechanoreceptors found in and around the peripheral joint.^{164, 167, 175-177} When intact, mechanoreceptors are believed to supply the CNS with information about joint position and joint movement based on tissue strain.^{164, 178} After a joint injury such as an ACL tear¹⁷⁸ or ankle sprain,¹⁷² the mechanoreceptors appear to provide less afferent information to the CNS.^{172, 178} A decrease in afferent input to the CNS is believed to contribute to changes in motor output, as motor output is modified based on afferent information.¹⁶⁴ This is important because prolonged alterations in motor output may contribute to maladaptive neuroplasticity, such as changes in muscle activation and strength.¹⁶⁴

CNS re-organization in individuals with ankle instability has been studied by comparing motor thresholds in injured and uninjured individuals.^{155, 173} Motor thresholds are a measure of how much brain stimulation is required to elicit a muscle contraction and these serve as an indicator of the relationship between the brain and motor output.¹⁶⁴ A higher motor threshold indicates that more brain stimulation is needed to elicit the same sEMG-measured muscle contraction.¹⁶⁴ Pietrosimone et al.¹⁷³ reported that individuals with CAI had significantly higher motor thresholds for fibularis longus contraction than subjects without CAI. Higher motor thresholds for fibularis longus were again reported in 2015 by McLeod et al.¹⁵⁵ These reports^{155, 173} may provide some evidence of CNS re-organization for what is believed to be a key muscle in ankle stability,¹⁷⁹ but there is limited evidence for this when proximal muscles are studied.^{155, 174} In the same study that found changes in motor threshold for fibularis longus, McLeod et al.¹⁵⁵

found no evidence of changes in motor threshold for vastus medialis. A pilot study published in 2021 examined motor threshold of gluteus maximus in individuals with CAI, lateral ankle sprain copers, and controls.¹⁷⁴ This study found that there were no differences on motor thresholds between the groups with and without CAI.¹⁷⁴ The copers group did demonstrate an increased motor threshold for gluteus maximus when compared to the control group, however.¹⁷⁴ This finding lead authors to hypothesize that the copers may have developed a beneficial CNS adaptation following lateral ankle sprain.¹⁷⁴ It should be noted that this study's sample size was limited to 30 participants and results may or may not be clinically meaningful or relevant to the neuromuscular changes occurring at the hip in this population.¹⁷⁴ Though this route of neuroplasticity is frequently suggested as a pathway for the neuromuscular changes that have been observed in individuals with CAI,^{11, 13, 14, 24-26, 28, 29, 34, 90, 164, 167, 171} direct evidence for this phenomenon is limited.

5.4 Clinical significance

The results of this study support the previous work showing that there are neuromuscular changes at the hip in individuals with CAI. These changes included increased gluteus maximus muscle activation during the SLST and decreased strength of the hip lateral rotators and extensors. These results may be clinically meaningful for physical therapists that are managing patients with CAI. Our finding that individuals with CAI performed the SLST with increased gluteus maximus muscle activation suggests that physical therapists may be able to use the SLST to assess gluteus maximus activation in their patients with CAI. Further study is needed before implementing this, however, as this study did not determine whether these differences could be

detected clinically. Our finding that the CAI group had less strength of the hip lateral rotators and extensors than the groups without CAI supports the recommendation from the 2021 Academy of Orthopedic Physical Therapy clinical practice guideline for ankle instability⁷⁹ that physical therapists assess hip strength in this population.

5.5 Limitations

There were several limitations to this study. Potential limitations that should be considered when interpreting the findings of this study will be discussed in the following sections. Factors that decreased the trustworthiness of our experimental findings, including factors related to the subjects being studied, factors related to the researcher, and factors related to the procedures will be discussed as threats to internal validity. Factors that limit the generalizability of our results beyond the study sample will be discussed as threats to external validity.

5.5.1 Threats to internal validity

Threats to internal validity include factors related to the subjects being studied, factors related to the researcher, and factors related to the experimental procedures. Each of the following factors could have impacted the study's ability to determine if there was a relationship between proximal neuromuscular activation, strength, and instability status.

5.5.1.1 Factors related to subjects

One of the major threats to the internal validity in this study included the sampling strategy. This study used a convenience sample of volunteers who were primarily health sciences

students at two Universities. It is possible that these individuals were not representative of the population and that the relationship that was identified between instability status and proximal neuromuscular status could be attributed to a confounding factor. To minimize this possibility, this study measured and compared factors that were suspected to play a role in strength and performance on the SLST, applied exclusion criteria to limit the introduction of known confounders, and utilized published criteria for group assignment. Factors that were suspected to play a role in performance on the SLST and in hip muscle strength such as age, sex, height, weight, body mass index (BMI), and level of activity were compared between the groups. There were no statistical differences between the groups on these factors, which decreased the likelihood that these subject attributes influenced our results. Other factors that had potential to play a role in the relationship between the independent and dependent variables included history of lower extremity surgery and the presence of a current or recent activity-limiting lower extremity injury. Both of these factors were exclusion criteria for participation in the study and were therefore unlikely to be valid threats to internal validity. The use of published guidelines^{16, 74} for group assignment was also intended to decrease threats to internal validity related to subject attributes. These guidelines^{16, 74} utilized valid and reliable instruments that objectively measure ankle instability^{134, 180} and foot and ankle functional ability^{135, 136} to assign subjects to groups. The use of statistical comparisons between the groups on factors that were suspected to be involved in the relationship between hip muscle function and ankle instability and the application of careful selection criteria were used to decrease threats to internal validity that could have been related to subject attributes.

5.5.1.2 Factors related to the researcher

The primary researcher performed subject recruitment, group assignment, data collection, data processing, and data analysis. This introduced the possibility of researcher bias to our study based on grouping criteria. Because the primary researcher was responsible for determining a volunteer's eligibility for the study and assigning the volunteer to a group, it was impossible for the researcher to be blinded to a subject's group assignment during data collection, processing, and analysis. To minimize the researcher's ability to recall a subject's group assignment during data collection, screening of the subjects to determine whether they met inclusion criteria and could be assigned to a group took place at least 1 day prior to the subject presenting for participation in the study. To further minimize the chance of researcher bias, the researcher performed processing of the electromyographic data and data analysis using subject identification numbers that were not linked to group assignment. Though this does not eliminate the possibility that the researcher was aware of a subject's group assignment and processed the data in light of that knowledge, this practice was implemented to improve the trustworthiness of the study's results given the responsibilities of the primary researcher.

Another threat to internal validity included the researcher's skills in data collection. Proof of the primary researcher's ability to collect data precisely and consistently is supported by their clinical experience and demonstrated reliability in the study's measures. The primary researcher was a physical therapist with 11 years of experience, a manual therapy certification, and a board certification in orthopedics. Reliability of the researcher's strength testing was excellent ($ICC(3,3) = .962 - .985$).

5.5.1.3 Factors related to the procedures

Factors related to the experimental procedures that could be considered threats to internal validity included the SLST itself and the order of maximum voluntary isometric contraction (MVIC) testing. There are many variations of the SLST and performance on the SLST has, at times, been associated with ankle dorsiflexion range of motion (ROM) impairments.^{68, 181, 182} Because individuals with ankle instability are likely to present with impaired ankle dorsiflexion ROM,⁷⁹ this study utilized a SLST protocol that was previously shown not to be influenced by factors such as ankle dorsiflexion ROM and foot posture.⁷⁸ The present study did not measure ankle dorsiflexion, however, so the influence of ankle dorsiflexion ROM on SLST performance cannot be conclusively eliminated. Another aspect of the experimental design that could have influenced results was the order of maximum voluntary isometric contraction (MVIC) and SLST testing. MVIC testing had to take place prior to SLST performance so that the sEMG data could be normalized to each subject's MVIC activation. It is possible that performing strength testing prior to the SLST could have influenced subjects' performance on the SLST. All subjects in the study completed MVIC testing prior to performing the SLST, however, so it is unlikely that this impacted the groups in an unequal manner. Another factor related to the order of MVIC testing is the order in which strength of the lateral rotators, extensors, and abductors was tested. Performing these tests in the same order for each subject could have introduced systematic error in the validity of the strength measurements. For example, if muscles were tested in the same order for each subject, fatigue could decrease maximum isometric output for the last muscle that was tested, resulting in lower-than-actual strength measures for this muscle. This study randomized the order of strength testing in order to minimize this potential threat to validity.

The instrumentation, including the handheld dynamometer and the sEMG system, used in the study also could have threatened internal validity. The handheld dynamometer that used to measure force output was calibrated by the manufacturer and was said to be accurate within +/- 2%, minimizing this as a threat to internal validity. The sEMG system could have also been a threat to internal validity in this study. Key factors that could have influenced the quality of the sEMG signal include impedance, noise, and individual subject variations.¹⁴⁶ Impedance, including skin-electrode impedance and input impedance, can influence how well the motor unit action potentials are received and recorded by the system.¹⁴⁶ Skin-electrode impedance is affected by skin preparation and electrode selection.¹⁴⁶ It is recommended by the manufacturer of our sEMG equipment that skin-electrode impedance be < 30 kOhms.¹⁸³ To minimize skin-electrode impedance and its related threat to internal validity, this study used gelled electrodes, which are associated with reduced impedance.¹⁸⁴ This study also checked skin-electrode impedance for each subject using the myoMUSCLE software (myoMUSCLE version 3.16, Noraxon USA, Scottsdale, AZ) and ensured that it was < 30 kOhm prior to performing any testing. Input impedance is a characteristic of the sEMG amplifier and it is recommended that the input impedance be at least 1 - 10 MegaOhm.¹⁸⁵ The sEMG unit (Desktop DTS, Noraxon USA, Scottsdale, AZ) used in this study has an input impedance of >10 MegaOhm, further helping to minimize this threat to internal validity. Noise is another factor that could have impacted our ability to measure muscle activation. Major sources of noise include noise related to the sEMG equipment, ambient noise, and noise from signal artifact.¹⁴⁶ Because the same sEMG equipment was used for all subjects, noise related to the sEMG equipment was unlikely to be a substantial threat to internal validity in this study. Ambient noise is noise caused by environmental

electromagnetic radiation such as fluorescent lighting and electrical cables.¹⁴⁶ This type of noise is considered to be nearly impossible to avoid.¹⁴⁶ Because this study was performed in two separate University laboratories, it was possible that the ambient noise was variable and impacted our results. To decrease this threat to internal validity, the researcher checked baseline noise and found it to be < 5 microvolts for each subject prior to beginning testing.¹⁸³ Motion artifact, which is noise occurring at 0-20Hz,¹⁸⁴ could have also been a source of noise that could have threatened internal validity. This noise was minimized by following recommendations to use double-sided tape to secure the components to the skin¹⁸⁴ and by applying a high-pass filter at 20Hz during data processing. Factors related to the ability of the instrumentation to accurately measure the individuals participating in the study could have also served as a threat to internal validity. An individual subject's subcutaneous fat, muscle geometry, and unintended differences in the recording set up could have impacted our ability to answer the research question.¹⁴⁶ These differences were accounted for in data processing by normalizing sEMG output to each individual's maximum voluntary isometric contraction activation. This allowed for comparison between subjects despite variations in a subject's anthropometrics or sEMG set-up.¹⁴⁶ Pearson correlation coefficients were calculated to measure the reliability of the sEMG system in detecting muscle activation during the SLST. The calculated correlation coefficients demonstrated a strong, positive relationship between mean muscle activation occurring during the second and third squat repetitions and that which occurred during the fourth and fifth squat repetitions ($r = .941 - .968$). Though there were several possible threats to internal validity related to the instrumentation used in this study, these threats were minimized through the use of calibrated, high-quality equipment and the implementation of signal checks, signal

normalization, and the measurement of the system's reliability in measuring muscle activation during the SLST.

5.5.2 Threats to external validity

External validity relates to the ability of the research to be generalized to a population outside of the study sample. Threats to external validity in this study primarily included factors related to the study sample. The age and physical activity level of the sample should be taken into account when results are generalized. The sample used in this study had a median age of 22 years, so application of these results to subjects that are much younger or older may be inappropriate. The subjects used in the sample were generally active individuals, with Tegner Activity Scale scores ranging from 3 - 10 and a median score of 5 for the entire sample. Application of these results to subjects that score < 3 on the Tegner Activity Scale may not be valid, as activity level may play a role in the relationship between muscle activation, muscle strength, and instability status. Additionally, although the subjects in the study's CAI group reported impaired function on both the FAAM-ADL and the FAAM-sports, they were not currently seeking care for their CAI. This may limit the ability of physical therapists to generalize these results to their patients with CAI, as the study sample may not be reflective of patients seeking care.

5.6 Suggestions for future work

This study provided evidence that individuals with CAI may use different activation patterns of the gluteus maximus muscle during the SLST. However, this study did not relate this

increase in muscle activation to a clinical measure, such as visual observation, which limits the immediate clinical application of these findings. Future research should seek to determine whether clinicians can reliably use visual observation of the SLST to identify impairments in gluteus maximus muscle activation. Future research should also include measurement and evaluation of the kinetics and kinematics during the SLST in this group so that the biomechanical factors contributing to SLST performance can be further understood. Finally, future research should investigate whether an intervention program aimed at improving gluteus maximus function and SLST performance is effective at improving function in this population.

This study also provided evidence that individuals with CAI had weaker hip lateral rotators and extensors than individuals without CAI. Future work should attempt to determine if a hip strengthening program could improve instability and function in this group. There is preliminary evidence that a rehabilitation program improving hip strength can improve functional outcomes.⁹² In 2018, Smith and colleagues⁹² reported that a 4-week hip strengthening program could improve both strength and functional outcomes in individuals with CAI. Further investigation into optimal exercise prescription, including a study of mode, frequency, intensity, and duration of exercise, to address hip strength deficits in this population should be pursued.

Finally, future research should include long-term studies that provide insight into the development of CAI. Although several studies have investigated risk factors for lateral ankle sprains^{2, 159, 186-190} and one study has followed subjects from index sprain to development of CAI,¹⁷ a high-quality prospective study following subjects from prior to an index sprain to development of CAI or copers status has not been published. This type of study could improve the understanding of ankle instability, its risk factors, its development, and the role that rehabilitation

may play in this process. Ultimately, information from this type of study could be used to develop ankle sprain and ankle instability prevention programs that could decrease the societal burden of these disorders.

5.7 Conclusions

1. The CAI group performed the SLST with increased gluteus maximus activation when compared to both the coper and control groups. There was no difference in gluteus maximus activation during the SLST when the coper and control groups were compared. There was no difference between the groups when gluteus medius activation during the SLST was measured.

2. The CAI group demonstrated decreased strength of both the hip lateral rotators and the hip extensors when compared to both the control group. There was no significant difference between the coper and control groups on either of these strength measures. There was no significant difference between any of the groups when hip abductor strength was measured.

3. Future research is needed to determine whether the SLST can be used reliably in a clinical setting to identify gluteus maximums activation impairments and to determine optimal exercise prescription of hip strengthening exercises for this population.

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