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Original Research

The lateral meniscus extrudes with and without root tear evaluated using ultrasound

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ABSTRACT

Purpose: The purpose of the current study was to measure extrusion of the intact lateral meniscus as a function of knee flexion angle and loading condition and to compare the changes in extrusion with a posterior root tear using a robotic testing system and ultrasound.
Study design: Controlled laboratory study.
Methods: Eight fresh-frozen cadaveric knees were subjected to external loading conditions (passive path position (no external load), 200 axial compression, 5-N-m internal tibial torque, 5-N-m valgus torque) at full extension, 30°, 60° and 90° of flexion using a robotic testing system. A linear array transducer was placed in the longitudinal orientation. Extrusion and kinematics data were recorded for two meniscus states: intact and posterior lateral root deficiency. Therefore, a complete radial root tear in the lateral meniscus at 10 mm from the tibial insertion was made in all 8 cadaveric knees using arthroscopy. The resultant forces in the lateral meniscus were also quantified by reproducing recorded paths after the removal of the lateral meniscus.
Results: A lateral meniscus root tear resulted in a statistically significant increase (up to 250%) of extrusion for the lateral meniscus ($p < 0.05$) in comparison to the intact lateral meniscus for all externally applied loads. Without external load (passive path position), significant differences were also found between the intact and posterior lateral root deficient meniscus except at full extension (1.0 ± 0.7 mm vs. 1.9 ± 0.4 mm) and 30° of flexion (1.4 ± 0.5 mm vs. 1.8 ± 0.5 mm). Overall, with increasing flexion angle, lateral meniscus extrusion decreased for the intact as well as for the posterior lateral root deficient meniscus, with the lowest measurements in response to internal tibial torque at 90° of flexion (-3.3 ± 1.1 mm). Knee kinematics were similar whether intact or posterior lateral root tear (n.s.). Ultrasound measurement of lateral meniscus extrusion showed good inter-rater (0.65 [0.30–0.97]–0.71 [0.34–0.94]) and excellent intra-rater reliability (0.81 [0.43–0.99]).
Conclusion: Dynamic Ultrasound is a reliable diagnostic modality to measure the lateral meniscus extrusion which can be helpful in the diagnosis and quantification of lateral meniscal root tears.
Level of evidence: III.

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What are the new findings

In this study, the intact lateral meniscus extruded in response to external loading. Lateral meniscus root tear resulted in significantly increased meniscal extrusion in response to all loading conditions, compared to the intact lateral meniscus.

Introduction

Radial tears located within 1 cm from the meniscal attachment or root avulsions are defined as meniscal root tears [1]. The prevalence of meniscus posterior root tears is reported to be 5.9% for medial and 3.5% for lateral root tears [2]. From a biomechanically perspective, this injury leads to compromised hoop stresses resulting in decreased tibiofemoral contact area and increased contact pressures in the involved compartment [3,4]. Additionally, a posterior root tear resulted in rotatory instability of the knee when concomitant with an anterior cruciate ligament (ACL) injury, suggesting its pivotal role in normal knee biomechanics [5,6]. Various repair techniques have been reported to improve clinical outcomes, decrease meniscal extrusion and slow the onset of degenerative changes, but the reported healing rate is low with persistent meniscus extrusion in 40% and failure rate between 15 and 34% [7,8].

The current standard method to evaluate the meniscus is magnetic resonance imaging (MRI) with the patient supine, the knee fully extended and no external loads applied. Especially for lateral meniscus root tears, MRI is associated with a decreased sensitivity of 0.600 (in comparison sensitivity for medial meniscus root tear 0.824), indicating a higher risk for overseen lateral meniscus root tears [2]. Meniscal extrusion has been described as an indirect sign of meniscal pathology and osteoarthritis [9–11]. However, meniscus extrusion greater than 3 mm in MRI can be detected only in 20% of lateral meniscus root tears [12].

Previously, an age- and load- dependent physiologic dynamic extrusion for the healthy medial meniscus has been evaluated using ultrasound [9–11]. Further studies demonstrated an ultrasound measurement of the medial meniscus extrusion to be a reliable option to assess intact medial meniscus function and the effect of a medial meniscus root tear [11]. Most previous ultrasound studies pertain to medial meniscus likely due to the relative ease of visualising the medial meniscus [13–15]. Measuring extrusion may help physicians to assess the influence of a tear on knee kinematics preoperatively, as well as the success and status of the repair postoperatively. Although lateral meniscus extrusion has previously been assessed in lateral meniscus transplants [16,17], normal lateral meniscus extrusion in response to posterior root tear at various knee flexion angles under various loading conditions remain unknown.

Therefore, the aim of the present study was to validate and assess the use of ultrasound to quantify lateral meniscus extrusion under externally applied loading conditions including axial compression, valgus torque and internal rotation torque (simulating bipolar standing and valgus alignment) using a robotic testing system. It was hypothesised that lateral meniscus extrusion is a sign of loss of function of the lateral meniscus, determined by the increased medial tibial translation and internal rotation. Based on the results of the medial meniscus, it was also hypothesised that increased lateral meniscus extrusion would be present in the posterior lateral deficient root state with axial compression, valgus torque and increased flexion angle (0–90°).

Material and methods

Specimen preparation

Eight fresh frozen human knees (mean age 57.6 ± 4.7 years old; range 53–66 years) were used in this study after the approval from the ethical oversight committee on decedents (CORID number 501). Specimens were stored at −20 °C and were thawed at room temperature for 24 h before testing [18]. The tibia and femur were cut 20 cm from the joint line and specimens were examined manually and arthroscopically before testing to exclude any specimens with ligamentous, meniscal or osseous abnormalities. The fibula was fixed to the tibia using a tricortical screw to maintain its anatomic position. The femur and tibia were potted in an epoxy compound (Bondo. 3M. Minnesota, USA) and secured within custom-made aluminium clamps. The knee was mounted in a robotic testing system.

The robotic testing system (MJT model FRS2010, Technology Service Ltd., Chino, Japan) consists of a 6-degree-of-freedom manipulator and a universal force-moment sensor (Delta IP60 [SI-660-60] and ATI Industrial Automation, North Carolina, USA) that is utilised to provide feedback to the controller. Control of the system is accomplished through a LabVIEW program (National Instruments) designed for knee-joint biomechanical testing and utilises hybrid velocity impedance control. The position and orientation repeatability of the robotic manipulator are less than ±0.015 mm and ±0.01°. The measurement uncertainty of the universal force sensor (UFS) is approximately 1% of full scale [19].

Test protocols

The 6 degree-of-freedom path of passive flexion-extension for the intact knee from full extension to 90° of knee flexion was initially determined [20]. The positions of zero forces and moments across the joint through the whole range of motion were determined as the path of passive flexion-extension. Four loading conditions were then applied to the intact knee and the resulting kinematics were recorded: (1) Passive path position (no external load), (2) 200N of axial compression, (3) 5-Nm internal tibial torque and (4) 5-Nm valgus torque applied at full-extension, 30°, 60° and 90° of knee flexion (Table 1, Kinematics A). Each load was applied three times and afterwards the lateral meniscus extrusion was measured. The order of the loading conditions was changed randomly for each specimen to minimise the effect of order.

Meniscus extrusion and kinematics data were recorded at each flexion angle for each loading condition. Then a posterior lateral meniscus root tear was created arthroscopically by transecting the meniscus tissue at 10 mm lateral from the tibial insertion. The same set of data was recorded for the meniscus with a posterior lateral root tear (Table 1, Kinematics B). Afterwards, the lateral meniscus was completely resected utilising a 3 cm anterolateral incision and previously recorded kinematics A and B were repeated to record new forces and moments. The resultant force within the lateral meniscus was determined by the principle of superposition [21–23].

Table 1
Test protocol and data acquisition.

Status of knee	Test protocol	Acquired data
Intact knee	1. Passive path position (no external load)	Meniscus extrusion
	2. Valgus Torque = 5Nm	
	3. Internal Tibial Torque = 5Nm	Intact knee motion (A)
	4. Axial Compression = 200N	
	Ultrasonographic evaluation	Meniscus extrusion
Injured knee -posterior root deficient knee-	1. Passive path position (No external load)	Meniscus extrusion
	2. Valgus Torque = 5Nm	
	3. Internal Tibial Torque = 5Nm	Injured knee motion (B)
	4. Axial Compression = 200N	
	Ultrasonographic evaluation	Meniscus extrusion
Injured knee -total lateral meniscectomy-	Replay knee kinematics A and B	Resultant force of the lateral meniscus

Ultrasound measurement

All ultrasound measurements were performed using a 4–15 MHz broad-spectrum linear matrix array transducer (LOGIQ S8, GE Healthcare, GE, USA). Prior to ultrasound assessment, the lateral femoral epicondyle, Gerdy's tubercle and fibular head were palpated and marked on the skin. A trans-tibial ACL drill guide (Acufex Director; Smith & Nephew Arthroscopy, Andover, MA, USA) was placed at the lateral side of the tibia, posterior to the Gerdy's tubercle. Then a k-wire was drilled from the medial side to the posterior half of Gerdy's tubercle about 3–5 mm below the tibial plateau. To improve the repeatability of the ultrasound measurements, the landmark was enlarged with a 2.8 mm reamer. This guide mark was created to ensure a consistent transducer position throughout testing, due to the variable anatomy of the lateral femoral condyle.

After the external load was applied the third time, the linear transducer was placed in an oblique longitudinal orientation, straddling over the lateral femoral condyle proximally to the guide mark (Fig. 1). To reduce attenuation, the centre frequency was set at 8 MHz for improved penetration and two forces were used as a default. The lateral meniscus was visible as a triangular, echogenic structure between the lateral femoral condyle and the tibial plateau. For each external loading condition, a 3–5 s cine loop was recorded. Lateral meniscus extrusion was defined as the distance between two parallel lines, tangent to the margin of the lateral tibial cortex and the outermost edge of the lateral meniscus (Fig. 1) [9,24].

Three orthopaedic surgeons with 2–9 years of experience with musculoskeletal ultrasound, who were blinded to the meniscus status and applied condition, independently measured the lateral meniscus extrusion in each video. The lateral meniscus extrusion for each video was measured one time by three different observers (ST, RT, TD) to determine inter-rater reliability and three times by one observer (TD) to assess intra-rater reliability.

Statistical analysis

Intraclass correlation coefficients were calculated to determine the consistency and reproducibility of the extrusion measurements. The inter-rater reliability coefficient was calculated using values from the ANOVA table corresponding to a model with a fixed-rater effect and a random subject and subject*rater effect using the formula given by $\widehat{\rho}_{intra,fixed}$ in Eliasziw et al. [25] (SAS 9.4, SAS Institute Inc, North Carolina, USA). The same model was used to calculate the intra-rater reliability coefficient given by $\widehat{\rho}_{intra,fixeg}$ in Eliasziw et al. [25]. The average extrusion of the three raters (TD, ST and RT) was taken as the true extrusion value for each loading condition, flexion angle and meniscus state. The Shapiro–Wilk was used to test for normality of the data. The difference between the intact and root tear state was tested using a linear mixed model with a random specimen effect. The difference was tested for each combination of flexion angle and loading condition and the Holm-Bonferroni method was used to adjust for multiple testing. A Wilcoxon signed rank test was used to test for differences between the intact and root tear state for each combination of flexion angle and loading condition. A linear mixed model with a random specimen effect was used to test differences between the four loading conditions (unloaded, axial, internal and valgus) for each combination of flexion angle and meniscus state (intact or root tear) and the Holm-Bonferroni method was employed to adjust for multiple comparisons.

Results

Extrusion for the intact lateral meniscus in comparison to the posterior lateral root tear was significantly different in response to 200N axial tibial load, 5 Nm valgus tibial torque and 5 Nm internal tibial torque at 0°, 30°, 60° and 90° of flexion ($p < 0.05$). With the increasing flexion angle, lateral meniscus extrusion significantly decreased for the intact (Fig. 2A) and posterior lateral root tear (Fig. 2B) meniscus in response to all applied loads.

Ultrasound lateral meniscus extrusion in response to 200 N axial tibial load

With the increasing flexion angle, the lateral meniscus extrusion for the intact and posterior lateral deficient meniscus decreased in response

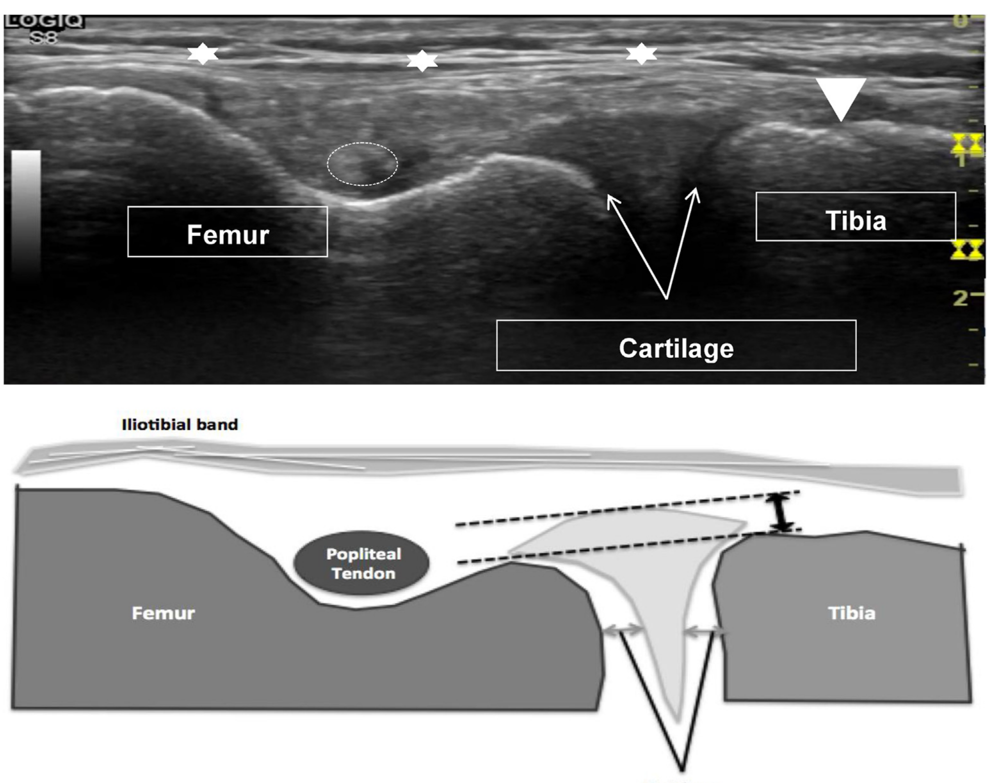

Fig. 1. A: Optimal ultrasound image for analysis: The image shows: (1) clear margins of both tibial and femoral cartilage (white arrows), (2) popliteal hiatus (popliteal tendon white circle), (3) lateral femoral epicondyle, and (4) posterior half of Gerdy's tubercle denoted by the guide mark (white arrow head). The stars mark the iliotibial band. **B**: Schematic drawing of an optimal ultrasonographic image. A black arrow corresponds to the measured lateral meniscus extrusion. The measurement was performed using a built-in caliper on the ultrasound machine (LOGIQ S8, GE Healthcare, Milwaukee, WI, USA).

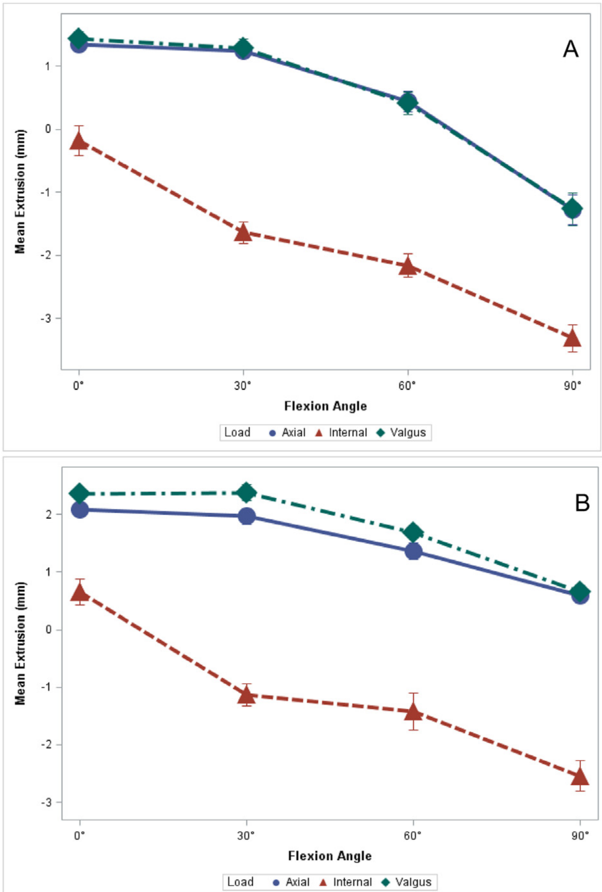

Fig. 2. Mean lateral meniscus extrusion for the intact (**A**) and posterior lateral meniscus tear (**B**) at 0°, 30°, 60° and 90° of flexion in response to 200 N axial tibial load, 5 Nm valgus tibial torque and 5 Nm internal tibial torque. Ultrasound lateral meniscus extrusion in response to 200 N axial tibial load.

Table 2
Lateral meniscus extrusion for the intact and posterior lateral deficient meniscus at 0°, 30°, 60° and 90° of flexion in response to 200 N axial tibial load.

Flexion angle	Lateral meniscus extrusion (mean ± SD; mm)		Holm-Bonferroni adjusted p-value
	Intact	Posterior lateral root tear	
0°	1.4 ± 0.6	2.1 ± 0.4	0.004
30°	1.2 ± 0.6	2.0 ± 0.7	0.004
60°	0.4 ± 0.8	1.4 ± 0.7	<0.002
90°	−1.3 ± 1.1	0.6 ± 0.6	<0.002

to 200 N axial tibial load (Table 2). Posterior lateral meniscus tear resulted in significant higher extrusion for all flexion angles (up to 250%) in comparison to the intact meniscus.

Ultrasound lateral meniscus extrusion in response to 5 Nm valgus tibial torque

Tibial valgus torque at full extension in the posterior lateral deficient meniscus resulted in the highest lateral meniscus extrusion for all tested conditions (Table 3). In comparison to lateral meniscus extrusion in response to axial tibial load, extrusion in response to valgus torque was not significantly different ($p > 0.05$).

Ultrasound lateral meniscus extrusion in response to 5 Nm internal tibial torque

Internal tibial torque at 90° of flexion in the intact meniscus resulted in the lowest lateral meniscus extrusion for all tested conditions

Table 3
Lateral meniscus extrusion for the intact and posterior lateral meniscus tear at 0°, 30°, 60° and 90° of flexion in response to 5 Nm valgus tibial torque.

Flexion angle	Lateral meniscus extrusion (mean ± SD; mm)		Holm-Bonferroni adjusted p-value
	Intact	Posterior lateral root tear	
0°	1.4 ± 0.6	2.4 ± 0.5	<0.002
30°	1.3 ± 0.7	2.4 ± 0.7	<0.002
60°	0.4 ± 0.8	1.7 ± 0.5	<0.002
90°	−1.3 ± 1.3	0.6 ± 0.5	<0.002

(Table 4). Lateral meniscus extrusion in response to internal tibial torque was significantly different from extrusion in response to axial tibial load and valgus tibial torque, respectively ($p < 0.05$). In comparison to axial tibial load and valgus tibial torque, internal tibial torque resulted in the lowest extrusion measurements.

Ultrasound lateral meniscus extrusion without external load (position)

Without external load (passive path position), again there was a significant difference for the lateral meniscus extrusion between the intact and the posterior lateral deficient meniscus at 60 and 90 degree of flexion but not at full extension and 30° of flexion (Table 5). Comparison of extrusion in passive path position to axial tibial load or valgus tibial torque revealed no significant difference, except for lateral meniscus extrusion in response to valgus tibial torque for the posterior lateral deficient meniscus at 30° of flexion (33.3%, $p = 0.02$).

Repeatability

Inter-rater intra-class correlation coefficient (ICC) was moderate averaged for individual knee flexion angles and states. ICC between the three observers was similar for the overall measurements (ST vs. RT 0.65 [0.30–0.97]; TD vs. RT 0.68 [0.29–0.91]; TD vs. ST 0.71 [0.34–0.94]). Intra-rater ICC was good as determined using three measurements by a single observer (TD 0.81 [0.43–0.99]). Inter-rater ICC for the posterior lateral meniscus tear was greater than in the intact state (Intact 0.84 [0.67–0.99]; root tear 0.78 [0.43–0.97]).

Kinematics

Posterior lateral deficient meniscus resulted in increased medial tibial translation compared to the intact meniscus at all flexion angles, but the difference was not significant (n.s.). The highest medial tibial translation was demonstrated at 90° of flexion for the posterior lateral deficient meniscus (5.7 ± 4.0 mm) in response to internal tibial torque.

With increasing flexion angle, valgus rotation of the tibia increased in response to 200 N axial compression, 5 Nm valgus torque and 5 Nm internal tibial torque, respectively. In response to 5 Nm valgus torque, valgus angulation of the knee was highest at 90° of flexion, for the intact lateral meniscus (2.9° ± 1.3°) as well as for the posterior lateral deficient meniscus (4.1° ± 1.1°). Overall, posterior lateral deficient meniscus resulted in increased valgus rotation at all flexion angles in response to 5 Nm valgus torque compared to the intact meniscus, but the difference

Table 4
Lateral meniscus extrusion for the intact and posterior lateral meniscus tear at 0°, 30°, 60° and 90° of flexion and in response to 5 Nm internal tibial torque.

Flexion angle	Lateral meniscus extrusion (mean ± SD; mm)		Holm-Bonferroni adjusted p-value
	Intact	Posterior lateral root tear	
0°	−0.2 ± 1.2	0.7 ± 1.1	0.002
30°	−1.6 ± 0.8	−1.1 ± 0.9	0.058
60°	−2.2 ± 0.9	−1.4 ± 1.6	0.004
90°	−3.3 ± 1.1	−2.5 ± 1.3	0.002

Table 5
Lateral meniscus extrusion for the intact and posterior lateral meniscus tear at 0°, 30°, 60° and 90° and in passive path position (no external load).

Flexion angle	Lateral meniscus extrusion (mean ± SD; mm)		Holm-Bonferroni adjusted p-value
	Intact	Posterior lateral root tear	
0°	1.0 ± 0.7	1.9 ± 0.4	0.058
30°	1.4 ± 0.5	1.8 ± 0.5	0.313
60°	0.5 ± 1.0	1.4 ± 0.6	0.044
90°	−1.3 ± 1.4	0.4 ± 0.8	<0.002

was not significant (n.s.).

5 Nm internal tibial torque resulted in no significant difference in internal tibial rotation between the intact and posterior lateral meniscus deficient state (n.s.).

The resultant force in the lateral meniscus was highest in full extension in response to 200 N axial compression (105.5 ± 43.8 N). Overall, posterior lateral deficient meniscus led to decreased resultant forces in the lateral meniscus, but only the difference in resultant force in response to 5 Nm valgus torque for higher flexion angles (60°: 77.9 ± 33.9 N to 37.2 ± 29.1 N; 90°: 63.4 ± 26.5 N to 32.1 ± 27.6 N) was significant different between the intact lateral meniscus and posterior lateral root tear state.

Discussion

The most important finding of the present study is that the intact lateral meniscus demonstrates an extrusion in response to external loading, as already shown for the medial meniscus [10,26]. Posterior lateral root tear resulted in significantly increased meniscal extrusion in response to all external loading conditions compared to the intact lateral meniscus. In contrast, for the passive path position (no external load) at full extension and 30°, no significant difference between intact and posterior lateral deficient meniscus was found. In opposition to the original hypothesis, with increasing flexion angle, lateral meniscal extrusion significantly decreased for the intact as well as for the posterior lateral deficient meniscus, with the lowest measurements in response to internal tibial torque at full flexion. Knee kinematics were similar whether intact or posterior lateral root tear.

In comparison to the medial meniscus extrusion, the measurements for the lateral extrusion resulted in lower repeatability measurements [26]. Beside the more C-shaped appearance, the lateral meniscus is also not directly attached to the lateral joint capsule and has a higher mobility within the knee joint [14,27]. During knee flexion, the lateral meniscus is moving posteriorly and medially between 4 and 9 mm with the anterior horn moving the most [28]. The lateral meniscus is pulled inside the knee joint and the soft tissue between the lateral meniscus and the capsule make it harder to define its border. In the posterior lateral deficient meniscus, the distance between the meniscus and capsule decreased and the border could be identified more easily, which is reflected by the increased ICC for the posterior deficient extrusion measurements. The extrusion measurements are also influenced by the bony morphology and reference landmarks. On the lateral side, the transducer is placed more anterolateral due to the fibula head, straddling over the lateral femoral epicondyle and the popliteal hiatus. The hiatus popliteus has a broad variety in deep and size and is affected by osteoarthritis changes [29] and thereby influences the lateral extrusion measurements. In contrast, the bony anatomy of the medial tibiofemoral joint demonstrates less bony variability and this could be another explanation for the lower ICC on the lateral side.

In terms of mobility during flexion and extension, the lateral meniscus moves almost twice the distance of the medial meniscus [27]. The greatest posterior translation of the lateral meniscus is present during flexion in the anterior horn of the lateral meniscus, 6.3 mm in the non-weight bearing knee and 9.5 mm under full weight bearing [28]. Negative extrusion of the meniscus at higher flexion angle could therefore be explained by the anterior transducer position and the increased posterior translation of the lateral meniscus. Although several studies reported a significant contribution to knee joint stability for the lateral meniscus in the ACL deficient or ACL reconstructed knee [30,31], the knee kinematics were not significantly affected by the posterior lateral meniscus root tear with an intact ACL. Similar to the results of present study, the effect of different lateral meniscus allograft techniques on knee joint stability was examined and showed no significant difference for internal rotation in the ACL intact knee after lateral meniscectomy [32].

From a clinical point of view, posterior lateral meniscus root tears are mostly caused traumatically [33,34]. A lateral root tear concomitant with an ACL injury is observed in up to 12% of patients [35,36]. In general, a complete posterior root tear results in a significant increase in lateral meniscus extrusion measured on MRI compared to the intact state, but with a lower cut-off value (1.1 mm) than the medial side (3 mm) [37,38]. Interestingly, a partial posterior root tear of the lateral meniscus did not cause a significant increase of meniscus extrusion (partial 0.4 mm vs. intact – 0.1 mm) [37]. In a recent MRI study, an extrusion greater 3 mm was found in over 70% of medial meniscus root tears, but only in 20% of lateral meniscus root tears [12]. In line with this work, the present study demonstrated no difference of the lateral meniscus extrusion at full extension and 30° of flexion at the passive path position (no external load), but significantly increased meniscal extrusion quantified with ultrasound in response to all loading conditions. Based on this, the predictive value of lateral meniscus extrusion might be improved by including loaded measurements as well as the difference between loaded and unloaded measurements (Δ-Extrusion). Moreover, internal tibial torque of 5 N-m resulted in the lowest lateral meniscus extrusion compared to all other external loading conditions and was able to almost completely neutralise the elevated lateral meniscus extrusion in the posterior root deficient state. An internally rotated knee during MRI examination might therefore also result in an underestimated lateral meniscus extrusion.

It is acknowledged that this study has some limitations. First, current results are based on a time-zero cadaveric study with middle-aged specimens and did not consider muscle activity nor the healing seen following a meniscal root tear. The complete posterior lateral root tear was created arthroscopically, but the mensicofemoral ligaments were not considered. The applied external loading conditions were taken due to previous biomechanical studies [32], but do not consider full body weight transmitted across the knee.

Conclusion

The intact lateral meniscus demonstrates an extrusion in response to external loading, as shown for the medial meniscus. A posterior lateral deficient meniscus resulted in significantly increased meniscal extrusion in response to all external loading conditions at full extension and all degrees of flexion and even without external load (the passive path position) except at lower flexion angles (full extension and 30° of flexion). With increasing flexion angle, the lateral meniscal extrusion significantly decreased for the intact as well as for the posterior root cut meniscus, with the lowest measurements in response to internal tibial torque. Knee kinematics were similar between intact or posterior lateral root tear states. Ultrasound evaluation of the lateral meniscus under load can be a helpful tool to clinically assess meniscus function.

Conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Not applicable

Ethical approval

Approval was obtained from the Committee for Oversight of Research and Clinical Training Involving Decedents (CORID #501) from the University of Pittsburgh.

Consent to participate

Not applicable.

Consent to publish

Not applicable.

Authors contributions

TD, KO, RED and VM conceived and planned the experiments. TD, RET, ST, TS and CC carried out the experiments. TD, KW, KO and RED contributed to the interpretation of the results. TD, RET and VM. took the lead in writing the manuscript. All authors provided critical feedback and helped shape the research, analysis and manuscript.

Availability of data and materials

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

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