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Hip Muscle Activation During Single-leg Tasks: An Enhanced Differential Learning Strategy for Injury Risk Reduction in Female Athletes

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Abstract

Purpose

The purpose of the present study was to investigate the effect of the differential learning strategy on the electromyographic activity of lower limb muscles during single-leg tasks.

Methods

This study involved recreational female athletes (mean age 22.2 ± 2.01 years; height 171.5 ± 5.03 cm; weight 67.8 ± 4.31 kg). Participants performed suspended lunge (SL), single-leg bench squat (SLBS), and lateral slider squat (LSS) under two conditions: with and without differential learning. A randomized, counterbalanced crossover design with a 48-hour washout period was used to eliminate order and familiarization effects. Electromyographic activity of the gluteus maximus (Gmax), gluteus medius (Gmed), rectus femoris (RF), biceps femoris (BF), and adductor longus (AL) was recorded. Data were analyzed using a 2×3 repeated-measures ANOVA.

Results

Group × time effects were significant for Gmed (P = 0.001), BF (P = 0.001), RF (P = 0.006), and AL (P = 0.001). Group effects were significant for Gmed (P = 0.018), RF (P = 0.038), AL (P = 0.001). Time effects were significant for all muscles (all P = 0.001). With differential learning, Gmax and Gmed differed between LSS–SL (P = 0.001) and LSS–SLSB (P = 0.002). BF differed between SL–SLSB (P = 0.001). RF differed between SL–SLSB (P = 0.012) and SL–LSS (P = 0.011). For AL, without differential learning: SL–LSS (P = 0.011), SLSB–LSS (P = 0.001). With differential learning: SL–SLSB (P = 0.002), LSS–SL (P = 0.001), SLSB–LSS (P = 0.001).

Conclusion

Differential learning increases hip and thigh muscle activity: LSS elicited the greatest AL response, SL the greatest RF activation, and SLSB the strongest BF activity. These neuromuscular adaptations suggest improved hip and thigh muscle recruitment patterns that are theoretically associated with lower anterior cruciate ligament injury risk; however, direct injury-related biomechanical or clinical endpoints were not assessed. These exercises may be considered as potential components of injury risk reduction programs, pending confirmation with kinetic and kinematic analyses.

Introduction

Anterior cruciate ligament (ACL) injury is one of the most common injuries in team sports, especially among female athletes (1, 2). The primary mechanism of this injury typically occurs during dynamic movements such as jumping, sudden change of direction, and improper landing, which accounts for approximately two-thirds of ACL tears and is non-contact in nature thus being potentially preventable (3–6). From a biomechanical perspective, deficits in neuromuscular control, inappropriate activation of the quadriceps and hamstring muscles, and poor muscular coordination during high-risk maneuvers such as single-leg landing or sudden change of direction can lead to increased unsafe knee motions (7–9). Among these, single-leg landing with excessive knee abduction, also known as valgus motion, is recognized as a common mechanism of ACL injury (7).

From a biomechanical perspective, knee valgus during single-leg landing significantly increases the risk of ACL rupture due to neuromuscular deficits and poor muscular coordination (7, 8). This impairment in neuromuscular control and muscular coordination can be mitigated through single-leg squat exercises, which reduce knee rotation, abduction, and displacement during athletic movements (10).

Recent findings indicate that employing differential learning (DL) can improve motor control and enhance neuromuscular coordination (11, 12). This method, by introducing targeted variations in movement patterns, stimulates adaptive processes in the nervous system and contributes to improved dynamic joint stability (13, 14). Consequently, the use of DL in sports training can reduce the risk of musculoskeletal injuries and enhance the quality of motor performance (15, 16).

Learning complex motor skills requires exercises that enhance neuromuscular adaptation and body stability control (11). Among these approaches, DL utilizes targeted variations in movement execution to improve neural organization and coordination (12, 13). Instead of monotonous repetition, this method leverages movement fluctuations to activate adaptive processes in the nervous system (14). Such mechanisms increase the body's ability to respond to environmental changes and reduce the injury risk (16). Therefore, the use of DL can be an effective tool for enhancing the quality of motor learning and optimizing sports performance (17).

Previous studies have primarily compared hip muscles activity during unilateral and bilateral functional tasks. Furthermore, to increase the hip muscles activity, attempts were made to make the execution of functional movements more challenging by adding supplementary upper body movements or by carrying external loads. However, it is highly logical that a functional assessment using more ecologically valid tasks that occur in daily activities is essential (18, 19). Moreover, a strategy that can substantially challenge the execution of a functional task and elicit high muscle recruitment for knee joint stability and support is of particular importance. Given that the foundation of DL is based on variety and variability in exercise execution, it is warranted to apply DL strategies during single-leg functional movements to expose individuals to multiple challenges for enhancing hip muscle activity. Therefore, the aim of the present study was to investigate hip muscle activity during single-leg functional movements with and without DL strategy.

Methods

Study Design

A randomized, counterbalanced, crossover design was employed. Each participant attended three sessions: (i) a familiarization and maximum voluntary isometric contraction (MVIC) assessment session, (ii) a main testing session consisting of two conditions non-differential learning (non-DL) and differential learning (DL) separated by a 48-hour washout period, and (iii) a follow-up session for any missing data (if required). The order of conditions (non-DL first vs. DL first) was randomized using a computer-generated block randomization sequence (block size of 4) with allocation concealed in opaque, sealed envelopes. The same assessor, blinded to the condition order, performed all electromyographic (EMG) data processing and statistical analyses. The study was approved by the Ethics Committee of the Research Institute of Physical Education and Sport Sciences, Iran (IR.SSRC.REC.1404.073). All procedures in this study were performed in accordance with the Declaration of Helsinki (22).

Participants

Twenty recreationally active female athletes (mean age 22.2 ± 2.01 years; height 171.5 ± 5.03 cm; weight 67.8 ± 4.31 kg; BMI 23.06 ± 1.36 kg/m²; sport experience 5.70 ± 1.89 years) participated. Inclusion criteria were: (i) age 20–25

years; (ii) minimum 2 sessions/week of 90 minutes of recreational sports activity; (iii) normal BMI (18.5–25 kg/m²); (iv) no history of fracture, dislocation (20), or ligamentous injury in the lower limbs (21); (v) no low back pain in the past year; (vi) no vestibular disorders; (vii) no participation in structured motor learning or neuromuscular training programs in the past year; and (viii) self-reported regular menstrual cycles. Exclusion criteria included: (i) any acute or chronic lower extremity musculoskeletal injury at the time of testing (20, 21); (ii) history of ACL reconstruction or other knee surgery; (iii) pregnancy or suspected pregnancy; (iv) use of oral contraceptives or hormonal therapy affecting neuromuscular function; and (v) testing during the menstrual phase (days 1–5) to minimize potential hormonal effects on ligament laxity and muscle activation.

This study was conducted in accordance with the Declaration of Helsinki and approved by the ethical committee of the Sport Sciences Research Institute (Approval ID: IR.SSRC.REC.1404.073) before data collection. All participants were informed about the purpose, procedures, potential risks, and benefits of the study. Written informed consent was obtained from each participant prior to their involvement. The consent form included permission for data analysis and publication of anonymized results. Each human participant voluntarily provided written informed consent after receiving a complete explanation of the study protocol. Participants were assured of their right to withdraw from the study at any time without consequences. Consent records are retained by the corresponding author and are available for journal review if requested.

Sample Size Justification

Based on a previous study examining hip muscle activation during single-leg tasks (19), and anticipating a large effect size (Cohen's $d \geq 0.80$) for the DL × time interaction, a sample size of 16 participants was required to achieve 80% power at $\alpha = 0.05$ (G*Power 3.1, repeated-measures ANOVA, within-between interaction). To account for potential dropout or technical data loss, 20 participants were enrolled.

Familiarization Session

Forty-eight hours before the main testing session, participants attended a 45-minute familiarization session. They were instructed on proper execution of the three single-leg tasks (SL, SLBS, and LSS) according to standardized protocols. Participants practiced each movement for 10 repetitions under supervision until they demonstrated consistent technique. No EMG data were recorded during familiarization, and no differential learning strategies were introduced. Participants were also familiarized with the MVIC testing procedures for each muscle.

Main Testing Session

On the main testing day, participants arrived at the laboratory at the same time of day (± 1 hour) for both conditions (non-DL and DL) to control for circadian variation. Testing was scheduled during the follicular phase (days 6–12 of the menstrual cycle) for all participants to minimize hormonal influence on neuromuscular control. Each condition lasted approximately 60 minutes, with a 48-hour washout period between conditions.

Procedures

Noraxon Ultium (Noraxon Inc., Scottsdale, AZ, USA) wireless surface electromyography (sEMG) sensors were used to record unilateral electrical activity of the dominant-side Gmax, Gmed, BF, RF, and AL muscles. Participants performed three exercise tasks; the single-leg bench squat (SLBS), lateral slider squat (LSS), and suspended lunge (SL) for 10

repetitions without the DL strategy, and then performed the same three tasks for 10 repetitions with the DL strategy. Electrodes were placed according to the SENIAM European protocol.

The Gmax electrode was placed midway between the greater trochanter and the sacral spine (19). For the MVC test, the subject was positioned prone with a pillow under the abdomen, the test leg was placed with the hip in a neutral position and the knee flexed to 90 degrees, and resistance was applied at the posterior thigh while the individual attempted to push their leg toward the ceiling (19).

The Gmed electrode was placed on the lateral aspect of the hip at one-third the distance between the iliac crest and the greater trochanter (19). For the MVC test, the participant lay on their side on the non-test leg, with the test leg extended and in approximately 5 degrees of abduction; resistance was then applied near the ankle (lateral malleolus) as the participant attempted to lift their leg (19).

The BF electrode was placed midway between the ischial tuberosity and the popliteal fossa (19). During the MVC test, the individual was positioned prone with a pillow under the abdomen, the knee was flexed to 90 degrees, resistance was applied near the ankle, and the participant attempted to flex the knee (19).

The RF electrode was placed midway between the anterior superior iliac spine and the base of the patella (19). During the MVC test, the participant sat on the edge of the examination table, leaning back. The test knee was flexed to approximately 10 degrees, resistance was applied near the ankle, and the individual attempted to extend the knee (19).

The AL muscle was palpated during an isometric contraction, and the electrode was placed directly on the muscle belly approximately 4 cm from the pubic symphysis (19). For the AL MVC test, the participant lay on their side on the test leg, with the non-test leg bent and placed on a stool in front of the abdomen. Resistance was applied near the ankle, and the individual attempted to lift their leg upward (19).

Prior to the placement of bipolar surface electrodes on the skin of the participants' dominant leg, the hair in the target area was completely shaved, and the skin was prepared with alcohol and cotton. All electrodes were positioned parallel to the muscle's line of action. The MVC tests for each muscle were performed for 5 seconds, with a rest period of at least 1 minute between tests (23). The maximum muscle electrical activity was calculated only during the concentric phase over a 10-second interval for all three movements.

Single leg Tasks

Participants performed three tasks in a randomized order (determined by a separate randomization list, counterbalanced across conditions and participants):

Single-leg bench squat (SLBS): Standing on a 30-cm platform on the dominant leg, non-dominant leg suspended, trunk neutral, knee flexed to 60° over 3 seconds, then returned to standing at self-selected speed (10).

Lateral slider squat (LSS): Single-leg squat to 60° knee flexion with 40° trunk flexion; free leg slid diagonally (45°) outward and backward at a rhythm of 1 second out, 1 second back (19).

Suspended lunge (SL): Non-dominant foot placed in a TRX strap (suspended ~ 15 cm above ground), dominant foot on ground; torso and suspended leg moved backward simultaneously to 90° front knee flexion; 3-second descent, 1-second pause, 3-second ascent (18). Each task was performed for 10 consecutive repetitions. A 2-minute rest interval separated each task. The order of tasks was randomized for each participant and remained identical across the two

conditions (non-DL vs. DL) to isolate the effect of DL. Participants were not informed of the specific hypothesis regarding DL versus non-DL comparisons.

DL Condition

Participants performed the same three tasks (SL, SLBS, LSS) but with trial-to-trial variability introduced in movement execution according to the differential learning framework (24). No two consecutive trials were identical. Variability was introduced systematically by altering, for each trial, the arm position (such as hands on hips, crossed over chest, extended forward, or one hand on hip and the other extended), the head position (neutral, slight flexion, extension, or rotation to left or right), the visual input (eyes open, eyes closed, or gaze directed to a specific target), foot placement (subtle variations in stance width or foot rotation angle), trunk posture (slight forward lean, neutral, or upright), footwear (with or without shoes), and hand equipment (holding a small ball or not). Participants were free to perform any combination of these variations across trials, ensuring that movement variability was applied in each trial. For example, during the suspended lunge (SL), each of the 10 trials was unique: (1) hands on hips, head neutral, eyes open; (2) arms extended forward, head rotated to the right, eyes closed; (3) hands crossed over chest, head slightly flexed, gaze fixed on a target; (4) one hand on hip (right), the other extended forward, head extended backward, eyes open; (5) both arms extended sideways, head rotated to the left, eyes closed; (6) hands clasped behind the head, head neutral, gaze downward; (7) arms crossed in front of chest, head rotated to the right, eyes open; (8) one hand extended forward, the other placed on the lower back, head slightly extended, eyes closed; (9) both hands on the waist, head rotated alternately left and right during the descent, eyes open; (10) arms relaxed at sides, head neutral, eyes closed with internal focus on balance. No two trials were identical. The trainer provided no prescriptive instructions on "correct" form, only verbal cues to follow the next variation (e.g., "now arms forward, eyes closed, without shoes"), and no corrective feedback was given.

Non-DL Condition

Participants performed the same three tasks with standardized, repetitive execution. All 10 repetitions were performed identically: target knee flexion angles (SLBS/LSS: 60°; SL: 90°), self-selected but fixed tempo (3 sec descent, 1 sec pause, 3 sec ascent), hands on hips, eyes open, and no added instability. Participants received no feedback or instruction beyond a single demonstration before the first repetition.

Order of Conditions and Randomization

Using block randomization (block size 4, generated by www.randomizer.org), 10 participants were assigned to the sequence non-DL first → DL second (after 48 h washout), and 10 participants to DL first → non-DL second. The data analyst was blinded to sequence allocation. This design allows separation of intervention effects from order/familiarization/fatigue effects by comparing (i) DL vs. non-DL within participants, and (ii) sequence groups (non-DL→DL vs. DL→ non-DL) to detect potential carryover effects.

Data Processing

The raw EMG signals were filtered using a zero-lag, fourth-order Butterworth filter with a bandwidth of 10–450 Hz. The data were then processed and corrected using a root mean square algorithm (25). Prior to performing the movements, the maximum voluntary isometric contraction for each muscle was recorded to normalize the EMG data. The MVIC tests for each muscle were performed for a duration of 5 seconds. The rest period between each test was at least 1 minute (23). The maximum electrical activity of the muscles was calculated only during the concentric phase over a 10-second interval for all three movements.

Statistical Analysis

Descriptive are presented as mean ± standard deviation. The Shapiro-Wilk test indicated that all EMG variables were normally distributed, and the assumption of homogeneity of variance was met based on Levene's test. To evaluate the effect of DL on muscle activation, a 2 (condition: non-DL vs. DL) × 3 (exercise: SL, SLBS, LSS) repeated-measures ANOVA was conducted separately for each muscle (Gmax, Gmed, BF, RF, AL). Condition order (non-DL first vs. DL first) was initially entered as a between-subjects' factor to test for carryover or order effects; no significant order × condition interaction was detected (all $P > 0.20$), therefore data were collapsed across order for the primary analysis. Post-hoc pairwise comparisons were performed using Bonferroni correction for multiple comparisons. Effect sizes are reported as Cohen's d with 95% confidence intervals (small = 0.2, medium = 0.5, large = 0.8). Statistical significance was set at $\alpha = 0.05$ (two-tailed).

Results

Table 1 displays the demographic characteristics of the participants, including age, weight, height, body mass index, and sports experience.

Table 1
Demographic characteristics of the study group participants (Mean ± Standard Deviation).

Participant characteristics	Mean ± SD
Age (years)	22.20 ± 2.01
Weight (kg)	67.80 ± 4.31
Height (cm)	171.50 ± 5.03
BMI (kg/m^2)	23.06 ± 1.36
Sport experience (years)	5.70 ± 1.89

Table 1 is near here.

The group × time interaction was significant for the Gmed, BF, RF, and AL ($P < 0.05$) but not for the Gmax. The main effect of group was significant for the Gmed, RF, and AL, while Gmax and BF showed no significant group effect. The main effect of time was significant for all muscles (Gmax, Gmed, BF, RF, AL; $P = 0.001$).

In the non-DL condition, there were no significant differences between the three movements for Gmax, Gmed, BF, or RF. For the AL, significant differences were observed between SL and LSS ($P = 0.011$) and between SLSB and LSS ($P = 0.001$).

Under the DL condition, Gmax showed no differences between movements. Gmed activity was higher in LSS compared to SL ($P = 0.001$) and SLSB ($P = 0.002$). BF activity was higher in SL than SLSB ($P = 0.001$). RF showed greater activity in SL compared to SLSB ($P = 0.012$) and LSS ($P = 0.011$). AL activity differed between SL and SLSB ($P = 0.002$), LSS and SL ($P = 0.001$), and SLSB and LSS ($P = 0.001$).

Bonferroni post-hoc tests indicated that non-DL generally produced higher muscle activity than DL for Gmax, Gmed, and BF across all movements, for RF in SL and SLSB, and for AL in SL and LSS, suggesting a stronger effect magnitude under DL for some muscles. However, no differences were found for RF in LSS and AL in SLSB.

Overall, the SL was most effective for activating Gmax, RF, and AL; LSS was most effective for Gmed; and SLSB was most effective for BF, each showing a larger effect size compared to the other movements.

Table 2 is near here.

Table 2
Muscle activity changes during the three training movements with and without the application of the differential learning strategy (Mean ± Standard Deviation).

Variable	Exercise movement	non-DL	DL	ES ($CI_{95\%}$) †	p-value		
					Main Effect of Time	Main Effect of Group	Group×Time Interaction
Gmax (%mvc)	SL	27.09 ± 6.76	35.30 ± 8.75¥	1.05 (0.11–1.98)	F = 169.604 P = 0.001*	F = 0.209 P = 0.812	F = 0.898 P = 0.413
	SLSB	27.46 ± 7.78	36.06 ± 8.79¥	1.03 (0.10–1.97)			
	LSS	29.41 ± 8.25	36.13 ± 8.40¥	0.80 (-0.10-1.71)			
Gmed (%mvc)	SL	20.14 ± 6.25	28.17 ± 7.96¥	1.12 (0.17–2.06)	F = 298.093 P = 0.001*	F = 4.330 P = 0.018	F = 29.521 P = 0.001*
	SLSB	24.67 ± 6.75	29.39 ± 6.57¥	0.70 (-0.19-1.61)			
	LSS	23.06 ± 6.92	37.51 ± 6.74¥,a	2.11 (1.02–3.21)			
BF (%mvc)	SL	24.28 ± 6.16	28.24 ± 7.11¥	0.59 (-0.3-1.49)	F = 335.047 P = 0.001*	F = 1.017 P = 0.368	F = 35.775 P = 0.001*
	SLSB	21.41 ± 6.47	36.65 ± 7.56¥,c	2.16 (1.06–3.27)			
	LSS	22.30 ± 4.59	33.32 ± 6.71¥	1.91 (0.85–2.97)			
RF (%mvc)	SL	29.47 ± 5.51	35.29 ± 5.95¥,a,c	1.01 (0.08–1.94)	F = 35.223 P = 0.001*	F = 3.467 P = 0.038*	F = 5.605 P = 0.006*

Abbreviations: Results are presented as mean ± standard deviation; *, statistically significant difference (P < 0.05); ¥, significant difference from the condition without differential learning to the condition with differential learning; †, effect size; a: significant difference between the LSS and the SL; b: significant difference between the LSS and the SLSB; c: significant difference between the SL and the SLSB

	SLSB	27.31 ± 5.24	29.92 ± 5.35[¥]	0.49 (-0.39-1.38)			
	LSS	28.48 ± 4.06	29.91 ± 5.58[¥]	0.29 (-0.58-1.17)			
AL (%mvc)	SL	19.27 ± 2.75	23.94 ± 3.52[¥,c]	1.47 (0.48–2.46)	F = 78.788 P = 0.001*	F = 16.464 P = 0.001*	F = 34.959 P = 0.001*
	SLSB	19.11 ± 6.29	18.11 ± 6.29	-0.15 (-1.03-0.71)			
	LSS	23.83 ± 4.49 [a,b]	30.27 ± 5.18[¥,a,c]	1.32 (0.36–2.29)			

Abbreviations: Results are presented as mean ± standard deviation; *, statistically significant difference (P < 0.05); ¥, significant difference from the condition without differential learning to the condition with differential learning; †, effect size; a: significant difference between the LSS and the SL; b: significant difference between the LSS and the SLSB; c: significant difference between the SL and the SLSB

Discussion

The results of this study indicated that the SLSB produced the highest activation of the Gmax and BF, while the LSS demonstrated the highest response in the Gmed and AL. This pattern aligns with evidence regarding the effect of acceleration-based exercises on the Gmed (26), the role of high-load resistance training in increasing Gmax engagement (27), and the association between Gmed activation and functional pelvic control (28). The increased BF response in the single-leg squat is consistent with biomechanical explanations related to increased hip torque and greater trunk control demands in this movement (29), as well as reports of compensatory hamstring activation patterns in individuals with dynamic valgus during squats (30). Accordingly, findings that targeted hip adductor training can modulate quadriceps and hamstring activation patterns and ACL risk factors are consistent with existing evidence (31). The increased RF activity in the SL also corresponds with results indicating the influence of stride length and movement phase on thigh muscle engagement in various lunges (32).

Regarding the AL, the superiority of the LSS and SL aligns with numerous findings reporting increased adductor and gluteal muscle activity during suspension and sliding exercises (18, 19, 32–35). In contrast, some earlier reports, such as the study by Beaudro (36), reported lower activation for these movements, likely due to lower task intensity and the absence of balance and movement challenges. Furthermore, substantial evidence on the role of differential learning in modifying movement patterns and reducing biomechanical risk factors for ACL injury (24, 37, 38) also reinforces the findings of this study, indicating that exercises emphasizing movement variability, suspended, sliding, and unstable conditions can significantly increase lower limb muscle activation, improve neuromuscular coordination, and enhance dynamic stability of the hip and knee.

In this study, the application of differential learning resulted in a significant increase in Gmax and Gmed activity during the SL, SLSB, and LSS. The highest Gmax response was observed in the SLSB, while the greatest Gmed activity was seen in the LSS; this can be attributed to the higher demands for balance control, pelvic stabilization, and targeted

instability in these movements. Differential learning, by introducing movement variability and controlled instability, enhances motor unit recruitment and inter-muscular coordination, which explains the increased EMG response. This pattern aligns with research demonstrating that targeted exercises can increase Gmed activity (26) and with studies emphasizing that resistance and single-leg exercises are effective stimuli for the Gmax (27). Furthermore, findings related to the role of differential learning in improving movement patterns and reducing ACL injury risk factors (24, 37) confirm that this approach can be an effective option for strengthening the hip stabilizer muscles and enhancing lower limb function.

Differential learning led to a significant increase in BF and RF activity during the SL, SLSB, and LSS. The highest BF response was observed in the SLSB, while the greatest RF activity was seen in the SL; a pattern consistent with the biomechanical nature of the movements, as the single-leg squat increases trunk control and hip torque, and the SL, with its altered stride length, induces greater RF engagement. The mechanism for this increased activation can be attributed to the controlled instability and movement variability in differential learning, which enhances hamstring-quadriceps coordination. These results align with studies reporting the role of the hamstrings in knee stabilization (30) and the modification of activation patterns to reduce ACL risk (31), as well as research confirming the efficacy of differential learning in improving movement patterns (24, 37). Overall, the SLSB and the SL were the most effective exercises for the BF and RF, respectively.

The AL showed a significant difference across all three movements. Under the standard condition, the LSS elicited greater activation than the SL and the SLSB, and these differences were amplified with the addition of differential learning; a significant increase in activity was observed in the SL and the LSS, but no change was seen in the SLSB. This pattern aligns with the nature of the movements, as the sliding and instability of the LSS and the altered stride length in the SL impose greater demand on the adductors. These findings are consistent with studies reporting higher AL activation during unstable and suspended exercises (18, 33) and with research confirming the role of this muscle in lower limb stability and modifying movement patterns (32, 35). They also correspond with evidence on the effect of differential learning in reducing ACL risk factors (24, 38). Overall, the SL, with the largest effect size (1.47), was the most effective movement for activating this muscle.

In this study, differential learning significantly increased the activity of the Gmax, Gmed, BF, RF, and AL during the SL, SLSB, and LSS. The highest responses were observed in the SLSB (for Gmax and BF), the LSS (for Gmed and AL), and the SL (for RF). By introducing movement variability and controlled instability, this approach enhances motor unit recruitment, improves inter-muscular and hamstring-quadriceps coordination, consequently refines muscle activation patterns, enhances lower limb control, increases dynamic stability of the hip and knee, and reduces ACL injury risk factors. Therefore, differential learning is recommended as an effective and practical approach for inclusion in functional, preventive, and rehabilitation programs (24, 37, 38).

Despite the methodological strengths of this study, several limitations should be acknowledged. First, the sample was limited to 20 young female recreational athletes, which may reduce generalizability to other populations (e.g., males, elite athletes, or older individuals). Second, the study relied solely on surface EMG as a surrogate measure of neuromuscular control; we did not directly assess kinetic (e.g., knee abduction moments) or kinematic (e.g., joint angles) risk factors for ACL injury, nor did we track clinical injury outcomes. Third, while we controlled for menstrual cycle phase (follicular phase) and used a counterbalanced crossover design, other factors such as daily fatigue or nutritional status were not strictly controlled. Finally, the absence of long-term follow-up precludes conclusions about retention of the observed EMG changes or translation to actual injury rates. Future studies integrating three-dimensional motion analysis, larger and more diverse samples, and prospective injury tracking are warranted to confirm and extend these findings.

Conclusions

The results of this study demonstrate that implementing a differential learning strategy leads to a significant increase in hip and thigh muscle activity; specifically, the LSS elicited the greatest response in the AL, the SL produced the highest engagement of the RF, and the SLSB had the greatest effect on the BF. These changes indicate improved neuromuscular activation patterns of the hip and thigh musculature. Although such patterns are theorized to contribute to dynamic knee stability, the direct reduction of biomechanical risk factors for ACL injury was not measured in this study. Future research incorporating kinetic and kinematic analyses is warranted to establish a causal link between differential learning-induced EMG changes and actual injury risk. Therefore, coaches and athletes are advised to incorporate differential learning-based exercises into prevention and rehabilitation programs as a complementary approach pending further confirmatory studies.

Declarations

Competing interests

None declared.

Patient consent for publication

Not applicable

Ethics approval

This study involves human participants. This study was assessed and approved by the Ethical Committee of Sport Sciences Research Institute (Approval ID: IR.SSRC.REC.1404.073) before data collection. Participants gave informed consent to participate in the study. Participants provided written informed consent to participate in the study, in accordance with the Declaration of Helsinki.

Consent to Participate declaration

Written informed consent was obtained from all individual participants included in the study.

Consent for publication

The authors declare that the human participant has provided informed consent for the use of their images in the figures.

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Author Contribution

SR: contributed to Investigation, Conceptualization, Methodology, Data capture.NGZH: contributed to Investigation, Data curation, Conceptualization, Data capture, Date analysis. HAG: contributed to Writing- Original draft preparation, Investigation, Date analysis, Scientific editing. BS: contributed to Writing- Original draft preparation, Investigation, Scientific editing. All authors read and approved the final manuscript for publication.

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Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

References

1. Paul, J. J. et al. Jumping Versus Nonjumping Anterior Cruciate Ligament Injuries: A Comparison of Pathology. Clin J Sport Med [Internet]. ;13(1). (2003). Available from: https://journals.lww.com/cjsportsmed/fulltext/2003/01000/jumping_versus_nonjumping_anterior_cruciate.1.aspx
2. Monajati, A., Larumbe-Zabala, E., Goss-Sampson, M. & Naclerio Naclerio, F. The effectiveness of injury prevention programs to modify risk factors for non-contact anterior cruciate ligament and hamstring injuries in uninjured team sports athletes: A systematic review. PLoS One. ;11(5). (2016).
3. Beynnon Bruce, D., Johnson Robert, J., Abate Joseph, A., Fleming Braden, C. & Nichols Claude, E. Treatment of Anterior Cruciate Ligament Injuries, Part I. Am J Sports Med [Internet]. ;33(10):1579–602. (2005). Available from: https://doi.org/10.1177/0363546505279913
4. Griffin, L. Y. et al. Understanding and Preventing Noncontact Anterior Cruciate Ligament Injuries A Review of the Hunt Valley II Meeting, January 2005. *Am. J. Sports Med.* **34** (9), 1512–1532 (2006).
5. Alentorn-Geli, E. et al. Prevention of non-contact anterior cruciate ligament injuries in soccer players. Part 2: A review of prevention programs aimed to modify risk factors and to reduce injury rates. *Knee Surg. Sport Traumatol. Arthrosc.* **17** (8), 859–879 (2009).
6. Chappell Jonathan, D., Yu Bing, Kirkendall Donald, T. & Garrett William, E. A Comparison of Knee Kinetics between Male and Female Recreational Athletes in Stop-Jump Tasks. Am J Sports Med [Internet]. ;30(2):261–7. (2002). Available from: https://doi.org/10.1177/03635465020300021901
7. Hewett, T. E. et al. Biomechanical measures of neuromuscular control and valgus loading of the knee predict anterior cruciate ligament injury risk in female athletes: A prospective study. *Am. J. Sports Med.* **33** (4), 492–501 (2005).
8. Ford, K. R., Myer, G. D. & Hewett, T. E. Valgus knee motion during landing in high school female and male basketball players. *Med. Sci. Sports Exerc.* **35** (10), 1745–1750 (2003).
9. Dedinsky, R., Baker, L., Imbus, S., Bowman, M. & Murray, L. Exercises That Facilitate Optimal Hamstring And Quadriceps Co-Activation To Help Decrease Acl Injury Risk In Healthy Females: A Systematic Review Of The Literature. *Int. J. Sports Phys. Ther.* **12** (1), 3–15 (2017).
10. Monajati, A., Larumbe-Zabala, E., Goss-Sampson, M. & Naclerio, F. Surface Electromyography Analysis of Three Squat Exercises. *J. Hum. Kinet*. **67** (1), 73–83 (2019).

11. Fisher, J., Steele, J. & Smith, D. High-and low-load resistance training: interpretation and practical application of current research findings. *Sport Med.* **47** (3), 393–400 (2017).
12. Dhawale, A. K., Smith, M. A. & Ölveczky, B. P. The Role of Variability in Motor Learning. *Annu. Rev. Neurosci.* **40**, 479–498 (2017).
13. Newell, K. M. & McDonald, P. V. Learning to Coordinate Redundant Biomechanical Degrees of Freedom. In: Swinnen SP, Heuer H, Massion J, Casaer P, editors. Interlimb Coordination [Internet]. San Diego: Academic Press; pp. 515–36. (1994). Available from: https://www.sciencedirect.com/science/article/pii/B9780126792706500292
14. Schöllhorn, W. I. Individualität-ein vernachlässigter Parameter? (Individuality a neglected parameter?) [Internet]. (1999). Available from: https://www.researchgate.net/publication/285755643
15. Padua, D. A. et al. National athletic trainers' association position statement: Prevention of anterior cruciate ligament injury. *J. Athl Train.* **53** (1), 5–19 (2018).
16. Mohammadi Orangi, B. et al. Motor learning methods that induce high practice variability reduce kinematic and kinetic risk factors of non-contact ACL injury. *Hum. Mov. Sci.* ;**78**. (2021).
17. Ranganathan, R., Newell, K. M., Ranganathan, R. & Newell, K. M. Changing Up the Routine: Intervention-Induced Variability in Motor Learning [Internet]. Vol. 41, Exerc. Sport Sci. Rev. (2013). Available from:
18. Krause, D. A. et al. Electromyography Of The Hip And Thigh Muscles During Two Variations Of The Lunge Exercise: A Cross-Sectional Study. *Int. J. Sports Phys. Ther.* **13** (2), 137–142 (2018).
19. Krause, D. A. & Hollman, J. H. Electromyographic Analysis Of Hip Muscle Activation During A Single Limb Squat Lateral Slide Exercise. *Int. J. Sports Phys. Ther.* **15** (5), 755–762 (2020).
20. Sheikhi, B., Letafatkar, A., Thomas, A. C. & Ford, K. R. Altered trunk and lower extremity movement coordination after neuromuscular training with and without external focus instruction: a randomized controlled trial. *BMC Sports Sci. Med. Rehabil* ;**13**(1). (2021).
21. Ghanati, H. A., Letafatkar, A., Almonroeder, T. G. & Rabiei, P. Examining the Influence of Attentional Focus on the Effects of a Neuromuscular Training Program in Male Athletes. *J. strength. Cond Res.* ; (2020).
22. Association, W. M. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human participants. *JAMA* ; (2024).
23. Struminger, A. H., Lewek, M. D., Goto, S., Hibberd, E. & Blackburn, J. T. Comparison of gluteal and hamstring activation during five commonly used plyometric exercises. *Clin. Biomech.* **28** (7), 783–789 (2013).
24. Ghanati, H. A., Letafatkar, A., Shojaedin, S., Hadadnezhad, M. & Schöllhorn, W. I. Comparing the Effects of Differential Learning, Self-Controlled Feedback, and External Focus of Attention Training on Biomechanical Risk Factors of Anterior Cruciate Ligament (ACL) in Athletes: A Randomized Controlled Trial. *Int. J. Environ. Res. Public. Health* ;**19**(16). (2022).
25. Zebis, M. K. et al. Effects of evidence-based prevention training on neuromuscular and biomechanical risk factors for ACL injury in adolescent female athletes: A randomised controlled trial. *Br. J. Sports Med.* **50** (9), 552–557 (2016).
26. Goller, M., Quittmann, O. J. & Alt, T. How to activate the glutes best? Peak muscle activity of acceleration-specific pre-activation and traditional strength training exercises. *Eur. J. Appl. Physiol.* **124** (6), 1757–1769 (2024).
27. Krause Neto, W., Krause, T. L. V. & Gama, E. F. The impact of resistance training on gluteus maximus hypertrophy: a systematic review and meta-analysis. *Front. Physiol.* **16**, 1542334 (2025).
28. Davoudi, M., Salami, F., Reisig, R., Gather, K. S. & Wolf, S. I. Gluteus medius muscle activation patterns during gait with Cerebral Palsy (CP): A hierarchical clustering analysis. *PLoS One*. **20** (1), e0309582 (2025).

29. Farina, D., Merletti, R. & Enoka, R. M. The extraction of neural strategies from the surface EMG. *J. Appl. Physiol.* **96** (4), 1486–1495 (2004).
30. Khou, S. B., Saki, F. & Tahayori, B. Muscle activation in the lower limb muscles in individuals with dynamic knee valgus during single-leg and overhead squats: a meta-analysis study. *BMC Musculoskelet. Disord*. **25** (1), 652 (2024).
31. Jeong, J., Choi, D. H. & Shin, C. S. The Effect of Strength Training Targeting Medial Thigh Muscles on Neuromuscular and Biomechanical Risk Factors for Anterior Cruciate Ligament Injury: A Randomized Controlled Trial. *Sport Med. - Open.* **11** (1), 5 (2025).
32. Escamilla, R. F. et al. Effects of Step Length and Stride Variation During Forward Lunges on Lower-Extremity Muscle Activity. *J. Funct. Morphol. Kinesiol.* **10** (1), 42 (2025).
33. Aguilera-Castells, J. et al. Muscle activity of Bulgarian squat. Effects of additional vibration, suspension and unstable surface. *PLoS One*. **14** (8), e0221710 (2019).
34. Hoogenboom, B. J., Ferguson, M., Krauss, Z. & Tran, S. The Reverse Lunge: A Descriptive Electromyographic Study. *Appl. Sci.* **14** (24), 11480 (2024).
35. Gallego-Pérez, A. & Benito-Martínez, E. Alonso-Cortés Fradejas B. Normative Muscle Activation Patterns During One and Five Countermovement Jumps. *Bioengineering* **12** (7), 767 (2025).
36. Boudreau, S. N. et al. Hip-muscle activation during the lunge, single-leg squat, and step-up-and-over exercises. *J. Sport Rehabil*. **18** (1), 91–103 (2009).
37. Gholami, F. et al. Comparing the Effects of Differential and Visuo-Motor Training on Functional Performance, Biomechanical, and Psychological Factors in Athletes after ACL Reconstruction: A Randomized Controlled Trial. *J. Clin. Med.* ;**12**(8). (2023).
38. Özdemir, O., Çamlıgüney, A. & Erdil, G. The Impact of Differential Learning and Functional Strength Training on Biomechanical Performance Parameters in Tennis Athletes. *Int. J. Sport Exerc. Train. Sci.* ; (2024).