Out of Lab Longitudinal Gait Assessment of Participants Pre and Post Anterior

Cruciate Ligament Reconstruction Surgery: An Observational Longitudinal Study

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All authors contributed to the interpretation of the data and critically revised the manuscript for important intellectual content. All the authors finally approved the manuscript. Arielle Fischer was responsible for obtaining project funding and takes responsibility for the integrity of the work as a whole. All authors have read and agreed to the published version of the manuscript.



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2	Objective:	To evaluate	the	longitudinal	changes	in	knee	sa

Objective: To evaluate the longitudinal changes in knee sagittal kinematics pre- and post-anterior cruciate

ligament reconstruction (ACLR) during varying walking speeds in non-laboratory environments. A secondary

objective describing the hip and ankle joint kinematics.

5 **Design:** Longitudinal observational study.

6 **Setting:** Hospital.

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7 Patients or Other Participants: Forty ACLR patients and 17 healthy matched controls were recruited.

Main Outcome Measures: Knee joint sagittal kinematics measured using seven inertial measurement units at

pre-surgery, three-, and five-months post-surgery while walking at slow, normal, and fast speeds.

Results: At pre-surgery, compared to the contralateral limb, the injured knee exhibited greater minimum flexion

during normal and fast walking (p≤.008) and exhibited less knee flexion at the first peak (p=.006). SPM

revealed significant differences throughout the gait cycle at all speeds (p≤.033). Compared to controls, the

injured knee had greater minimum flexion during normal and slow walking (p≤.025). At three months,

compared to the contralateral limb, the injured knee showed increased minimum flexion across all speeds

(p≤.005) and exhibited less knee flexion at the first peak during fast walking (p<.001). SPM indicated

significant differences throughout the gait cycle at all speeds (p≤.028). Compared to controls, the injured knee

remained more flexed at the minimum angle across all speeds (p<.001) and exhibited less knee flexion at the

first peak during slow walking (p=.031). At five months, differences between limbs were reduced, with

significant differences in minimum flexion at all speeds (p≤.027). SPM differences were limited to specific gait

cycle portions during normal and fast walking (p≤.011). Compared to controls, the injured knee remained more

flexed at the minimum angle during slow and normal walking (p≤.005). Lastly, hip adaptations resolved while

ankle asymmetries persisted during terminal stance.

Conclusion: ACLR patients demonstrated progressive improvements in knee sagittal kinematics, indicating a

recovery trend. However, the recovery was non-linear across different walking speeds.

Keywords: Anterior cruciate ligament; Gait; Walking; Kinematic analysis; Lower limb

28 Introduction

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Anterior cruciate ligament reconstruction (ACLR) often results in lasting kinematic asymmetries while walking, reduced quality of life, and increased risk of post-traumatic osteoarthritis (PTOA). 1-3 Early gait assessment within the first six months post-ACLR is critical for guiding rehabilitation and reducing long-term joint degeneration risks, as these asymmetries can significantly affect knee function and potentially accelerate PTOA development.^{2,4} Despite its clinical importance, comprehensive biomechanical assessment accounting for pre-operative status, varying walking speeds, and hip and ankle compensatory adaptations remains underexplored. Moreover, traditional analyses focus primarily on discrete timepoints of the gait cycle, limiting our understanding of continuous movement patterns in non-laboratory settings. The role of pre-operative biomechanical status is often underexplored, with few studies providing pre- and post-operative assessments. This gap is significant as pre-operative biomechanics can influence the post-operative recovery outcomes. 6-8 Specifically, Büttner et al. (2024) demonstrated that gait asymmetries are present pre-operatively and persist through recovery timepoints at 2, 4, 6, and 12 months post-ACLR. Further, Davis-Wilson et al. (2020) observed bilateral gait abnormalities at six and twelve months post-ACLR compared to uninjured controls, 10 emphasizing the need to assess recovery across multiple stages and walking speeds. Walking speed influences lower extremity biomechanics after ACLR, with faster speeds typically exacerbating biomechanical deficits. 11-13 This is particularly important as daily activities require walking at varying speeds—from slow ambulation during routine tasks to fast walking when commuting or exercising. By assessing these speed-dependent changes, clinicians can better understand how patients adapt their movement strategies in real-world contexts and can adjust rehabilitation interventions to improve performance across all activity

levels. Yet only a handful of studies assessed the influence of walking speed on gait kinematics after ACLR. 11-13 Understanding how the relationship between walking speed and biomechanics changes throughout rehabilitation might lead to a better insight into recovery patterns and real-world mobility demands. While knee kinematics is a primary concern post-ACLR, assessing the hip and ankle joints alongside it provides insight into compensatory strategies and overall lower limb adaptation. Previous studies have shown that ACLR patients display compensatory sagittal plane adaptations, specifically, reduced hip flexion and ankle dorsiflexion, contributing to asymmetrical gait patterns. 14-16 These adaptations may contribute to altered joint loading patterns throughout the kinetic chain. Movement analysis often focuses on specific gait events, such as minimum and maximum points, providing important snapshots of movement. However, continuous analysis methods like statistical parametric mapping (SPM) offer a more comprehensive view of movement mechanics and can be used alongside traditional discrete point analysis.¹⁷ Specifically, SPM addresses these limitations of discrete analysis by examining continuous data, allowing the evaluation of how joint angles change over time and identifying differences or asymmetries that discrete analysis might miss. For example, SPM analysis of walking revealed that knee sagittal kinematic asymmetries after ACLR occur during extended portions of the stance phase rather than just at discrete peak angles. 11,18 Despite these advantages, SPM remains underutilized in gait analysis, particularly with IMU data in ACLR populations. ¹⁹ Traditional optoelectrical camera systems, while accurate, are expensive and confined to laboratory environments, reducing ecological validity. 20,21 In contrast, wearable technologies like inertial measurement units (IMUs) allow real-world kinematic data collection, although normative values are needed to interpret differences from optoelectrical data.²² Although no

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studies directly link IMU-based gait analysis to ACLR and PTOA outcomes, IMU systems have shown promise in evaluating gait deviations associated with OA. When paired with SPM, wearable sensors have demonstrated the ability to identify knee OA gait deviations characterized by asymmetries in ankle, knee, and hip sagittal kinematics during stance and swing phases²³; Higher asymmetries were significantly associated with lower quality of life scores, measured by the knee injury and hip disability OA outcome scores (KOOS and HOOS) suggesting that IMU-based gait analysis can provide meaningful insights into biomechanical asymmetries and related quality of life measures.

This study's primary aim is to evaluate knee sagittal kinematic differences between injured

and contralateral limbs and healthy controls at pre-surgery, three and five months post-ACLR using an IMU system during slow, normal, and fast walking speeds. The secondary aim is to describe potential compensatory adaptations of hip and ankle sagittal kinematic differences during the same timepoints.

We hypothesize that ACLR patients will show significant differences in knee sagittal kinematics between their injured and contralateral limbs and when compared to healthy controls, with these differences decreasing but persisting by five months post-surgery. Additionally, we expect faster walking speeds will magnify these between-limb differences compared to normal or slow walking speeds.

96 Methods

The data for this study were obtained from a randomized clinical trial (NCT05001594) assessing the effect of local muscle vibration on the lower limb kinematics of ACLR patients.

The ---will be inserted after blind review--- Helsinki Committee (0089-21-RMB) approved this study.

Following Lakens' (2022) framework for sample size justification in resource-constrained studies derived from previous trials, we performed a sensitivity power analysis for our sample of 40 participants.²⁴ This analysis revealed that with $\alpha = 0.05$, our study had 80% power to detect effect sizes (ES) of 0.45 or larger, 90% power for ES of 0.53 or larger, and 95% power for ES of 0.58 or larger in our between-limb comparisons.

Study Design

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In accordance with the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines, we conducted a longitudinal observational study.²⁵ The study involved individuals scheduled for ACLR surgery, with assessments conducted presurgery and three and five months post-surgery. Gait kinematics at three different walking speeds were collected using an IMU motion capture system (Xsens Technologies, B.V., The Netherlands). Participants were recruited from the ---will be inserted after blind review--between 2020-2023 during their scheduled pre-surgery meetings. Gait analysis was performed at three-timepoints: pre-surgery and at three and five months post-surgery followup visits. The study included male and female participants aged 18-40 who were scheduled for an ACLR using hamstrings, bone-patellar tendon-bone (BTB), or quadriceps grafts. The surgeries took place between 2020 and 2023 and were conducted by a team of five experienced surgeons, each with 7-25 years of senior surgical experience at ---will be inserted after blind review---. Participants were excluded if they had sustained multi-ligament injuries, significant meniscus

damage, or any other concurrent injuries that altered the standard weight-bearing protocol.

Additionally, individuals with a history of previous knee surgeries or fractures in either leg,

known neuropathies, active cancer, inflammatory arthritis, or who received an injection for

pain in the last six months were not eligible for the study. Those who declined to participate were also excluded. All participants followed a standardized baseline rehabilitation protocol (Appendix 1).

Additionally, healthy participants matched by sex, weight, and height were recruited and underwent a similar gait assessment protocol in a single testing session. The inclusion criteria for the control group were males or females, aged 18-40 with no self-reported lower limb or back pain in the last six months. The exclusion criteria were a history of any lower limb or back fractures, surgeries, known neuropathies, active cancer, or inflammatory arthritis.

All the participants provided written informed consent before participating in the study.

Gait Assessment

Each participant was fitted with seven IMUs: One on each foot, one just below each knee, one on each mid-thigh, and one on the pelvis (Xsens Awinda, The Netherlands). Data was collected, automatically processed, and filtered using proprietary software at a sampling rate of 100 Hz (Xsens MVN analyze, version 2023.0.0). The system captures acceleration, angular velocity, and magnetic field data and exports them to a computer, process them using biomechanical modeling algorithms and proprietary software to compute joint angles, spatiotemporal parameters, and segment orientations in 3D.²⁶ This system was shown to be reliable compared to traditional motion capture.²⁷

Before data collection, a calibration process was performed to ensure accurate sensor

alignment. Next, the participants were asked to walk across a 20-meter corridor at three speeds: slow, normal, and fast. The following standardized verbal instructions were used: for the slow speed: "Walk across the walkway at a slow speed", "Walk across the walkway at your normal speed", and "Walk as fast as possible". 28 Each speed was repeated three times. The main discrete outcome was the knee sagittal angles, which were defined as the first peak

angle – the first peak of knee flexion during the early stance phase, the second peak angle – the peak of knee flexion occurring during the swing phase, and the minimum flexion angle – the lowest knee flexion angle reached during the gait cycle (Figure 1). The secondary outcome was the hip and ankle's minimum and maximum sagittal angles. All outcomes were averaged and used for the final analysis. The participants were asked to bring their own walking shoes and wear the same shoes at each visit.

Statistical Analysis

Descriptive statistics are presented as median [minimum, maximum]. The Wilcoxon signed ranks test with the exact method was used to compare the injured and contralateral limbs at each timepoint, and the Mann-Whitney U test was used to compare the injured limb and the left limb of the healthy participants. The Friedman test was used to assess differences between walking speeds.

We utilize the Benjamini-Hochberg procedure to control for multiple comparisons and false

discovery rates. We applied the Benjamini-Hochberg procedure separately for each aim, ensuring that the control of the false discovery rate is appropriate for each hypothesis and maintaining statistical power without the results of one aim affecting the other. We set the FDR level at 0.1, balancing the need to detect true effects.²⁹ The results are presented as mean difference (M), standard error of the difference (SE), Rank biserial correlation effect size (ES), and p-values (p).

Linear mixed-effects models were used to assess longitudinal changes in the lower limbs. Each joint angle was the independent variable, the timepoint was added as a fixed effect, and the ACLR participants were added as random effects. When significant differences were detected between timepoints, the Bonferroni post-hoc test was used for pairwise comparisons

between the different timepoints. The analyses were carried out using SPSS Statistics (version 29; IBM Corp, Armonk, NY) and Jamovi (version 2.4; The Jamovi project).

Before performing the SPM analysis, all time-series data were normalized to 101 data points, representing 0-100% of the gait cycle. The data was interpolated to 101 evenly spaced points for each gait cycle using a linear interpolation function (scipy.interpolate.interp1d). Discrete outcome measures were then extracted from these normalized gait cycles, with the knee sagittal angles defined as the first peak angle (the first peak of knee flexion during the early stance phase), the second peak angle (the peak of knee flexion occurring during the swing phase), and the minimum flexion angle (the lowest knee flexion angle reached during the gait cycle). Next, One-dimensional statistical parametric mapping (spm1d) was used to evaluate differences in the full kinematic wave during walking. The spm1d non parametric paired-sample t-test with 1000 permutations was used to assess the limb differences (version 3.7; Python Software Foundation). The alpha level for all tests was set at 0.05.

Results

Participants Demographics and Walking Speeds

Table 1 provides the demographic details of the ACLR patients and healthy participants. Knee kinematics are presented in Table 2. There was no difference in sex, age, height, weight, and graft between the participants who attended the five months follow-up and those who were lost to follow-up at five months post-ACLR (Appendix 2).

Figure 2 shows the difference in walking speeds at different timepoints. The results of the Friedman test found statistically significant differences between slow, normal, and fast walking speeds within each timepoint (all p values <.001). When comparing the same speed

- categories across timepoints, no significant differences were found in slow (p=.390), normal
- 197 (p=.113), or fast (p=.193) walking speeds, implying that it was sustained over the three
- timepoints. The hip and ankle kinematics are detailed in Appendices 3 and 4, respectively.

199 Knee Joint Pre-Surgery

- 200 Compared to the contralateral limb, the discrete analysis showed that during normal walking,
- the injured limb exhibited significantly greater knee flexion at the minimum flexion angle (M
- $= 1.7^{\circ}$, SE = 0.7, ES = 0.48, p = .006). This pattern was also present during fast walking (M =
- 1.7° , SE = 0.7, ES = 0.57, p = .008). Further, the injured knee showed lesser knee flexion at
- 204 the first peak angle (M = -1.7°, SE = 0.8, ES = -0.49, p = .006).
- The SPM analysis (Table 3) revealed that the injured knee was significantly less flexed than
- the contralateral limb across multiple portions of the gait cycle. Figure 3 shows that, during
- slow walking, differences were observed during 12-54% of the gait cycle (t = 2.48, p = .001).
- During normal walking (Figure 4), significant differences spanned 24-48% of the cycle (t =
- 2.75, p = .001). Differences were noted during fast walking (Figure 5) in two areas: 30-51%
- and 90-94% of the gait cycle (t = 2.79, p = .002 and .033, respectively).
- When comparing the injured limb to healthy controls, the knee of the ACLR patients was
- more flexed at the minimum flexion angle than controls at normal walking ($M = 2.2^{\circ}$, ES =
- 213 0.37, p = .025) and slow walking (M = 2.7° , ES = 0.38, p = .021).

Knee Joint Three Months Post-Surgery

- 215 Compared to the contralateral limb, the discrete analysis revealed that the injured limb
- 216 displayed increased knee flexion at the minimum flexion angle during slow walking (M =
- 4.3°, SE = 0.6, ES = 0.96, p < .001) and lesser knee flexion at the second peak angle (M = -
- 4.7° , SE = 0.8, ES = -0.92, p < .001). During fast walking, the injured limb had greater knee
- flexion at the minimum flexion angle (M = 4.3° , SE = 0.8, ES = 0.98, p < .001) and lesser

- knee flexion at the first (M = -5.2° , SE = 0.9, ES = -0.86, <= .001) and second peak angles
- 221 $(M = -2.2^{\circ}, SE = 0.89, ES = -0.60, p = .005).$
- The SPM analysis (Table 3) showed multiple periods where the injured knee was less flexed
- across all walking speeds. During slow walking (Figure 3), differences between limbs
- occurred across three clusters of the gait cycle: 0-3%, 19-61%, and 92-99% (t = 2.62, p =
- 225 .028, .001 and .001, respectively). Figure 4 shows differences in normal walking across 26-
- 226 59% and 90-99% of the cycle (t = 2.78, p = .001). During fast walking (Figure 5), significant
- differences spanned 30-62% and 90-97% of the gait cycle (t = 2.86, p = .001 and .008,
- 228 respectively).
- 229 Compared to controls, the injured knee remained more flexed at the minimum flexion angle
- 230 at slow (M = 5.3° , ES = 0.78, p < .001), normal (M = 5.5° , ES = 0.77, p < .001) and fast (M =
- 4.9°, ES = 0.71, p < .001) speed. Further, the knee was less flexed at the first peak during slow
- 232 walking (M = 4.6° , ES = 0.39, p = .031).

233 Knee Joint Five Months Post-Surgery

- Compared to the contralateral limb, the discrete analysis showed that the injured limb was
- significantly more flexed at the minimum flexion angles during slow ($M = 1.5^{\circ}$, SE = 0.6, ES
- 236 = 0.59, p = .027) and normal walking speeds (M = 2.1° , SE = 0.7, ES = 0.76, p = .003).
- Further, the knee was less flexed at the first peak angle (M = -1.9° , SE = 1.0, ES = -0.53, p =
- 238 .048), but this was non-significant after correction.
- Similarly, at fast walking, the injured knee was significantly more flexed at the minimum
- flexion angle (M = 2.3° , SE = 0.6, ES = 0.86, p < .001) and less flexed at the first peak angle
- 241 $(M = -5.1^{\circ}, SE = 1.0, ES = -0.83, p < .001).$
- Figure 4 and Figure 5 show that differences were now limited to 93-95% of the gait cycle
- during normal walking (t = 2.86, p = .003) and 38-61% during fast walking (t = 2.83, p = .003)

.011). The knee remained less flexed during the weight acceptance phase at fast walking than the contralateral limb, indicating that some asymmetry persisted, mostly at higher walking speeds. Compared to healthy controls, the injured knee was more flexed at the minimum angle at slow ($M = 3.2^{\circ}$, ES = 0.57, p = .003) and normal speeds ($M = 3.3^{\circ}$, ES = 0.54, p = .005).

As seen in Table 4, significant changes were observed in knee kinematics from pre-surgery to three months post-surgery. At a slow walking speed, knee flexion was significantly increased for the minimum angle (p=.006). Similar patterns were observed at normal and fast walking speeds (p<.001, p=.006, respectively).

Hip and Ankle

After correction for multiple comparisons, at pre-surgery, significant differences were observed in the injured ankle that demonstrated increased plantarflexion (slow speed, p= .003) compared to healthy controls. No significant differences were noted between the injured and contralateral limbs. By three months post-surgery, the ankle of the injured limb exhibited increased plantarflexion (slow speed, p=.002. Next, the hip showed less flexion across all walking speeds (normal and slow, p= .001. fast, p=.008) and less extension at normal speed (p= .004) when comparing the injured limb to the contralateral limb.

By five months post-surgery, the injured ankle showed significantly less dorsiflexion and increased plantarflexion at a slow walking speed (p < .05) compared to healthy controls, but those differences were not significant after correction for multiple comparisons. No differences were found in the hip joints. The detailed results of the SPM analysis are available in Table 3, and the discrete analysis is provided in Appendices 3-6.



269 Discussion

This study assessed sagittal plane kinematics during gait, pre- and post-ACLR, using IMUs in a non-laboratory setting. Our primary findings indicate progressive improvement in knee sagittal kinematics symmetry post-ACLR, particularly at three months post-surgery, with compensatory adaptations observed in hip and ankle kinematics. Few studies have examined pre- and early post-ACLR gait kinematics, the impact of walking speed on gait asymmetries, or utilized SPM in this population. 5,11,12,19,30

Pre-Surgery Knee Kinematics

The injured knee exhibited a more flexed position during most of the stance phase across all speeds and just before the weight acceptance phase at a fast speed. These results are consistent with Gao et al. (2010), who observed that patients with less than a year since their ACL injury presented with a less extended knee during the stance phase.³¹ The SPM analysis revealed that these asymmetries occurred predominantly during mid-stance and terminal stance, highlighting how weight-bearing activities specifically challenge movement symmetry even before surgery. Increased knee flexion during stance may represent a protective mechanism to reduce tibial translation and minimize ACL strain in the absence of a functional ACL.³² The persistence of this pattern across different walking speeds suggests it's a robust compensatory strategy rather than a speed-dependent adaptation.

Three Months Post-Surgery Knee Kinematics

Now extending into the swing phase, the discreet analysis found that three months following ACLR, the participants demonstrated less knee flexion at the second peak during slow and normal walking speeds in their injured limb compared to the contralateral side. During fast walking, differences between limbs extended to include reduced flexion at the first peak in the injured limb.

While Williams et al. (2023) reported reduced first peak knee flexion angle in the involved limb during self-selected walking speeds ($17.7^{\circ}\pm 6.7$ and $20.9^{\circ}\pm 4.7$), we only observed this difference during fast walking.³³ This discrepancy may be explained by walking speed differences, as their participants' self-selected speeds were faster than ours, falling between our normal and fast conditions. Further, Ferber et al. (2002) found no difference in first peak knee flexion angle at a self-selected walking speed but did not report on the actual walking speed of the participants($16.1^{\circ}\pm 2.1$ and $19.7^{\circ}\pm 5.3$).³⁴

The SPM analysis revealed that the injured knee was more flexed during most of the stance and the beginning of the swing across all walking speeds, while peak flexion during the swing was also reduced. Neal et al. (2021) used SPM to assess knee kinematics three months post ACLR and reported increased knee flexion between 31-51% of the cycle¹⁸, consistent with our findings. On the contrary, they found decreased knee flexion between 6.5% and 20% of the gait cycle. Notably, the self-selected speed in Neal's study was closer to our participants' fast walking speed, partially explaining this discrepancy.

These findings reinforce the speed-dependent nature of kinematic asymmetries post-ACLR, suggesting that movement deficits may be less apparent at slower walking speeds but become more pronounced as task demands increase. Furthermore, the late stance and terminal swing asymmetries might highlight challenges in preparing for the next gait cycle.

Five Months Post-Surgery Knee Kinematics

By five months post-ACLR, the injured limb improved, but asymmetries persisted in midstance at faster walking speeds. This speed-dependent pattern demonstrates how increasing task demands can reveal persistent movement asymmetries that might not be apparent in less challenging walking. Discreet analysis revealed that the knee was more flexed at the minimum point and less flexed at the first peak at normal and fast speeds compared to the contralateral limb. These results differ from those of Georgoulis et al. (2003)³⁵, who found no differences at any discreet point of the gait cycle with a walking speed similar to ours. One possible explanation is that Georgoulis compared two separate groups (different participants for the pre- and post-ACLR groups), whereas we performed within-subject comparisons.

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Neal et al. assessed participants six months after ACLR and found differences between 5-18% and 36-48% of the gait cycle. 18 Similarly to the three-month comparison, the selfselected speed in Neal's study was faster than ours. The differences in sample size, movement capture systems, and slightly different timepoints after the ACLR can further explain the different results.

While improvements during the first five months post-ACLR are evident, knee kinematics do not fully return to pre-injury levels even one year after surgery. Studies show that despite reconstructive efforts, peak knee flexion remains lower in ACLR limbs compared to controls, which could contribute to less-than-optimal recovery.2 Further, The persistence of asymmetries primarily during mid-stance at faster speeds might suggest that the neuromuscular system still struggles with higher mechanical demands, such as excessive tibial translation.³⁶

Associated Hip and Ankle Adaptations

As a secondary finding, we observed several adaptations in hip and ankle kinematics, possibly as a compensation strategy for the changes in the knee joint. Pre-surgery, hip asymmetries were limited to slow walking during early to mid-stance. Ankle asymmetries during this period were evident in the mid-stance when walking slowly and at normal speed. At three months post-ACLR, hip asymmetries persisted in mid-stance and late stance across all walking speeds. Similarly, ankle asymmetries were observed in mid-stance and terminal stance across all walking speeds. By five months, hip asymmetries had resolved across all speeds, while ankle asymmetries persisted during terminal stance at normal and fast walking.

The resolution of hip asymmetries by five months, compared to the persistence of ankle asymmetries, suggests a joint-specific recovery pattern. These findings suggest compensations still exist, particularly under higher demands, which can lead to higher joint loading.³⁷

Clinical Implications

The comparison of our study with others highlights the need for transparent, reproducible methodologies. As some don't report walking speeds, and others don't describe the exact verbal cue given to their participants, a heterogeneous comparison between studies is hard to make.

Our findings using IMU-based gait analysis align with previous evidence supporting the clinical utility of wearable sensors in assessing knee pathologies. Recent studies have demonstrated that IMU-derived gait parameters can discriminate between individuals with and without knee osteoarthritis. Additionally, IMU measurements have shown significant associations with patient-reported outcomes and pain levels, suggesting their potential value as objective markers of functional recovery. While previous research has primarily focused on primary knee osteoarthritis, our study extends these findings to the ACLR population, demonstrating that IMU-based gait analysis can capture subtle biomechanical changes and potentially early identification of patients at risk for PTOA development.

Our findings have important implications for post-ACLR rehabilitation protocol design and implementation. The non-linear recovery patterns across walking speeds align with previous studies showing increased asymmetries at faster speeds. 11,30,41 Our secondary findings of

compensatory mechanisms, particularly evident at higher speeds, may lead to excessive joint loading that could increase the risk of early degenerative changes in both the hip and ankle joints. ⁴¹⁻⁴⁴ Further, different walking speeds can reveal important insights: Slower walking speeds may highlight joint patterns similar to those seen in osteoarthritis, showing how ACLR patients adapt their gait. ⁴⁵ Additionally, faster walking speeds place greater demands on joint moments, muscle activation, and range of motion. ³⁰

Limitations

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We acknowledge some limitations in our study. While our sample size was determined by a previous trial rather than an a-priori power calculation, sensitivity power analyses demonstrated our study had sufficient power (80%) to detect effect sizes of 0.45 or larger, aligning with the effects we observed (ranging from 0.48 to 0.98). We used a single-center design with a limited follow-up period. However, the study aimed to describe the kinematic changes during the pre-ACLR and early to middle stages of the ACLR rehabilitation process. We did not account for different graft types and sex differences, possibly introducing a bias in our results. Further, the participants available for this study had varied times between their injury and ACLR, which can influence their biomechanics and ACLR outcomes. Next, Multiple comparisons of hip, knee, and ankle joint kinematics might increase type I errors. However, we pre-defined the knee joint kinematic analysis as the primary outcome of this study and adjusted the results accordingly. Additionally, a high drop-out rate may introduce attrition bias, potentially linked to participants missing pre-defined check-up meetings with their surgeon rather than the study itself. Lastly, although there were no statistically significant demographic differences between the healthy control group and the ACLR cohort, the healthy participants were older, shorter, and weighed less, which could have influenced their gait.

In conclusion, our study highlights several key findings regarding ACLR patients' gait recovery. We observed symmetry improvements in gait post-surgery compared to the contralateral limb and controls. However, these improvements were inconsistent across different walking speeds, suggesting that varying walking speeds may challenge patients differently. While hip and ankle adaptations were noted, the primary focus on knee kinematics revealed persistent asymmetries at faster walking speeds. These findings underscore the importance of integrating varied speed regimens in rehabilitation protocols to facilitate comprehensive recovery of knee function.

Key Points

- ACLR patients showed significantly improved symmetry in knee flexion angles at three and five months post-surgery during various walking speeds.
- Significant asymmetries in knee, hip, and ankle kinematics were observed presurgery, indicating altered movement patterns that persisted post-surgery.
- The non-linear recovery process across different walking speeds highlights the need for assessing and incorporating varied walking speeds in rehabilitation protocols.

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Figure Legend

Figure 1. Knee sagittal movement angle during the gait cycle of healthy participants, together with commonly analyzed discrete points

Normalized gait cycle of a healthy participant. Orange = Stance phase. Blue = Swing phase. Vertical dashed line = The point between the stance and swing phase.

Figure 2. Median walking speed for each walking condition grouped by timepoint

Each violin plot represents the probability density of the data at different speed values, with the width of the violin indicating the frequency of observations at each speed. Black lines within the violins indicate the median speed and interquartile range of each group. Overlaid on the violin plots are individual data points (gray circles), representing the inter-participant differences in speed values.

Figure 3. One Dimension Statistical Parametric Mapping of the Gait Cycle: Comparison between Injured and Contralateral Limbs during Slow Walking at Pre-ACLR, and Three and Five Months Post-ACLR

The shaded grey boxes equal statistical significance difference. Yellow line = ACLR limb. Blue line = Contralateral limb

Figure 4. One Dimension Statistical Parametric Mapping of the Gait Cycle: Comparison between Injured and Contralateral Limbs During Normal Walking Speed at Pre-ACLR, and Three and Five Months Post-ACLR

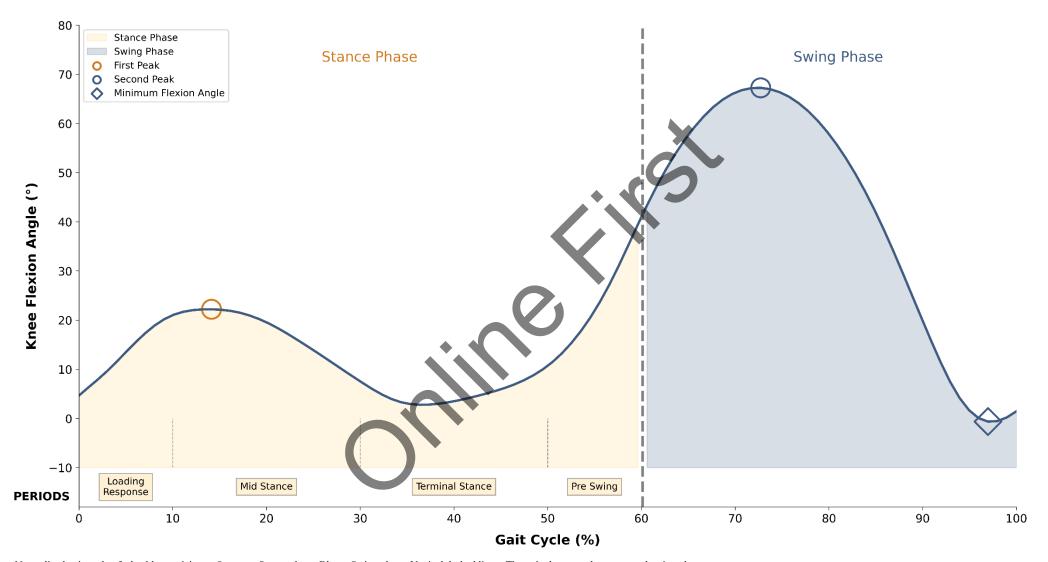
The shaded grey boxes equal statistical significance difference. Yellow line = ACLR limb. Blue line = Contralateral limb

Figure 5. One Dimension Statistical Parametric Mapping of the Gait Cycle: Comparison between the Injured and Contralateral Limbs During Fast Walking Speed at Pre-ACLR, and Three and Five Months Post-ACLR

The shaded grey boxes equal statistical significance difference. Yellow line = ACLR limb. Blue line = Contralateral limb

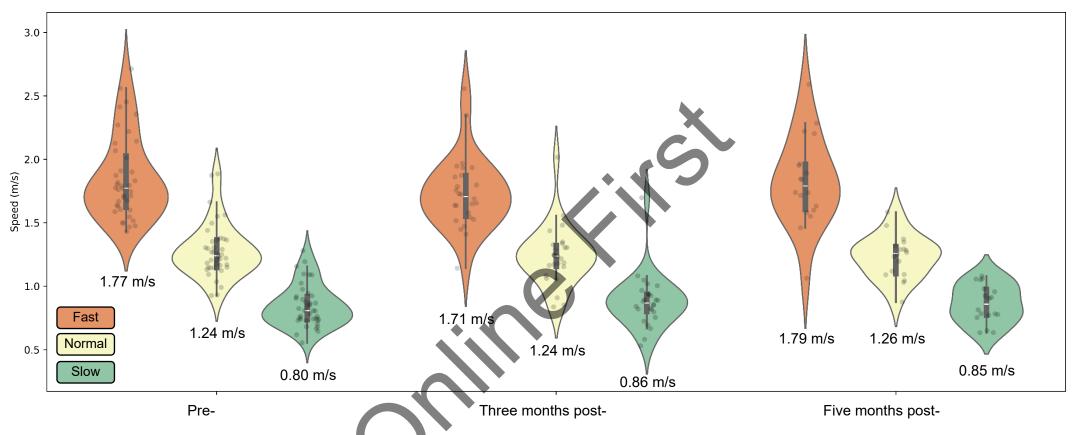


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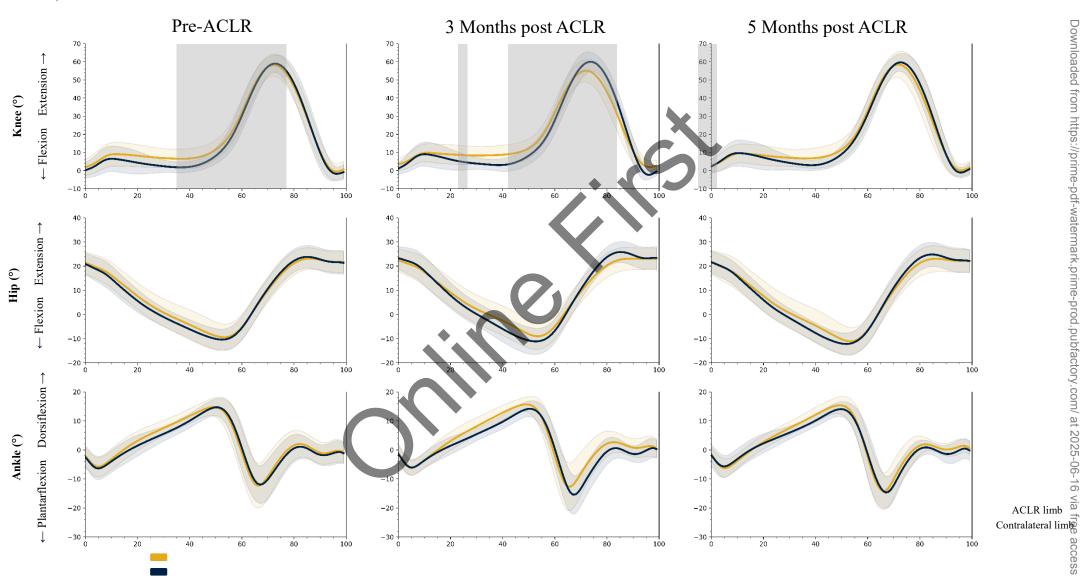
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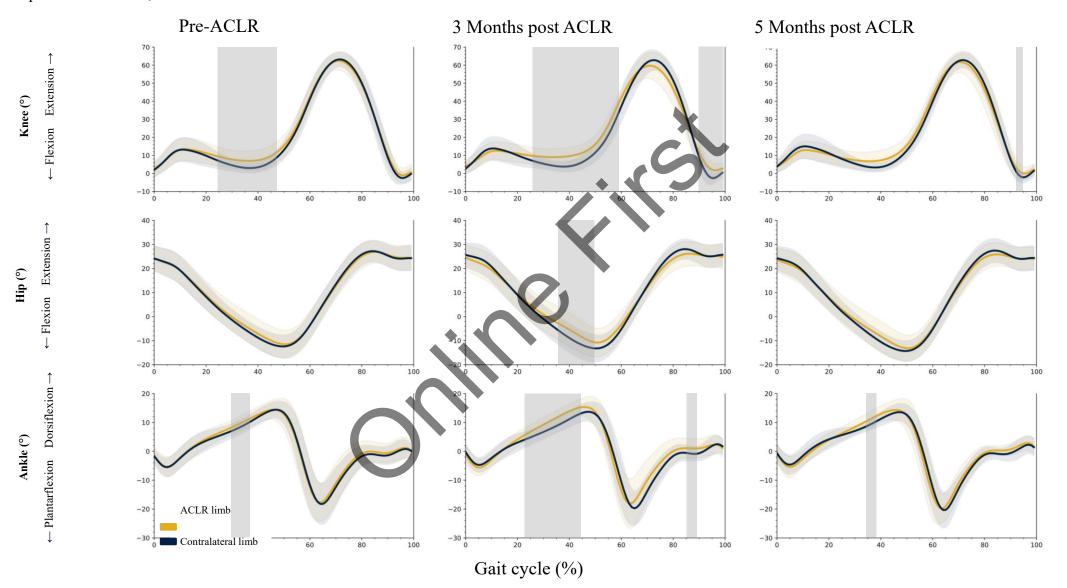
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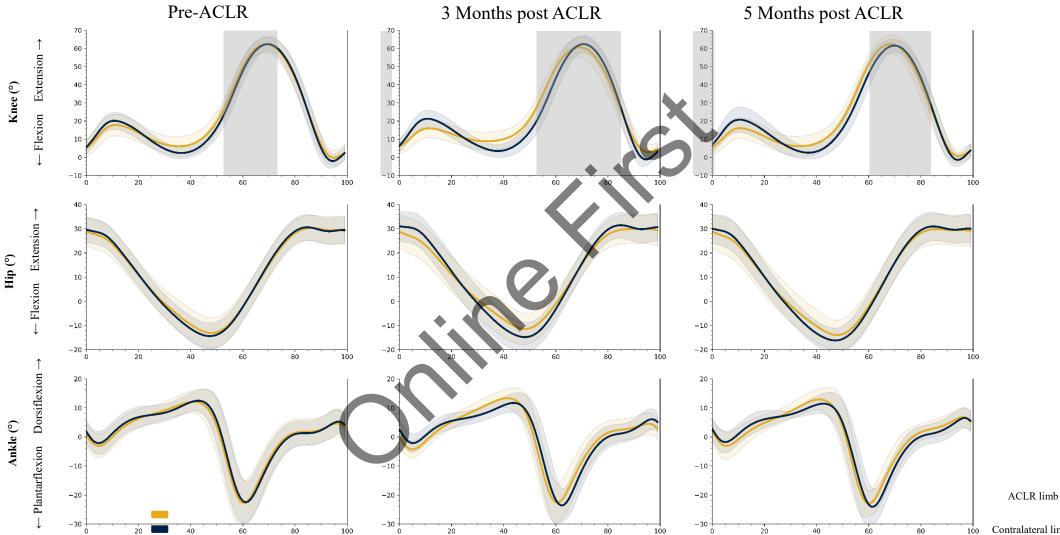
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Figure 5. One Dimension Statistical Parametric Mapping of the Gait Cycle: Comparison between the Injured and Contralateral Limbs During Fast Walking Speed at Pre-ACLR, and Three and Five Months Post-ACLR.



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100

The shaded grey boxes equal statistical significance difference. Yellow line = ACLR limb. Blue line = Contralateral limb



Table 1. Demographic of the Participants

	Participants with ACLR (n=40)	Healthy Participants (n=17)	p
Age (years)	23.9 ± 5.9	28.4 ± 5.9	.008
Sex (%)			
Males	33 (82.5)	12 (70.5)	.090
Females	7 (17.5)	5 (29.5)	
Height (m)	1.76 ± 0.1	1.70 ± 0.1	.052
Weight (kg)	76.3 ± 13.4	68.3 ± 15.4	.094
Injured Leg (%)			
Left	18 (45%)	N/A	N/A
Right	22 (55%)		
Time between injury and	191 [45-698]	NT/A	NT/A
reconstruction (days)		N/A	N/A
Graft (%)			
Hamstrings	26 (65%)		
ВТВ	8 (20%)	N/A	N/A
Quadriceps	5 (12.5%)		
Allograft	1 (0.02%)		

 χ^2 Goodness of Fit. Independent samples T-test.

Table 2. Description of the Knee Sagittal Movements While Walking at Three Different Speeds at Three Timepoints Comparing the Involved Limb to the Contralateral Limb and a Healthy Cohort.

Walking Pace	Angle Control (n=17)					Pre-surgery (n=40)			Three months post-surgery (n=27)			Five months post-surgery (n=18)		
		Left (°)	Right (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value	
Slow	Minimum	-3.8 [-10.2, 1.8]	-3.7 [-9.6, 1.7]	.243	-1.3 [-7.5, 14.6]	-3.5 [-10.8, 5.5]	.053*	1 [-5.2, 7.9]	-3.5 [-7.9, 1.3]	<.001§*	-1.8 [-5.2, 6.9]	-2.7 [-6.5, 3]	.027§*	
	First Peak	5.4 [-6.2, 22.5]	7.1 [-3.6, 21.7]	.080	9.57 [-4.1, 21.6]	7.2 [-5, 21.5]	.068	11.2 [0.1, 22]	10.5 [-1.4, 23.5]	.628*	10 [-0.6, 21]	11.5 [-3, 21.4]	.609	
	Second Peak	58.6 [48.1, 65.7]	59.5 [52.7, 66.3]	.225	58.7 [44.8, 71.7]	59.9 [53.7, 69.2]	.062	55.9 [38.5, 66.6]	59.7 [53.7, 70]	<.001§	58.6 [48.3, 73.1]	59.5 [54.1, 70.9]	.369	
Normal	Minimum	-4.3 [-9.4, -0.6]	-3.0 [-10.8, 1.1]	.031§	-2.1 [-8, 14]	-3.9 [-11.7, 4.2]	.006§*	1.3 [-4, 9]	-4.2 [-8.2, 0.6]	<.001§*	-1.5 [-6.5, 6.6]	-3.2 [-7.4, 1.5]	.003§*	
	First Peak	13.7 [-2.6, 28.1]	13.4 [-2.6, 28.1]	.225	14.5 [-4, -25.3]	14.7 [-3.6, 27.9]	.618	13.1 [2.5, 23.8]	15 [-1.8, 24.6]	.258	13 [4.3, 21.6]	16.5 [0.7, 25.4]	.048§	
	Second Peak	60.7 [56.4, 69.4]	61.4 [54.3, 71.0]	.263	63.3 [49.3, 71]	63.2 [58.5, 71.5]	.132	60.2 [37.1, 71.7]	63.1 [55, 73.9]	.001§	62.6 [51.1, 73.8]	61.8 [57.7, 72]	.417	
Fast	Minimum	-2.9 [-12.7, 1.7]	-1.6 [-10.3, 2.8]	.132	-1.5 [-7, 12.4]	-3 [-11.1, 4.5]	.008§	1.8 [-3.1, 14.7]	-2.7 [-7.3, 3.6]	<.001§*	-0.6 [-5.7, 7.9]	-3.1 [-7.3, 2.2]	<.001§	
	First Peak	18.6 [5.2, 33.2]	21.8 [-1.0, 30.9]	.089	18.8 [-4.4, 28.9]	20.3 [9.8, 29.9]	.006§	17.1 [4.1, 29.8]	21.6 [12.2, 29.7]	<.001§	17 [2.2, 25.4]	21.5 [-0.2, 31.1]	<.001§	
	Second Peak	60.5 [55.2, 74.5]	59.8 [52.9, 70.4]	.782	63.4 [48.4, 71.4]	62.6 [53.7, 71.6]	.590	61.1 [51.7, 75.3]	61.9 [55.5, 71.2]	.005§	62.4 [54.1, 73.7]	61.3 [57.7, 70.2]	.468	

Angles are presented in degrees (°).

Comparison between limbs was done using the exact Wilcoxon signed-rank test.

Comparison between the injured limb of the ACLR group and the left leg of the control group was done using the exact Mann-Whitney U test.

The p value between represent the comparison between contralateral and involved limb

[§] Denotes a difference between the injured/non injured or right and left limbs at a single time point.

^{*}Denotes differences between the injured limb at the chosen time and the left leg of the control.

Table 3. Results of the non-parametric SPM1d{t} Analysis for Differences Between the Injured and Contralateral Limb During Walking at Three Different Speeds by Joint and Time Post-ACLR.

Walking Pace	Joint	Pre su (n=-			Three months (n=		ry	Five months post-surgery (n=18)			
	Joint	Clusters (% Cycle)	t value	p value	Clusters (% Cycle)	t value	p-value	Clusters (% Cycle)	t value	p value	
	Knee	(12,54)	2.48	.001	(0, 3) (19,61) (92,99)	2.63	.028 .001 .001	-	-	-	
Slow	Hip	(12,37)	2.56	.012	(24, 50)	2.68	.004	-	-	-	
	Ankle	(20,43)	2.69	.001	(19,48) (67,91)	2.89	.001 .020	-	-	-	
	Knee	(24, 48)	2.75	.001	(26, 59) (90,99)	2.78	.001 .001	(93, 95)	2.86	.003	
Normal	Hip	-	-	-	(34, 47)	2.71	.012	-	-	-	
	Ankle	(29, 36)	2.79	.032	(23,44) (85,88)	2.88	.001 .045	(34, 37)	2.98	.040	
				11							
	Knee	(30, 51) (90,94)	2.79	.002	(30,62) (90,97)	2.86	.001 .008	(38,61)	2.83	.011	
Fast	Hip	-		-	(34,57)	2.75	.006	-	-	-	
	Ankle	-	-		(27,41)	2.85	.001	(33,39)	3.03	.023	

The results of non-parametric paired t-tests using the One-Dimension Statistical Parametric Mapping (SPM1d). A permutation-based approach with 1000 iterations was used for cluster-level. Significant clusters were identified based on an alpha level of 0.05. '-' indicates no significant difference between limbs. A positive t value indicates a higher angle for the injured limb. Only statistically significant differences are reported in this table.

Table 4. Description of Changes in the Knee Sagittal Movements Between the Different Timepoints Before and After ACLR

Walking Pace	Angle	Pre surgery three months pos		Three months five months post-s		Pre surgery to five months post-surgery		
		Difference (°)	p value	Difference (°)	p value	Difference (°)	p value	
	Minimum	-2.5 ± 0.7	.006	2.2 ± 0.8	.045	-0.3 ± 0.9	1	
Slow	First Peak	-1.0 ± 0.9	.771	1.0 ± 0.7	.411	0.1 ± 1.0	1	
	Second Peak	3.0 ± 1.1	.037	-2.9 ± 1.4	.142	0.1 ± 1.2	1	
	Minimum	-2.9 ± 0.7	<.001	2.2 ± 0.8	.55	-0.7 ± 0.9	1	
Normal	First Peak	0.1 ± 0.9	1	0.3 ± 0.5	1	0.4 ± 0.9	1	
	Second Peak	3.0 ± 1.2	.067	-1.8 ± 1.5	.685	1.1 ± 0.9	.730	
	Minimum	-3.1 ± 0.9	.006	2.7 ± 1.0	.042	-0.4 ± 1.0	1	
Fast	First Peak	1.2 ± 1.0	.716	0.45 ± 1.0	1	1.6 ± 1.0	.426	
	Second Peak	1.8 ± 0.9	161	-1.6 ± 0.9	.305	0.1 ± 0.7	1	

[‡] Denotes significant differences between timepoints. Angles are presented in degrees (°).

Appendix 1. Baseline Rehabilitation Protocol

First week post-op:

- Static quadriceps exercises in straightening.
- Walking with crutches/walker while applying partial weight on the operated leg.
- Wearing elastic socks and exercising calf muscles to avoid deep vein thrombosis.

7 - 14 days post-op:

- Walking while fully stepping on the leg (can use crutches)
- Exercising to improve range of movement.
- After 2-3 weeks, maximal recommended range of movement is 0-90

Week 2-3:

- Wall slides & Heel Slides exercises.
- Static VMO exercises sitting down.
- Using rowing machine with minimal resistance and medium range of movement.

Week 4:

- Bending exercises 0-100 degrees (avoid over straightening).
- Static bicycles with low resistance.
- Up and down exercises with a low step.
- Squat exercises up to 45-60 degrees.

Week 8:

- Step machine.
- Squat exercises.

Week 10:

- Trampoline.

Week 12:

- Achieving a range of movement of 0-130 degrees is recommended.
- Advancing in strengthening quadriceps & hamstrings.

3-6 months:

- Increasing gradual activity.
- Easy running starting on the 5th month.
- Changing directions, accelerations, deceleration, running in the shape of 8.
- Preparing for a specific sport.

After 6 months:

- If everything is normal and the patient feels confident to return to the specific sport, gradually exercise the specific sport.

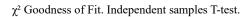
Between 7-9 months:

- Returning to the specific sport.



Appendix 2. Demographics of the Participants at Five Months Follow-up Compared to the Participants That Was Lost for Follow-up at Five Months

	Participants at five months post-op (n=18)	Participants who were lost to follow-up at five months (n=22)	p
Age (years)	24.1 ± 6.6	23.6 ± 4.6	.796
Sex (%)			
Males	13 (72.2)	20 (90.9)	.211
Females	5 (27.8)	2 (9.1)	
Height (m)	1.76 ± 0.1	1.76 ± 0.1	.929
Weight (kg)	74.1 ± 12.6	75.7 ± 13.0	.684
BMI (kg/m²)	23.9 ± 3.3	24.6 ± 4.5	.597
Injured Leg (%)			
Left	6 (33.3)	10 (45.5)	.526
Right	12 (66.7)	12 (54.5)	
Graft (%)			
Hamstrings	11 (61.1)	15 (68.2)	
BTB	3 (16.7)	5 (22.7)	.698
Quadriceps	3 (16.7)	2 (9.1)	
Allograft	1 (5.6)	0(0)	



Appendix 3. Description of Changes in the Hip Sagittal Movements Between the Different Timepoints Before and After ACLR

Walking Pace	Angle	Pre surger three months pos		Three month five months post-		Pre surgery to five months post-surgery		
		Difference (°)	p value	Difference (°)	p value	Difference (°)	p value	
Slow	Minimum	0.7 ± 1.2	1	1.5 ± 0.6	.123	2.2 ± 1.2	.262	
Slow	Maximum	-0.4 ± 0.4	.829	0.4 ± 0.5	1	-0.1 ± 0.5	1	
Normal	Minimum	0.1 ±1.4	1	1.6 ± 1.1	.496	1.7 ± 1.0	.268	
TOTMAT	Maximum	-0.1 ± 0.3	1	0.7 ± 0.4	.362	0.6 ± 0.4	.430	
Fast	Minimum	-1.0 ± 1.5	1	1.1 ± 1.1	.958	0.1 ± 1.2	1	
Fast	Maximum	-0.1 ± 0.4		-0.4 ± 0.5	1	-0.4 ± 0.6	1	

Angles are presented in degrees (°).

[‡] Denotes significant differences between timepoints.

Appendix 4. Description of Changes in the Ankle Sagittal Movements Between the Different Timepoints Before and After ACLR

Walking Pace	Angle	Pre surger three months pos		Three month		Pre surgery to five months post-surgery							
		Difference (°)	p value	Difference (°)	p value	Difference (°)	p value						
Slow	Minimum	-1.0 ± 0.6	.395	1.8 ± 0.8	.119	0.8 ± 0.7	.828						
Slow	Maximum	-0.2 ± 0.8	1	-0.3 ± 1.0	1	-0.6 ± 1.0	1						
Normal	Minimum	-1.0 ± 0.7	.506	1.8 ± 0.8	.104	0.8 ± 0.7	.907						
rvormar	Maximum	0.1 ± 0.8	1	-0.3 ± 0.9	1	-0.1 ± 0.8	1						
Fast	Minimum	-2.0 ± 0.8	.061	1.9 ± 1.1	.304	-0.1 ± 1.0	1						
1 ast	Maximum	-0.4 ± .08	1	-0.6 ± 1.4	1	-1.1 ± 1.3	1						

Angles are presented in degrees (°).

[‡] Denotes significant differences between timepoints.

Appendix 5. Description of the Hip Sagittal Movements While Walking at Three Different Speeds at Three Timepoints Comparing the Involved Limb to the Contralateral Limb and a Healthy Cohort.

Comparison between limbs was done using the exact Wilcoxon signed-rank test.

Comparison between the injured limb of the ACLR group and the left leg of the control group was done using the exact Mann-Whitney U test.

Walking Pace	Angle		Control (n=17)		Pre-surgery (n=40)			Three months post-surgery (n=27)			Five months post-surgery (n=18)		
		Left (°)	Right (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value
	Minimum	-11.2 [-27.6, 20.3]	-11.7 [-24.0, -5.1]	.071	-10.2 [-20.2 – 4.1]	-10.3 [-17.90.6]	.277	-10.6 [-17.8 – 4.2]	-10.7 [-25.34.3]	.052	-11.8 [-21 – 4.8]	-13.3 [-23.85.9]	.369
Slow	Maximum	25.4 [6.5, 31.4]	26.7 [13.9, 32.1]	.031 [§]	24.6 [15.1-34.9]	24.6 [16-37.1]	.277	24.3 [15.6 – 32.1]	25 [16.5 – 37.7]	<.001 [§]	23.1 [15.4 – 33.4]	23.3 [18.8 – 36.6]	.130
	Minimum	-13.2 [-26.3, -8.1]	-12.9 [-24.8, -7.2]	.080	-11.8 [-23.2 -4.85]	-12 [-24.63.5]	.202	-11.9 [-21.3 – 2.2]	-11.3 [-27.45.2]	.030§	-14.5 [-21 – 2.7]	-13.9 [-26.7 – 14.3]	.966
Normal	Maximum	28.6 [12.6, 34.8]	29.9 [17.0, 35.2]	.071	26.9 [18.6-38.8]	27.4 [19.3-41.5]	.295	27.6 [18.1 – 34.9]	27.9 [20–39.7]	<.001 [§]	26 [17.9 – 35.2]	26.5 [22.4 – 38.4]	.142
Fast	Minimum	-14.0 [-28.5, -9.3]	-13.6 [-25.5, -9.4]	.098	-14.2 [-26.9 -4.6]	-13.4 [-28.42.3]	.166	-13 [-23.7 – 2.2]	-13.6 [-32.17.4]	.004 [§]	-15.4 [-22.6 – 3.6]	-15.8 [-29.77.2]	.054
2 431	Maximum	34.3 [15.1, 42.4]	35.4 [16.7, 43.6]	.017 [§]	30.7 [18.5-43.8]	30.2 [20.9-44.6]	.375	30.5 [18.9 – 45.5]	31.4 [22.6 – 44.1]	.008§	31.5 [15.1 - 46.2]	32.6 [18.8 - 46.2]	.130

Angles are presented in degrees (°).

Comparison between limbs was done using the exact Wilcoxon signed-rank test.

Comparison between the injured limb of the ACLR group and the left leg of the control group was done using the exact Mann-Whitney U test.

[§] Denotes a difference between the injured/non injured or right and left limbs at a single time point.

^{*}Denotes differences between the injured limb at the chosen time and the left leg of the control.

Appendix 6. Description of the Ankle Sagittal Movements While Walking at Three Different Speeds at Three Timepoints Comparing the Involved Limb to the Contralateral Limb and a Healthy Cohort.

Comparison between limbs was done using the exact Wilcoxon signed-rank test.

Walking Pace	Angle		Control (n=17)	Pre-surgery (n=40)			Three months post-surgery (n=27)			Five months post-surgery (n=18)			
		Left (°)	Right (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value	Involved (°)	Contralateral (°)	p value
	Minimum	-20.3 [-44.7, -9.8]	-17.4 [-36.7, -7.5]	.005§	-13.4 [-30, 1]	-13.9 [-27.1, -0.2]	.858*	-16.1 [-27, -2.15]	-18.6 [-26.3, -5.4]	.046 ^{§*}	-17.1 [-28.7, -7.6]	-17.2 [-27.6, -7.2]	.495*
Slow	Maximum	14.0 [7.7, 17.9]	14.5 [4.5, 20.2]	.431	15.5 [7.3, 21.8]	14.7 [7.4, 21.8]	.590*	16.5 [11.6, 19.9]	14.8 [8.8, 19]	.016§*	15.5 [9.7, 21]	14.3 [11, 19.6]	.108*
	Minimum	-22.0 [-44, -15.2]	-21.2 [-41.1, -12.4]	.145	-18.7 [-33.8, -4.9]	-18.2 [-36.2, 16.9]	.921	-20.5 [-38.9, -4]	-22.2 [-32.8, -9.2]	.386	-20.1 [-35.1, -14.2]	-22.1 [-30.1, -11]	.671
Normal	Maximum	13.7 [10.6, 20.6]	14.8 [7.2, 21.1]	.548	16.1 [7.2, 21]	14.9 [6.6, 20.6]	.572	16.2 [10.7, 21.2]	14.4 [7.5, 18.8]	.044 [§]	15.4 [8.6, 20.6]	14.5 [8.1, 19.2]	.304
Fast	Minimum	-27.6 [-46.7, -17.2]	-24.8 [-45.2, -17.8]	.263	-24.1 [-46.5, -7.3]	-23.4 [-41.8, -13]	.973	-24.7 [-36.9, -8.5]	-28.7 [-33.7, -14.7]	.100	-24.8 [-30.1, -18.6]	-27.6 [-36.1, -18.9]	.284
rast	Maximum	12.9 [5.5, 18.0]	13.0 [8.4, 19.3]	.353	13 [6.8, 22.9]	13.1 [8.4, 19.9]	.666	13.7 [6.9, 20.6]	12.9 [6.3, 17.7]	.141	14.2 [8.6, 21.1]	12.3 [8.6, 17.3]	.090

Angles are presented in degrees (°).

Comparison between limbs was done using the exact Wilcoxon signed-rank test.

Comparison between the injured limb of the ACLR group and the left leg of the control group was done using the exact Mann-Whitney U test.

[§] Denotes a difference between the injured/non injured or right and left limbs at a single time point.

^{*}Denotes differences between the injured limb at the chosen time and the left leg of the control.