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Am. J. Sports Med. 2008; 36; 2158 originally published online Jul 31, 2008;
DOI: 10.1177/0363546508320572

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Effect of Tunnel-Graft Length on the Biomechanics of Anterior Cruciate Ligament–Reconstructed Knees

Intra-articular Study in a Goat Model

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Background: In anterior cruciate ligament (ACL) reconstruction using hamstring grafts, the graft can be looped, resulting in an increased graft diameter but reducing graft length within the tunnels.

Hypothesis: After 6 and 12 weeks, structural properties and knee kinematics after soft tissue ACL reconstruction with 15 mm within the femoral tunnel will be significantly inferior when compared with the properties of ACL reconstruction with 25 mm in the tunnel.

Study Design: Controlled laboratory study.

Methods: In an intra-articular goat model, 36 ACL reconstructions using an Achilles tendon split graft were performed with 15-mm (18 knees) and 25-mm (18 knees) graft length in the femoral tunnel. Animals were sacrificed 6 weeks and 12 weeks after surgery and knee kinematics was tested. In situ forces as well as the structural properties were determined and compared with those in an intact control group. Histologic analyses were performed in 2 animals in each group 6 and 12 weeks postoperatively. Statistical analysis was performed using a 2-factor analysis of variance test.

Results: Anterior cruciate ligament reconstructions with 15 mm resulted in significantly less anterior tibial translation after 6 weeks ($P < .05$) but not after 12 weeks. Kinematics after 12 weeks and in situ forces of the replacement grafts at both time points showed no statistically significant differences. Stiffness, ultimate failure load, and ultimate stress revealed no statistically significant differences between the 15-mm group and the 25-mm group.

Conclusion: The results suggest that there is no negative correlation between short graft length (15 mm) in the femoral tunnel and the resulting knee kinematics and structural properties.

Clinical Relevance: Various clinical scenarios exist in which the length of available graft that could be pulled into the bone tunnel (femoral or tibial) could be in question. To address this concern, this study showed that reducing the tendon graft length in the femoral bone tunnel from 25 mm to 15 mm did not have adverse effects in a goat model.

Keywords: double bundle; outcome; revision; ACL reconstruction; failure; rupture; tendon-to-bone healing

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No potential conflict of interest declared.

Reconstruction of the anterior cruciate ligament (ACL) with autologous tendon grafts is a well-accepted operative technique and aims to reestablish normal knee function.^{5,7} During the early postoperative period, the fixation of the graft to the bone tunnel is the primary factor in limiting early rehabilitation.^{23,24} With increasing time, healing of the graft to the tunnel plays a more and more important role to the fixation strength.^{1,23,24}

Various clinical scenarios exist in which the length of available graft that could be pulled into the bone tunnel (femoral or tibial) could be in question. For example, when a surgeon chooses to reconstruct the ACL with a hamstring graft, the graft can be looped to a quadruple graft to increase the diameter, thereby shortening the graft. As an alternative, the graft can be used as a triple graft with a smaller diameter but resulting in a longer graft length. Because the intra-articular length of the ACL is fixed and may not be influenced by the reconstruction technique, the graft length automatically affects the length of the graft within the tunnels.^{14,15,34,35} Likewise, this problem may also be important when the surgeon considers reconstructing the ACL in an anatomic approach, restoring the anteromedial and posterolateral bundle of the ACL separately using a double-bundle technique.^{3,14,22,28,30,31} Because of the short length of the femoral posterolateral bundle tunnel, the resulting graft length in this tunnel is approximately 15 mm.

Currently, no scientific rationale for the graft length within the femoral tunnel in ACL reconstruction can be found in the literature. Therefore, the aim of the current study is to determine if fixation strength of a soft tissue autograft at 6 and 12 weeks after implantation is adversely affected by reducing the length of tendon within a bone tunnel from 25 mm to 15 mm. To accomplish this, we performed ACL reconstructions in an intra-articular model in goats with 15 mm and 25 mm in the femoral tunnel and evaluated the knees using histologic analysis, stability measurements, in situ force assessments, and tensile testing after 6 and 12 weeks. We hypothesize that at 6 and 12 weeks after implantation, the properties after soft tissue ACL reconstructions with 15 mm within the femoral tunnel will be significantly inferior when compared with the properties of ACL reconstruction with 25 mm in the tunnel.

MATERIALS AND METHODS

Animal Model and Study Groups

The animal experiment was performed following the guidelines of the National Institutes of Health for animal care and was approved by the University of Pittsburgh's Institutional Animal Care and Use Committee. A total of 36 ACL reconstructions were performed in skeletally mature female Nubian goats (average weight, 46.3 ± 6.8 kg) in a bilateral approach.⁴ The goat model was chosen because ACL reconstructions have been deemed successful out to 3 years with very low rates of graft failure or severe cartilage damage.^{10,12,13} Animals were sacrificed 6 weeks and 12 weeks after surgery. In group 1 (n = 18), 15 mm of the graft was pulled into the femoral tunnel; in group 2 (n = 18), 25 mm was pulled into the femoral tunnel (Figure 1). Two separate control groups were used for this study: 1 control group (n = 5) to determine the kinematic data for the ACL-intact and ACL-deficient knees and 1 control group (n = 5) to determine the structural properties of the ACL.

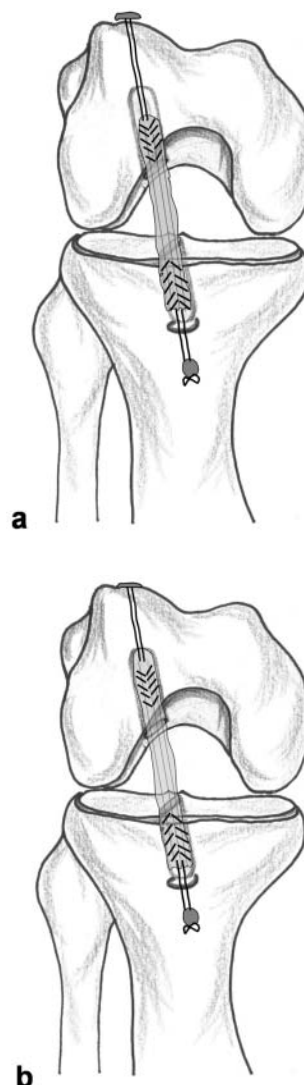


Figure 1. In group 1 (a), 15 mm of graft was pulled into the femoral tunnel; in group 2 (b), 25 mm was pulled into the femoral tunnel.

Surgical Technique

All surgical procedures were performed using sterile techniques, with the animals under general anesthesia. The knees of 9 animals per group underwent reconstruction of the ACL in an open fashion using an Achilles tendon split autograft. The Achilles tendon split graft was used as a soft tissue graft because the hamstring tendons in the goat model are too short and too thin to be a suitable autograft.^{9,23} Through a posterolateral skin incision, half of the Achilles tendon, approximately 6 cm in length, was harvested and grafts were sized to 6-mm diameter using sizing tubes. Both ends of the grafts were sewn in a baseball-stitch technique using a No. 5 FiberWire (Arthrex, Naples, Florida).

The knee joint was then exposed through an anteromedial incision. A tunnel of 5-mm diameter was drilled into the tibia at a 55° angle in the sagittal plane using a tibial aimer (Acufex, Smith & Nephew, Andover, Massachusetts). The tunnel was subsequently dilated to a diameter of 5.5 mm. With use of a 6-mm offset guide (Acufex, Smith & Nephew), a femoral tunnel (diameter, 5 mm) with a length of 25 mm was created in the 1 o'clock position in maximum knee flexion, leaving through an anteromedial portal position. Afterward the femoral tunnel was dilated to 5.5 mm and an EndoButton drill (Smith & Nephew Endoscopy, Andover, Massachusetts) was used to penetrate the cortex, and 15 mm (group 1) or 25 mm (group 2) of the graft was pulled into the femoral tunnel. Then the graft was fixed on the lateral femoral condyle using an EndoButton (Smith & Nephew Endoscopy). Before tibial fixation, a spring scale was fixed to the holding sutures at the tibial site and the graft was tensioned during 15 cycles of full knee flexion-extension under a constant load of 35 N. Afterward, the graft was fixed at the tibial site using a screw and washer.

Postoperatively, free cage activity was allowed (cage area, 3 m²) and the gait was documented as described previously (grade 0, 3-legged walking; grade 1, lameness during walking; grade 2, lameness during running; grade 3, bare lameness during running; and grade 4, no lameness during running).⁹ Six weeks after the ACL reconstruction on the 1 hind limb, the contralateral ACL was reconstructed.⁴ If the ligament was reconstructed with a 15-mm graft length during the first surgery, a graft length of 25 mm was used in the contralateral knee. Thus at the time point of sacrifice after 12 weeks from the first surgery, 1 leg represents the healing of 6 weeks whereas the other leg was subjected to 12 weeks of healing. After sacrifice, the hind limbs were disarticulated at the hip joint and stored at -20°C sealed in double plastic bags.²⁷

Robotic/Universal Force-Moment Sensor Testing System

To determine the knee kinematics, a testing system for knee kinematics that combines robotic technology with a universal force-moment sensor (UFS) was used as described previously.^{11,14,16,19,20,26,32,33} The robot (Stäubli RX-90, Stäubli Tec-Systems GmbH, Bayreuth, Germany) is a 6-joint, serially articulated manipulator that allows 6 degree of freedom movement of the knee (Figure 2). The robotic manipulator is capable of achieving positional control of the knee in 6 degrees of freedom, while the UFS can measure 3 forces and 3 moments along a Cartesian axis system with a repeatability of 0.2 N for forces and 0.01 N·m for moments. Simultaneously, this system is capable of operating in a force-controlled mode via the force feedback from the UFS to the robot.^{19,26}

Testing Protocol

Before testing, the specimens were thawed for 24 hours at room temperature.²⁷ During preparation and testing, the specimens were kept moist with 0.9% saline. The



Figure 2. Test set-up of the kinematic testing using a robotic/universal force-moment sensor testing system. The tibias of the specimens were fixed to the end effector of the robot via a custom-made clamp and the femur was mounted to the base of the robot.

tibia and femur were cut to a length of 10 cm, and surrounding tissue was dissected approximately 5 cm proximal and distal to the joint line. Bones were secured within thick-walled aluminum cylinders using 2-part epoxy resin (Bondo Corporation, Atlanta, Georgia). The knees were mounted to the robotic/UFS testing system, with the femoral cylinder fixed to the base of the robot a custom-made clamp and the tibial cylinder connected through a UFS (Figure 2).

The path of passive flexion-extension served as the starting point for application of external loads. Anterior-posterior displacement was used to simulate a clinical Lachman test, and anterior drawer examination was used to diagnose ACL deficiency. To perform anterior translation tests, the robot moved the joint to the desired flexion angle and applied an external anterior tibial loading of 67 N to the specimen at 30°, 60°, 75°, and 90° of flexion, while allowing 5 degree of freedom motion of the knee. The force of 67 N was used because these forces were reported to occur during trotting of adult goats.⁸

To determine the in situ forces in the ACL at time 0, all soft tissue structures except the ACL in and around the knee were dissected. To prevent a load transfer through a bone-to-bone contact, the articulating joint surfaces of the femoral condyles were removed. The robotic manipulator then repeated the previously determined kinematics of the intact knee in a position-control mode, while the UFS recorded a new set of force and moment data. The UFS records directly the in situ forces that are in the ACL as it is the only structure attaching the femur to the tibia. Knee kinematics and in situ forces of the reconstructed knees 6 and 12 weeks after reconstructions were performed as described above.

Cross-sectional Area Measurements

Before tensile testing, measurements of the cross-sectional area were performed in a noncontact mode. As a control group to the ACL replacement grafts of group 1 and 2 after 6 and 12 weeks, 5 intact ACLs were measured. The cross-sectional area and shape of the ACL replacement grafts were measured using a laser micrometer system (Digibot III, Digibotics, Austin, Texas).²⁵ Cross-sectional area and shape measurements were obtained at the midsubstance of the ACL replacement graft locations. Cross-sectional area measurements were averaged over 5 measurements with a distance of 1 mm.

Tensile Testing

To determine the structural properties of the femur-graft-tibia complex, the femur and tibia were cut 10 cm proximal and distal to the joint line. All tests were performed at room temperature using a material testing machine (ATM, Toronto, Ontario, Canada) and the specimens were moistened with saline solution buffer during mounting and testing. Load-to-failure tests were performed using a custom-made apparatus that gripped the femoral and tibial bones and allowed load transfer along the axis of the graft to simulate a worst-case scenario. To ensure that the tensile properties of the femoral graft-bone interface are measured, the femoral fixation device was removed before tensile testing.

Reconstructed knees were loaded to failure at a rate of 20 mm/min. The resulting load-elongation curve was recorded and the ultimate failure load was documented. Stiffness was determined as the linear region of the load-elongation curve and ultimate stress as the gradient of ultimate failure load and cross-sectional area. As a control group, the structural properties of 5 intact goat ACLs were determined.

Histologic Analysis

Two animals per group were used for histologic evaluation. The femur-graft complex was harvested, leaving the tendon-bone interface intact. A graft-bone block from the femur was sawed and soaked in decalcifying solution (ethylene diamine tetra-acetic acid [EDTA], 20%) for 3 to 5 days. The blocks were embedded in paraffin and

transverse sections were stained with hematoxylin and eosin. The specimens were sectioned parallel to the longitudinal axis of the femoral bone tunnel. Sections were made at the graft-bone interface. The histologic sections were analyzed with polarized light microscopy, and differences in the graft-bone interface were observed between groups.

Statistics

To assess the effect of healing time and knee flexion on the resulting knee kinematics and in situ forces in the intact ACL or the graft, statistical analysis was performed using a Student *t* test. A *P* value of < .05 was considered to be significant. A Statistica sampling plan (StatSoft Inc, Tulsa, Oklahoma) was used to perform a power analysis and determine a suitable number of specimens.¹⁰ The plan was set to detect a 30% difference from the mean values for stress as a measure of mechanical integrity, and anterior-posterior translation as a measure of functional integrity. The power analysis revealed that to detect a 30% difference with a power of 50%, 7 animals used for biomechanical analysis would be required.

RESULTS

In study group 1 (15 mm), 1 animal had to be excluded for anesthesia issues not related to the surgical intervention directly after the contralateral ACL reconstruction. For this animal, only the 6-week result could be reported. Additionally, because of a surgical error, 1 goat received 25-mm grafts in both knees and a mechanical error during testing resulted in the loss of 1 knee in each group. As a result of these complications, the total number of knees tested in each group were as follows: group 1 (15 mm), 6 weeks *n* = 7, 12 weeks *n* = 8; and group 2 (25 mm), 6 weeks *n* = 9, 12 weeks *n* = 8.

Recovery and Weightbearing

For all animals, with the exception of 1 that died from anesthesia-related issues, the arthrotomies and graft harvest sites healed without complications. Weightbearing of the animals improved from a mean grade of 1.2 (\pm 0.7) in group 1 (15 mm) and 1.4 (\pm 0.9) in group 2 (25 mm) 1 week after surgery to 3.5 (\pm 1.9) and 3.2 (\pm 2.0), respectively, after 4 weeks. Before surgery of the contralateral side, all animals showed a normal gait pattern. After the second intervention, mean weightbearing was 1.0 (\pm 0.8) and 1.1 (\pm 0.6) after 1 week in groups 1 and 2, respectively. After 4 weeks, the mean weightbearing improved to 3.2 (\pm 1.0) in group 1 and 3.4 (\pm 1.4) in group 2. Before sacrifice, all animals showed normal gait patterns.

Gross inspection of the knee joints at harvest showed no signs of failure of the grafts. The grafts were covered by synovial membrane including a mild inflammation. This inflammation was more pronounced at 6 weeks than at 12 weeks. No chondral or meniscal lesions were found in the 6-week and 12-week specimens.

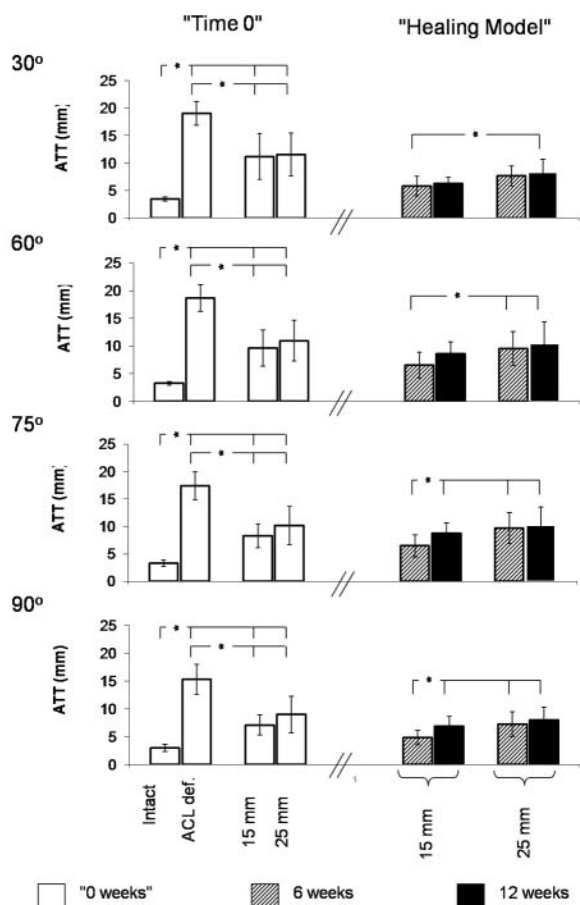


Figure 3. Anterior tibial translation (ATT) in mm under 67-N anterior tibial force. Data are shown for the healing specimens (15 mm 6 weeks [n = 7], 15 mm 12 weeks [n = 8], 25 mm 6 weeks [n = 9], and 25 mm 12 weeks [n = 8]), compared with a control group (n = 5) (intact/anterior cruciate ligament-deficient [ACL def]). Asterisks indicate statistically significant differences ($P < .05$).

Knee Kinematics

In the control group, anterior tibial translation (ATT) of the intact knee under 67-N anterior tibial load was a mean of 3.5 (± 0.4) at 30°, 3.2 (± 0.3) at 60°, 3.3 (± 0.5) at 75°, and 3.0 (± 0.6) at 90° of knee flexion (Figure 3). After transection of the ACL, the ATT significantly increased to a mean of 19.1 (± 2.1) at 30°, 18.7 (± 2.4) at 60°, 17.4 (± 2.6) at 75°, and 15.3 (± 2.7) at 90°. Anterior cruciate ligament reconstructions at time 0, as well as in the healing model in both groups, significantly reduced the increased ATT at all flexion angles ($P < .05$) (Figure 3).

After 6 weeks of healing with a graft length of 15 mm in the femoral tunnel (group 1), the ATT under 67-N anterior tibial load was a mean of 5.8 (± 1.8), 6.5 (± 2.3), 6.5 (± 2.0), and 4.8 (± 1.3) at 30°, 60°, 75°, and 90° of knee flexion, respectively (Figure 3). After 6 weeks of healing, the ATT of specimens reconstructed with 25 mm in the femoral tunnel (group 2) showed a mean ATT of 7.7 (± 1.9), 9.5 (± 3.1),

9.7 (± 2.8), and 7.2 (± 2.2) at 30°, 60°, 75°, and 90° of knee flexion, respectively (Figure 3). At this time point, the ATT of ACL reconstruction with 15 mm in the femoral tunnel (group 1) was statistically significantly lower than ACL reconstructions with 25 mm in the femoral tunnel at 60° ($P = .049$), 75° ($P = .023$), and 90° ($P = .025$) of knee flexion ($P < .05$). There was also a trend of increased ATT at 30° ($P = .064$) of flexion; however, this was not statistically significant. The ATT of both groups was statistically significantly higher than that in the intact control group (Figure 3).

After 12 weeks of healing, the ATT of specimens in group 1 was a mean of 6.3 (± 1.0) at 30°, 8.6 (± 2.2) at 60°, 8.8 (± 1.9) at 75°, and 6.9 (± 1.7) at 90°, whereas ATT of specimens in group 2 was a mean of 8.2 (± 2.5) at 30°, 10.4 (± 4.0) at 60°, 10.1 (± 3.4) at 75°, and 8.0 (± 2.3) at 90° (Figure 3). At this time point, the ATT of reconstructions in group 1 (15 mm) showed a trend of lower ATT compared with reconstructions of group 2 (25 mm); however, no statistically significant differences were recorded (30°, $P = .066$; 60°, $P = .292$; 75°, $P = .348$; and 90°, $P = .307$). After 12 weeks of healing, the ATT of reconstructions with 15-mm or 25-mm groups was statistically significantly higher than that in the intact control group (Figure 3).

The in situ forces of the intact ACL as measured in the control group were 65.5 (± 9.4) at 30°, 71.0 (± 6.4) at 60°, 72.9 (± 5.2) at 75°, and 79.1 (± 5.6) at 90° (Figure 4). Anterior cruciate ligament reconstructions in both groups showed significantly lower in situ forces at 6 and 12 weeks after reconstructions ($P < .05$). At 6 weeks and 12 weeks after reconstruction, the in situ forces of the ACL reconstructions in group 1 showed no statistically significant difference compared with group 2 (6 weeks: 30°, $P = .314$; 60°, $P = .525$; 75°, $P = .698$; 90°, $P = .367$; 12 weeks: 30°, $P = .514$; 60°, $P = .781$; 75°, $P = .391$; 90°, $P = .639$).

Structural Properties

Tensile testing of the intact ACL showed a mean stiffness of 48.3 (± 11.0) N/mm and ultimate force 462.3 (± 19.9) N (Figure 5). Ultimate stress resulted in 15.2 (± 2.2) N/mm². Stiffness, ultimate force, and ultimate stress were significantly larger compared with the ACL-reconstructed specimens after 6 and 12 weeks in both groups ($P < .05$). After 6 and 12 weeks, stiffness of the ACL reconstructions with 15 mm in the femoral tunnel (group 1) was not statistically significantly different compared with the reconstructions with 25 mm in the femoral tunnel (30°, $P = .610$; 60°, $P = .973$; 75°, $P = .685$; and 90°, $P = .928$) (Figure 5). The ultimate failure load 6 weeks after reconstruction of specimens in group 1 (15 mm) was 180.7 (± 89.9) N, whereas in group 2 (25 mm), the ultimate failure load was 165.8 (± 74.2) N. After 6 and 12 weeks, the ultimate failure load showed no statistically significant differences between group 1 and group 2 (30°, $P = .720$; 60°, $P = .633$; 75°, $P = .603$; and 90°, $P = .803$) (Figure 5). Ultimate stress 6 and 12 weeks after ACL reconstructions revealed no statistically significant differences in group 1 and group 2 (30°, $P = .197$; 60°, $P = .797$; 75°, $P = .874$; and 90°, $P = .421$) (Figure 5).

In both groups (15 mm and 25 mm), the predominant failure mode was a pull-out of the graft from the tunnels, a

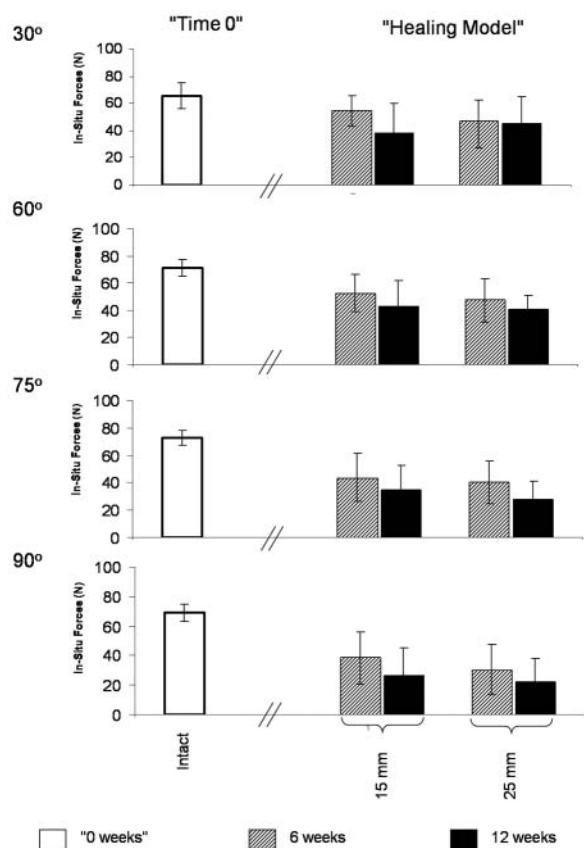


Figure 4. In situ force in newtons of the anterior cruciate ligament (ACL) and the ACL replacement graft for the control group ($n = 5$, intact) and the 2 healing groups with 15 mm and 25 mm in the femoral tunnel after 6 and 12 weeks (15 mm 6 weeks [$n = 7$], 15 mm 12 weeks [$n = 8$], 25 mm 6 weeks [$n = 9$], and 25 mm 12 weeks [$n = 8$]).

so-called “degloving” mechanism. After 12 weeks, the predominant failure mode was still a pull-out from the tunnels. There seemed to be a shift toward midsubstance failure; however, the difference was not statistically significant compared with the 6-week specimens.

Histologic Sections

After 6 weeks, in both groups, a granulation tissue filling the gap between the Achilles tendon split graft and the tunnel wall was found. The tendon-to-bone interzone was filled with Sharpey fibers, anchoring the tendon graft perpendicular to the tunnel wall. In the 15-mm and 25-mm groups, the interval between the Sharpey fibers was smaller close to the tunnel entrance than at the tunnel end (Figure 6).

After 12 weeks of healing, the same result was found. The Sharpey fibers were more dense at the articular tunnel entrance in the 15-mm and 25-mm groups when compared to the tunnel end.

DISCUSSION

The aim of the current study was to evaluate the effect of soft tissue graft length within the femoral tunnel in ACL reconstruction. We hypothesized that after 6 and 12 weeks, the structural properties and the knee kinematics after soft tissue ACL reconstructions with 15 mm within the femoral tunnel will be significantly inferior compared with the properties of ACL reconstruction with 25 mm in the tunnel. The results of the current study do not support our initial hypothesis. In fact, at 6 weeks postoperatively, the ATT under 67-N anterior tibial load was significantly lower for the reconstructions with 15 mm in the femoral tunnel compared with the ATT of reconstructions with 25 mm in the femoral tunnel at 60°, 75°, and 90° of knee flexion ($P < .05$). The in situ forces in the replacement grafts showed no statistically significant differences after 6 and 12 weeks between the 2 groups. The structural properties with regard to stiffness, ultimate failure load, and ultimate stress of reconstructions with 15 and 25 mm in the femoral tunnel also revealed no statistically significant differences at both time points.

Currently, there is a lack of scientific background for a recommendation of graft length in the femoral tunnel in ACL reconstruction using a soft tissue graft. In cruciate ligament reconstruction, there is the possibility of looping a hamstring graft to increase the diameter of the graft and thereby shorten it. By getting a sufficient graft diameter through this procedure, the surgeon may be able to avoid harvesting the gracilis tendon. In a recent prospective randomized clinical study, Gobbi et al⁶ reported significantly reduced internal rotation torque after ACL reconstruction using semitendinosus and gracilis tendon when compared with patients who underwent ACL reconstruction with only the semitendinosus tendon. This finding may also be important for the graft choice in multiligament reconstruction cases. A second clinical motivation for this basic research study was the reawakening of anatomic ACL reconstructions restoring the anteromedial and posterolateral bundle separately. Here, the femoral posterolateral bundle tunnel is usually short and results in a graft length of approximately 15 mm in the femoral tunnel.^{3,16,17,22,30,34,35}

To test our hypothesis, ACL reconstruction with 15 mm and 25 mm in the femoral tunnel were performed in an intra-articular goat model. The current study used a bilateral approach based on the findings of Cummings et al.⁴ These authors reported a large variation between subjects despite similar treatments and only little difference between ipsilateral and contralateral limbs despite different surgical treatments. They concluded that a subject-related factor may be responsible for regulating the quantity and quality of healing tissue. To account for this, a bilateral approach was used in the current study; however, to analyze the results accounting for this study design, a mixed-model analysis would have to have been performed. Upon expert statistical review, it was concluded that the more advanced statistical analysis would add confusion to the results and not add to the clinical significance of this study. After the first surgery, weightbearing of the animals improved constantly; before surgery of the

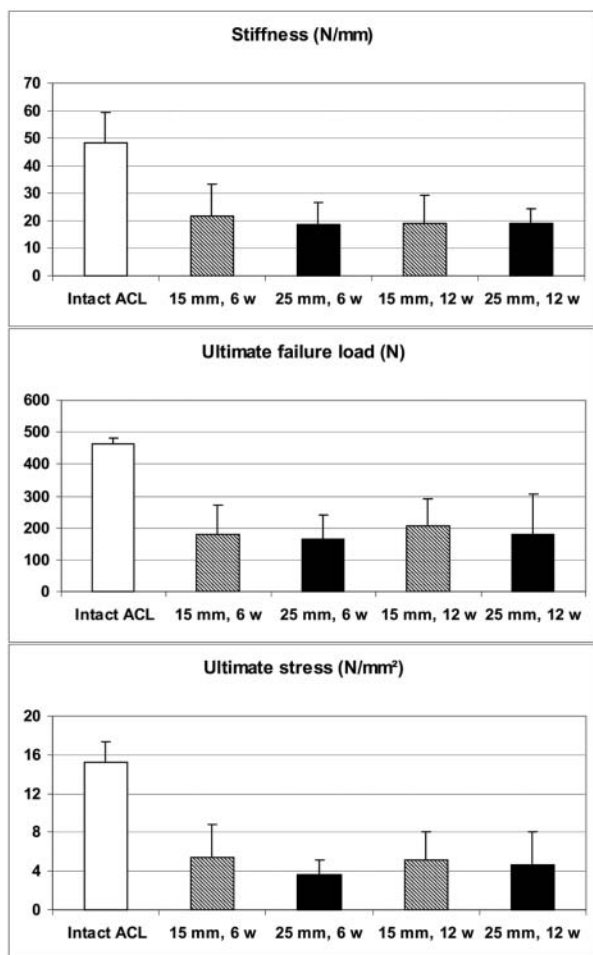


Figure 5. Structural properties of the intact goat anterior cruciate ligament (ACL) (control, n = 5) and the ACL reconstructions after 6 and 12 weeks (15 mm 6 weeks [n = 7], 15 mm 12 weeks [n = 8], 25 mm 6 weeks [n = 9], and 25 mm 12 weeks [n = 8]). Note that in the healing specimens, all fixation devices have been reharvested to evaluate the tendon-to-bone healing. With regard to stiffness, ultimate failure load, and ultimate stress, there was a statistically significant difference of the ACL-reconstructed specimens with 15 mm and 25 mm after 6 and 12 weeks, when compared with the intact knees. However, no statistically significant difference was found between the 2 tested groups. w, weeks.

contralateral side, all animals showed a normal gait pattern. The recovery to normal weightbearing after the second surgery was slower than for the first surgery; however, within the grading system, there was no statistically significant difference. Before sacrifice, all animals showed normal gait patterns. The results of this study support the hypothesis of Cummings et al that a bilateral reconstruction model with an interval of 6 weeks provides an opportunity to directly compare the subject and surgical treatment variances.⁴

To determine the ATT using a robotic/UFS testing system, an anterior tibial load of 67 N was used because these forces were reported to occur during trotting of adult

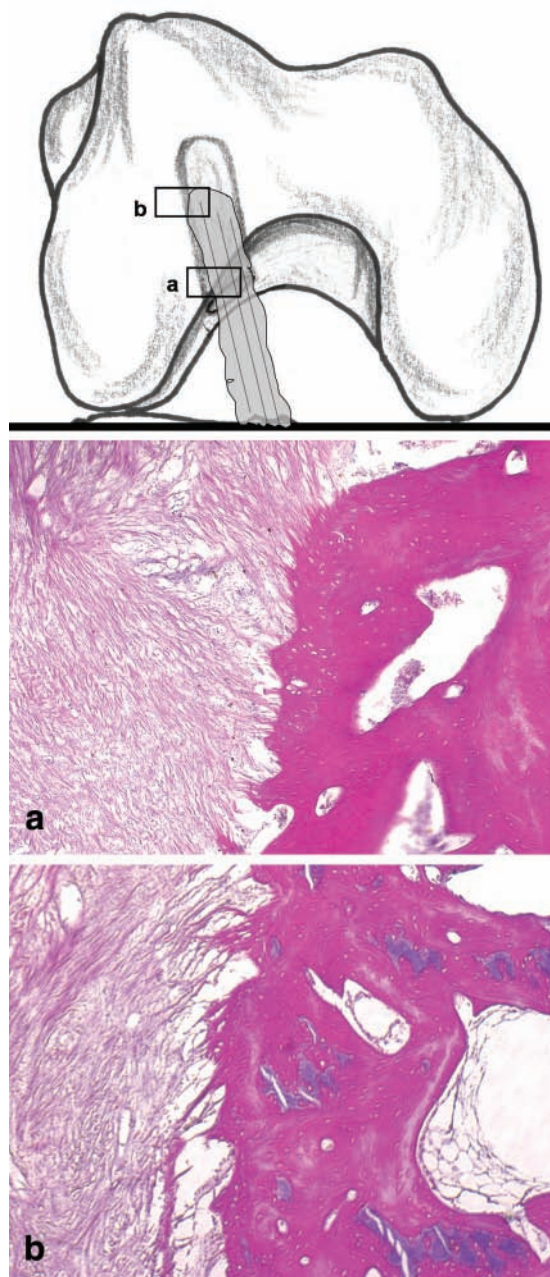


Figure 6. Typical example of tendon-to-bone healing of a specimen with 15 mm in the femoral tunnel after 6 weeks. Close to the intra-articular tunnel entrance (a), the Sharpey fibers appeared to be more dense than at the area 15 mm inside the tunnel (b).

goats.⁸ In contrast to our initial hypothesis, the ATT of reconstructions with 15 mm in the femoral tunnel was significantly lower than for reconstructions with 25 mm in the femoral tunnel after 6 weeks of healing; however, at 12 weeks of healing, this difference was no longer observed. The lack of significant difference observed in the 12-week group was more consistent with the results for the other

outcome measures (stiffness, ultimate load, ultimate stress) in which no significant differences between the 15-mm and 25-mm groups were recorded. Numerous speculations could be made regarding the significantly lower ATT with the 15-mm reconstruction at 6 weeks; however, without further investigation, it is thought that these speculations are not merited nor are they clinically significant, in light of the other findings of this study. After 6 and 12 weeks of healing in both groups, the ATT was significantly higher compared with the intact control group ($P < .05$). This may be due to the fact that in a goat model, the postoperative protocol is nearly impossible to standardize. After 4 weeks, the specimens showed a mean grade of weightbearing of 3.2 (± 1.0) in the 15-mm group and 3.4 (± 1.4) in the 25-mm group. At this time, the goats are able to bend their knees without any deficits in range of motion. Theoretically, a knee flexion of more than 120° may tension the ACL graft extensively and thereby may be responsible for the increased ATT in the reconstructed knees. The increased ATT after reconstruction is in accordance to the results of Weiler et al.²⁴ These authors reported a significantly increased anterior tibial displacement after ACL reconstruction in a sheep model up to a follow-up of 52 weeks compared with the intact knee.²⁴ For the clinical setting, there is a striking difference from the setting in an animal model. In most of the patients using a hamstring graft ACL reconstruction, a brace would be applied to protect the graft from extensive tensions.⁵

The structural properties of ACL reconstructions are important factors to determine the integrity of the reconstruction. In the current study, there was a statistically significant difference between the structural properties of the intact goat ACL and the ACL reconstructions. However, it has to be pointed out that those ultimate failure tests of the reconstructed knees included removal of the femoral graft fixation. The reason for this was that we tried to investigate the tendon-bone interface and the healed graft to the tunnel. After 6 weeks, there was no statistically significant difference between ACL reconstruction with 15 mm and 25 mm in the femoral tunnel. Twelve weeks after ACL reconstruction, there was likewise no statistically significant difference between the groups. Interestingly, there was no statistically significant difference in stiffness between the groups after 6 and 12 weeks.

The fact that nonintuitive/nonhypothesized results were observed for the ATT at 6 weeks and that otherwise no significant differences were observed between the 15-mm and 25-mm groups may suggest that tendon-to-bone healing occurs at the aperture site and that the length of the graft in the femoral tunnel is not significantly influencing the resulting biomechanical properties after ACL reconstruction. These findings were supported by the histologic analyses in which the anchoring of the graft to the tunnel wall using Sharpey fibers was more prominent at the tunnel entrance than at the tunnel end.

Similar findings were reported by Yamazaki et al,²⁹ when they studied the effect of graft length in the tibial tunnel following an ACL reconstruction in a canine model. Although Yamazaki et al were looking at tibial tunnel length rather than femoral tunnel and differences between

the surgical techniques and models existed, their ultimate conclusions were complementary to our findings. Similar to our findings, they observed no significant differences between their groups (5 mm and 15 mm) in the structural properties at 6 weeks of healing and their histologic results also pointed to healing initiating at the intra-articular space.

Berg et al² suggested that intra-articular tunnels heal from outside in and showed that the bone healing was slower and incomplete in the articular segment of the tunnel, close to the joint surface. In the study by Berg et al, the authors drilled a 3.2-mm intra-articular tunnel into the femoral condyles and analyzed the tunnel after 6 weeks using histomorphometric methods; however, no graft was pulled into the tunnel. With a graft in the tunnel, as was the case in this study, very different loading patterns would occur throughout the length of the tunnel and these differences could explain these contrary findings.

A few limitations also apply to the current study. First, we used an intra-articular model in goats. The range of motion of the goat knee shows an extension deficit of 30° and therefore the results of knee positions in extension cannot be reported. In this model, the hamstring tendons are too thin and short to provide a suitable graft. Therefore, we used an Achilles tendon split graft, which may not reflect the clinical setting in humans. Next, a standardized rehabilitation in this goat model could not be performed. However, ACL reconstructions in a goat model have been deemed successful out to 3 years with very low rates of graft failures or severe cartilage damage.^{10,13,18,21} Second, the current study reports data at 6 and 12 weeks. A longer follow-up would possibly result in higher structural properties; however, the follow-up was chosen according to the clinical relevance. Finally, we tested the femoral tendon-bone interface without any femoral fixation devices *in vivo*. Clinically, a graft failure may also occur at the tibial tunnel. However, when determining the graft length in the femoral tunnel, because a constant graft length was used in both groups, the tibial graft length and the length of the tibial linkage material is automatically affected. We therefore decided to evaluate the femoral graft length. Additionally, the results showed the ATT of reconstructions with 15 mm with no significant differences compared with 25 mm in the femoral tunnel after 12 weeks; however, the P value was .066. Theoretically, a higher number of specimens might show a higher statistical power and result in slight changes of the P value. However, power analysis revealed that to detect 30% difference with a power of 50%, 7 animals used for biomechanical analysis would be required.

This basic science study addressed the clinically relevant question: Is the fixation strength of a soft tissue autograft at 6 and 12 weeks after implantation adversely affected by reducing the length of tendon within a bone tunnel from 25 to 15 mm? Various clinical scenarios exist (some of which were previously discussed), in which the length of available graft that could be pulled into the tunnel (femoral or tibial) could be in question and it is currently unknown how this might affect outcomes. This study did not show any negative correlation with regard to knee kinematics and structural properties of a 15-mm length compared with a 25-mm length

in the femoral tunnel in a goat model. These findings in a goat model cannot be directly translated to the human setting; however, they do suggest that excessive graft length within the tunnel may be unnecessary (potentially removing the need to harvest the gracilis tendon) as there seems to be no kinematic or biomechanic benefit of inserting an extensively long graft into the femoral tunnel.

ACKNOWLEDGMENT

Thore Zantop, MD, was Research Fellow of the German Speaking Association of Arthroscopy (AGA) at the Department of Orthopaedic Surgery, University of Pittsburgh.

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