Can Markers Injected Into a Single-Loop Anterior Cruciate Ligament Graft Define the Axes of the Tibial and Femoral Tunnels? A Cadaveric Study Using Roentgen Stereophotogrammetric Analysis

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Lengthening of a soft-tissue anterior cruciate ligament (ACL) graft construct over time, which leads to an increase in anterior laxity following ACL reconstruction, can result from relative motions between the graft and fixation devices and between the fixation devices and bone. To determine these relative motions using Roentgen stereophotogrammetry (RSA), it is first necessary to identify the axes of the tibial and femoral tunnels. The purpose of this in vitro study was to determine the error in using markers injected into the portions of a soft-tissue tendon graft enclosed within the tibial and femoral tunnels to define the axes of these tunnels. Markers were injected into the tibia, femur, and graft in six cadaveric legs the knees of which were reconstructed with single-loop tibialis grafts. The axes of the tunnels were defined by marker pairs that were injected into the bones on lines parallel to the walls of the tibial and femoral tunnels (i.e., standard). By using marker pairs injected into the portions of the graft enclosed within the tibial and femoral tunnels and the marker pairs aligned with the tunnel axes, the directions of vectors were determined by using RSA, while a 150 N anterior force was transmitted at the knee. The average and standard deviations of the angle between the two vectors were 5.5 ± 3.3 deg. This angle translates into an average error and standard deviation of the error in lengthening quantities (i.e., relative motions along the tunnel axes) at the sites of fixation of (0.6 ± 0.8)%.

Introduction

Following anterior cruciate ligament (ACL) reconstruction, anterior laxity as measured by an anterior displacement of the tibia when subjected to an anterior force often increases over time and clinically important increases greater than 3 mm occur in 10–20% of cases [1–5]. Increased anterior laxity over time can be traced to lengthening of the graft construct (i.e., graft-fixation-bone complex) over time. Lengthening over time can result from the relative motion between the graft and the fixation devices, relative motion between the fixation devices and the bone, and/or lengthening of the graft substance between the fixations (e.g., due to remodeling). For any one of these three sources, lengthening over time can result either from plastic deformations, which increase the unloaded or slack length of the graft construct, and/or decreases in elastic stiffness, which lead to an additional increase in the length of the graft construct when loaded. One method that has been shown to quantify the relative motions is Roentgen stereophotogrammetric analysis (RSA) [6–9].

To quantify the relative motions between the graft and fixation devices and between the fixation devices and bone by using RSA, a vector that is parallel to the tunnel axes in each of the tibia and femur must be identified [6,9]. Each of the relative motions between the graft and fixation devices and between the fixation devices and bone will have two components, one parallel and one perpendicular to the tunnel axis. Because the fixations are either inside the tunnels or outside on the cortical bone and because the graft snugly fits in the tunnels, angulation of the graft in the tunnels is minimal at the sites of fixation, so that only the parallel component of any relative motion will cause an increase in the length of the graft construct. Knowledge of the vectors parallel to the tunnel axes is essential in determining the parallel component of each of the relative motions.

One proven method in defining vectors parallel to the tunnel axes is to inject two markers (i.e., marker pairs) into each bone by using a specially designed and constructed tool, so that the marker pairs lie on lines parallel to the axes of the tunnels [9]. An unproven method is to inject marker pairs into the portions of a single strand of a graft enclosed within the tibial and femoral tunnels and use the lines connecting these marker pairs to define the tunnel axes. The unproven method would eliminate the need to use the custom tool, hence simplifying the implantation process and would save surgical time in any future in vivo study. Therefore, the objective of this study was to determine the error in the direction of vectors determined from marker pairs injected into the portions of the graft enclosed within the tibial and femoral tunnels.

Materials and Methods

Experiments. Six cadaveric legs (average of 69 yr, range of 53–87 yr) were obtained and stored at −20°C. Radiographs and inspection of the knees at the time of ACL reconstruction did not reveal either moderate or severe degenerative arthritis, chondrocalcinosis, or torn menisci. The intact leg was thawed overnight before use.

The tibial metaphysis was reinforced with polyurethane foam to provide fixation structural properties in elderly cadaveric tibia similar to those in young human tibia [10]. The ACL was excised and tibial and femoral drill holes were made using a previously

Contributed by the Bioengineering Division of ASME for publication in the JOURNAL OF BIOMECHANICAL ENGINEERING. Manuscript received February 7, 2007; final manuscript received September 19, 2007; published online May 23, 2008. Review conducted by Jennifer S. Wayne.
respectively, while \( P^v \) vectors aligned with the axes of the femoral and tibial tunnels, \( P^G \) are markers attached to the tibial fixation, \( P^E \) is a marker attached to the femoral fixation, \( WL_1 \) and \( WL_2 \) drilled to the same diameter as the graft by using a tibial drill bit from the medial joint line of the tibia in the coronal plane, and \( H \) is perpendicular to the tibia in the sagittal plane.

Fig. 1 A-P view of the knee showing marker placement. \( T_1 – T_6 \) are markers implanted in the tibia, \( F_1 – F_6 \) are markers implanted in the femur, \( G_1 – G_8 \) are markers implanted in the graft, \( E \) is a marker attached to the femoral fixation, \( W_1 \) and \( W_2 \) are markers attached to the tibial fixation, \( H_1 \) and \( H_2 \) are vectors aligned with the axes of the femoral and tibial tunnels, respectively, while \( P^G_2 / G^2 \) is a vector computed using a pair of graft markers.

described transtubial technique that positions the graft without roof impingement, without PCL impingement, and with a tension pattern during passive flexion-extension [11] that is similar to that of the intact ACL [12]. A tibial guide (Howell 65 deg tibial guide, Arthrotek Inc., Ontario, CA) was used to place a tibial tunnel, so that the following criteria were met: (1) the tunnel axis was centered between the tibial spines and formed an angle of 65 ± 3 deg from the medial joint line of the tibia in the coronal plane, and (2) the tunnel was 5–6 mm posterior and parallel to the intercondylar roof at full extension in the sagittal plane. The tunnel was drilled to the same diameter as the graft by using a tibial drill bit (fully fluted cannulated tibial drills, Arthrotek, Inc.). A roof and wallplasty were performed to prevent impingement of the graft against the roof and wall of the intercondylar notch by inserting a metal rod, which has the same diameter as the tibial tunnel, so that it freely passed into the notch through the tibial tunnel with the knee in maximum hyperextension.

The femoral tunnel was positioned by inserting an over-the-top femoral aimer through the tibial tunnel (size-specific femoral aimer, Arthrotek, Inc.). An open-end femoral tunnel was drilled to 16 mm in diameter and a low-friction femoral bushing with an outer diameter of 16 mm and an inner diameter of 9 mm at its distal end was inserted into the femoral tunnel until flush with the intercondylar roof and fixed with bone cement [13]. This technique for endoscopically placing the tibial and femoral tunnels has good reproducibility between surgeons [14] and the tension pattern of the graft during passive flexion-extension [11] is similar to that of the intact ACL [12].

To establish a vector that is parallel to the tunnel axes in each of the tibia and femur (i.e., standard), two markers were placed in each of the tibia and femur. By using a tunnel vector tool [15], two 0.8 mm diameter tantalum markers (Tilly Medical Products AB, Lund, Sweden) spaced 2 cm apart were inserted into the posterior wall of the tibial tunnel (T1 and T2, Fig. 1). The tunnel vector tool ensured that the markers were inserted on a line that was parallel to the axis of the tunnel. Two more markers were placed in the low-friction femoral bushing to establish a line parallel to the axis of the femoral tunnel (F1 and F2, Fig. 1). Four more markers were implanted into both the femur and tibia. The use of six markers in each bone created an overdetermined system and reduced the error in determining the position of the markers [16].

To preserve the weight of the shank and water content in the specimens, one of the tibials was harvested from the previous specimen tested to serve as the graft in the subsequent specimen to be tested. The midportion of the tendon was exposed and the skin and surrounding tendon sheath were cut, exposing both the muscle and the distal insertion of the tendon. The distal insertion of the tendon was detached and the muscle was removed until the length of the tendon was a minimum of 27 cm, at which point the tendon was excised. The diameter of the tendon was trimmed until it slid without resistance through a 9 mm sizing sleeve when looped (Arthrotek, Inc.). Four cm from the end of each strand were whip stitched with a number-1 braided, absorbable suture (Polysorb; United States Surgical/ Syneture, Norwalk, CT) [10]. The tendon was then wrapped in a saline-soaked paper towel and stored at −20°C.

To determine the vectors defined by pairs of radio-opaque tantalum markers implanted into the graft, markers were injected into the portions of the tendon enclosed within the tibial and femoral tunnels by using the following techniques. The tendon was looped through the adjustable femoral fixation device with the thicker strand medially oriented until the strands were equal in length. The lengths of both tunnels and the intrarticular space were measured using a femoral tunnel depth gauge (Arthrotek, Inc.). The locations of the femoral and tibial tunnels were marked onto the graft. On the tibial end of the graft, two 0.8 mm diameter markers (G8 and G7) were implanted into the thinner strand and two 1.0 mm diameter markers (G1 and G2) were implanted into the thicker strand 5 mm proximal to the distal tibial tunnel mark and 5 mm distal to the proximal tibial tunnel mark (Fig. 1). Similarly, on the femoral end of the graft, two 0.8 mm markers (G5 and G6) were implanted into the thinner strand and two 1.0 mm markers (G4 and G3) were implanted into the thicker strand 5 mm distal to the marker on the femoral fixation (EL) and 5 mm proximal to the distal femoral tunnel mark (Fig. 1). Markers were implanted by injecting them \( \frac{\pi}{4} \) of the depth of the diameter of the graft with the injecting device (Tilly Medical Products AB, Lund, Sweden) perpendicular to the longitudinal axis of the tendon [17]. The use of markers with different diameters allowed the identification of each strand in the biplanar radiographs.

The graft was fixed in the femur and tibia by using the adjustable femoral fixation and WasherLoc fixation devices. The distal ends of the graft were passed from proximal to distal through the low-friction femoral bushing, intrarticular space, and tibial tunnel. Tension was distally applied to the graft and the knee was flexed and extended for 15 cycles. With the knee in maximum extension, a 110 N load measured using a load cell (Model L1650, Futek Inc., Irvine, CA) was applied to the distal end of the graft. The graft was fixed on the tibial side with an 18 mm extended-spiked WasherLoc (Arthrotek, Warsaw, IN).

A custom loading apparatus applied posterior and anterior forces to the proximal tibia of the leg specimens (Fig. 2). Forces perpendicular to the tibia at 12.5 cm distal to the joint line were applied by a pneumatic actuator, controlled to within ±1 N and held within this range for 10–20 s until simultaneous radiographs (described below) were taken. Three additional markers were fixed in a line along the pneumatic actuator to enable the identification of the loading axis from the radiographs.

To standardize the load transmitted at the line of the knee joint and perpendicular to the tibia in the sagittal plane (hereafter termed the A-P shear force transmitted at the knee), two load cells positioned in line with the attachment at the tibia and the attachment at the ankle were used (Fig. 2). The weight of the shank foot...
of each specimen was estimated using regression equations [18]. The sum of the two load cells was equal to the sum of the A-P shear force transmitted at the knee and the component of the weight perpendicular to the shank. A 150 N A-P shear force transmitted at the knee was standardized. This standardization ensured that the A-P shear force transmitted at the knee was the same for each specimen independent of the weight of the specimen.

The RSA system included several components. The calibration cage (Tilly Medical Products AB, Lund, Sweden) contained embedded markers at known positions. The calibration cage was modified to hold two X-ray cassettes and two scatter grids (Medical X-Ray Enterprises, Inc., Culver, CA) placed at right angles to each other (A-P and lateral views). The scatter grids were used to minimize exposure of the radiographic film from scattered X-rays and thus improve the image quality. The calibration cage surrounded the tibio-femoral joint such that all markers could be seen from each view. The portable X-ray machines (Model HF80H+, MinXray Inc., Northbrook, IL) were positioned perpendicular to, and at a distance between 80 cm and 100 cm from, each X-ray cassette. The parameters of the X-ray machines were initially set to 1.5 mA s and 80 kVp, and the parameters were adjusted until well-exposed radiographs were obtained.

Each specimen was inserted into the loading apparatus, the knee was centered in the calibration cage, and the knee was flexed to 25 deg, as measured with a goniometer. This flexion angle was selected because it is in the range where anterior knee laxity is the greatest [12]. Additionally, the tibia was placed in a position of neutral rotation because this is the position of greatest laxity [12]. The 150 N anterior shear force transmitted at the knee was applied and simultaneous radiographs were taken.

Data Analysis. Analysis of the radiographs was performed using a customized RSA data analysis system previously described [15]. Briefly, a digital image was obtained for each radiograph by using a backlit scanner (Epson 1600, Epson America Inc., Long Beach, CA). The two-dimensional centroid coordinates of the markers were measured from the digital image using a software program (SCION IMAGE 1.0, Scion Corporation, Frederick, MD). A customized computer program written in MATLAB (Version 6.0, The Mathworks Inc., Natick, MA) was used to determine the two vectors defined by the two pairs of markers injected into the tunnel walls and the four vectors defined by the four pairs of markers injected into the graft. This program initially computed the transformation of image coordinates to the calibration cage, the positions of the Roentgen foci, and the 3D position coordinates of all the markers in a laboratory coordinate system defined by the calibration cage [19].

To determine the error in the direction of vectors determined from marker pairs injected into the tendon, the tendon vectors (e.g., \( \mathbf{P}_{G2/G1} \), defined as the vector to marker G2 from G1) were computed. The tendon vectors for both pairs of markers in a particular tunnel (i.e., tibial or femoral) were then averaged by computing the averages of the corresponding coordinates (e.g., \( \left( \mathbf{P}_{G2/G1} + \mathbf{P}_{G7/G8} \right)/2 \)). To serve as the standard, the vectors defined by the marker pairs injected using the tunnel vector tool also were computed (e.g., \( \mathbf{P}_{T2/T1} \), defined as the vector to marker T2 from T1). The angle \( \theta \) between the average tendon vector and the standard vector was computed as

\[
\theta = \cos^{-1} \left( \frac{\left( \mathbf{P}_{G2/G1} + \mathbf{P}_{G7/G8} \right)/2 \cdot \mathbf{P}_{T2/T1}}{\left| \left( \mathbf{P}_{G2/G1} + \mathbf{P}_{G7/G8} \right)/2 \right| \cdot \left| \mathbf{P}_{T2/T1} \right|} \right)
\]

Because only the component of any relative motion between the fixation devices and graft and/or between the fixation devices and bone along the tunnel axis contributes to the lengthening of the graft construct [6], the error was computed from

\[
\text{error}(\%) = (1 - \cos \theta) \times 100
\]

Similar equations were written for both strands of the graft in the femoral tunnel. To quantify the error related to a change in length of the graft construct, the average error and the standard deviation of the error were computed. All statistics were computed using the data from both tunnels in all specimens.

Results

The average and standard deviations of the angle between the vector computed using the marker pairs injected into the wall of the tunnels and the average vector computed using the marker pairs injected into the tendon were 5.5 \( \pm \) 3.3 deg. This angle translates into an average error and a standard deviation of the error in lengthening quantities (i.e., relative motions along the tunnel axes) at the sites of fixation of (0.6 \( \pm \) 0.8\%)%.

Discussion

An increase in the unloaded and/or loaded length of an ACL graft construct over time causes a corresponding increase in anterior laxity following an ACL reconstruction [20] and a vector that is parallel to the tunnel axes must be identified to quantify the relative motions between the graft and fixation devices and between the fixation devices and bone by using RSA. Therefore, the objective of this study was to determine the error in using marker pairs injected into the tendon to determine the axes of the tibial and femoral tunnels. Our most important finding is that the vectors determined from marker pairs injected into the tendon result in small errors and therefore can be used to determine the axes of the tunnels.

The implication of this finding is that it will reduce surgical time in an in vivo RSA study. In an in vivo RSA study, the surgical time for the ACL reconstruction will be increased due to the injection of markers into the bone and into the tendon. Therefore, it is necessary to inject the markers as efficiently as possible. One way to minimize surgical time is to eliminate any unnecessary steps or procedures. Because markers will be injected into the graft for the RSA study, the added time to inject an additional four markers required to create marker pairs will be inconsequential. However, the time required to use the tunnel vector tool to place markers that define the axes of the tibial and femoral tunnels is substantial, requiring an extra 10–15 min. With an operating room cost estimated at $50/min, the extra time translates into a substantial extra cost.

There are two potential sources of error associated with our RSA analysis, the inherent error associated with the RSA system and the migration of markers in the tendon graft under cyclic load. The inherent error associated with the RSA system is 0.05 mm [15], while the migration of markers injected into single-looped
grafts is 0.10 mm [17]. Because both of these sources of error are minimal, high confidence can be placed in the determination of the vectors parallel with the axes of the tunnels.

The error in determining the axes of the tibial and femoral tunnels by using marker pairs injected into the tendon would not be expected to change during cyclic loading. This error could change if the markers moved or migrated within the tendon substance. However, migration of markers in single-loop tendon grafts subjected to 225,000 load cycles was minimal, as noted above [17].

While this study focused on single-loop tendon grafts, it is of interest to consider whether direct injection of marker pairs could also be used for double-loop grafts. As discussed previously [17], markers of the size used in our study cannot be reliably injected perpendicular to the long axis of the tendons used for double-loop grafts because of their smaller cross-sectional area. Hence, for either in vitro or in vivo study of double-looped grafts, the tunnel axes must be identified by marker pairs injected into the walls of the tunnels on lines parallel to the tunnel axes.

In conclusion, by using the methods described in this study, where marker pairs were injected perpendicular to the long axis of a single-loop tendon graft, the marker pairs can be used to determine vectors parallel with the tunnel axes with an angle difference of 5.5° on average. This angle difference translates into an average error of 0.6% when the vectors based on marker pairs injected into the graft are projected onto the axes of the tunnels. Hence, this angle difference will introduce a minimal error when computing the various causes of lengthening of a graft construct over time associated with the sites of fixation.

Acknowledgment

The authors thank the Musculoskeletal Transplant Foundation for financial support of this research project.

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