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# Biomechanical Testing of Three Alternative Quadrupled Tendon Graft Constructs With Adjustable Loop Suspensory Fixation for Anterior Cruciate Ligament Reconstruction Compared With Four-Strand Grafts Fixed With Screws and Femoral Fixed Loop Devices

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*Investigation performed at the Department of Research, Arthrex GmbH, Munich, Germany*

Please delete "different"; revised text should read "To biomechanically analyze 3 alternative tendon constructs fixed with adjustable suspensory fixation devices [...] and femoral fixed loop device."

**Background:** Quadrupled semitendinosus (ST) grafts for anterior cruciate ligament (ACL) reconstruction have advantages of greater graft diameter and gracilis (G) preservation compared with doubled ST-G grafts. However, a paucity of biomechanical data are available regarding different preparation techniques for these constructs.

**Purpose:** To biomechanically analyze 3 different alternative tendon constructs fixed with adjustable suspensory fixation devices on the femur and tibia compared with a matched 4-strand construct fixed with a tibial screw and femoral fixed loop device.

**Study Design:** Controlled laboratory study.

**Methods:** Three alternative quadrupled tendon preparation techniques with suspensory fixation (grafts constructs A, B, and C) were compared with a 4-strand screw-fixed loop device construct (graft construct D) in matched diameter bovine tendon graft and porcine tibia models. Graft constructs were tested with a 3-stage cyclic loading protocol (1000 cycles in position control and 1000 cycles each from 10 to 250 N and from 10 to 400 N), followed by a pull to failure. In graft construct A, the graft ends were whipstitched and tied over the tibial button; in graft construct B, the graft ends functioned as pulleys; and in graft construct C, a continuous loop was created. Initial, dynamic, and total elongation, stiffness, and ultimate failure load were recorded.

**Results:** Graft construct D had the highest initial ( $0.51 \pm 0.29$  mm) and total ( $3.53 \pm 0.98$  mm) elongation compared with the 3 quadrupled constructs ( $P < .001$  each). Graft construct B had lower total elongation ( $2.13 \pm 0.31$  mm) compared with graft construct A ( $2.40 \pm 0.30$  mm) ( $P = .004$ ) and graft construct C ( $2.53 \pm 0.21$  mm) ( $P = .007$ ). Graft construct C had a higher ultimate failure load ( $1097 \pm 79$  N) compared with graft construct A ( $988 \pm 112$  N) ( $P = .001$ ), graft construct B ( $973 \pm 137$  N) ( $P = .022$ ), and graft construct D, which had the lowest failure load ( $767 \pm 182$  N) ( $P < .001$ ).

**Conclusion:** The 3 quadrupled tendon suspensory fixation constructs exhibited small yet statistically significant biomechanical differences among each other. Constructs that used tibial screw fixation had lower ultimate failure load and higher total elongation compared with the quadrupled tendon constructs.

**Clinical Relevance:** Total elongation for the screw fixation group was higher than the threshold of clinical failure, which may allow for graft construct elongation during the postoperative rehabilitation phase. Biomechanical properties of the 3 quadrupled tendon suspensory graft constructs may be clinically comparable, albeit statistically different.

**Keywords:** ACL reconstruction; quadrupled graft; suspensory fixation; biomechanical testing

diameter was associated with decreased graft rupture after hamstring ACL reconstruction.<sup>18,19</sup> Increased graft diameter in hamstring grafts can be achieved via 5- or 6-strand semitendinosus (ST) and gracilis (G) grafts, or by quadrupling the ST and preserving the G tendon. Although the benefits of G preservation have been questioned,<sup>27</sup> harvest of both the G and ST has been linked to knee flexor weakness<sup>31,37</sup> and low rates of tendon regeneration,<sup>15</sup> suggesting that avoidance of unnecessary tendon harvesting warrants further examination.

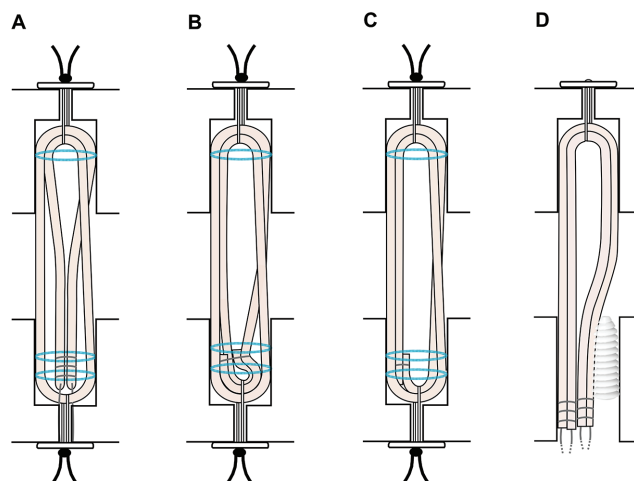
Although quadrupling of the ST increases the graft diameter, it also results in shorter grafts, producing unique fixation issues compared with traditional, longer, doubled ST-G grafts. Novel fixation techniques have been developed, such as adjustable suspensory fixation, that allow shorter grafts to be used, all-inside or outside-in, while obtaining fixation strength similar to that of traditional devices.<sup>8,12,20,30</sup> Although biomechanical data regarding these newer adjustable loop devices (ALDs) have been reported, a paucity of comparative data exist on the biomechanics of the optimum method of tendon quadrupling.

**[AQ-1]** The purpose of this biomechanical study was to examine 3 alternative quadrupled tendon constructs fixed with adjustable suspensory fixation compared with a benchmark, matched diameter tendon, 4-strand construct fixed with a tibial interference screw and a femoral fixed loop device.<sup>4,30</sup> Our hypothesis was that the 3 quadrupled tendon constructs would have similar biomechanical properties in regard to initial, dynamic, and total elongation, stiffness, and ultimate failure load compared with the 4-strand control group.

## METHODS

A partial construct testing method that entailed bovine tendons and porcine tibias was used. Porcine bone was chosen because similarities to the human knee of young adults have been reported.<sup>1,21</sup> The femoral side consisted of a custom-made acrylic block. ST and G grafts, which are commonly used for ACL reconstructions, have been described as being comparable with the bovine extensor digitorum tendons in regard to their mechanical properties.<sup>11</sup>

Three different preparation techniques to create quadrupled tendon constructs were biomechanically tested. Graft construct A, with graft ends whipstitched and the free sutures additionally tied over the



**Figure 1.** Quadrupled and 4-strand constructs tested. (A) Graft construct with whipstitched ends and additional knots of the whipstitching sutures tied over the tibial button. (B) Each graft end was sutured as a pulley over the tibial suture loop and each other. (C) Graft ends were stitched together to create a continuous loop. (D) Four-strand graft fixed to the tibia with an interference screw and to the femur with a fixed loop device. Graft ends were whipstitched together and tied for fixation to allow equal tensioning during screw insertion.

button; graft construct B, where the stitched graft ends function as pulleys; and graft construct C, a continuous loop technique. Graft construct D, a 4-strand control construct, was created to match the diameter of the quadrupled tendon constructs and to allow tensioning of all 4 strands during tibial screw fixation (Figure 1).

## Specimen Preparation

After porcine tibias were acquired from a local abattoir, attached soft tissue was removed and the bones were embedded in a 2-component fast cast resin system consisting of polyurethane (Huntsman Advanced Materials). The tibial plateaus were sectioned to create a constant total tunnel length of 40 mm. For biomechanical testing of the quadrupled tendon constructs (graft constructs A, B, and C), tibias were prepared by drilling a tunnel through the tibia, beginning at the medial plateau and ending at the footprint of the ACL tibial plateau. Then, a 9-mm drill was used

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to retro-drill a socket measuring 30 mm in depth. The socket distance was measured off the drill guide, and in this way, a consistent bone bridge of 10 mm was retained to mimic an all-inside technique. For the control, graft construct D, the 4-strand constructs were fixed with a screw on the tibial side, and a full tibial tunnel with a diameter of 9 mm was drilled.

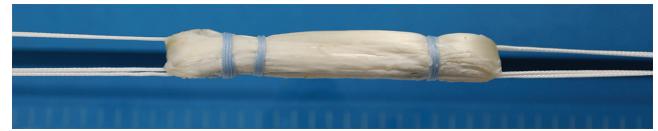
For bovine graft preparation, limbs from fresh 2-year-old cattle were obtained from an abattoir, and the lateral tendons of the common digital extensor muscle were harvested. The tendons were prepared to obtain tendon segments of 9 mm in diameter when quadrupled or in 4 strands. The final quadrupled tendon constructs (graft constructs A, B, and C) and 4-strand construct (graft construct D) had lengths of 65 mm and 80 mm, respectively. The 4-strand constructs were longer to obtain interference fixation along the entire screw length in the tibial tunnel.

## Graft Preparation

Eleven grafts were tested in each group. The grafts were prepared on a graft preparation system (Arthrex), and the quadrupled constructs were created with suspensory fixation using a no-button TightRope TN (Arthrex) on the tibial side and a TightRope RT (Arthrex) on the femoral side. In contrast, the control group used a  $9 \times 28$ -mm bio-composite interference screw (Arthrex) for tibial fixation and the RetroButton (Arthrex) as a femoral fixed loop device with 20-mm loop length.

**Graft Construct A.** This graft construct was prepared in accordance with a technique previously described by Smith and DeBerardino.<sup>30</sup> Both ALDs were secured on the graft preparation station. The graft was sectioned at 270 mm. Then, both ends were placed through the femoral ALD by the graft ends were stitched (Arthrex) in a Speedy suture. The tensioning of these sutures was done into the inside of the tunnel. A tension of 20 N was applied before the graft was placed on the tibial side. For this purpose, the needle was passed through 2 graft strands beginning on the inside of the construct; then it was stitched back through all 4 strands and wrapped around the whole construct to “link” them together. [AQ:2] The needle was then passed through the other 2 strands ending on the inside again, and a knot was tied and then pulled into the inside of the graft. After this was completed, 80 N of tension was placed on the construct and 1 cerclage stitch was placed on the femoral side and 1 more stitch on the tibial side. The sutures used for whipstitching the tendon ends were not cut but rather were preserved for later use to be tied as backup fixation on the tibial side.

**Graft Construct B.** Bovine grafts were sectioned to a length of 300 mm, and the tibial and femoral ALDs were positioned on the graft preparation station. One end of the graft was folded for 5 mm over the tibial ALD to create a pulley, sutured back onto the graft with a No.



**Figure 2.** Completed graft construct B with 2 linked stitches on the tibial side and 1 linked stitch on the femoral side.

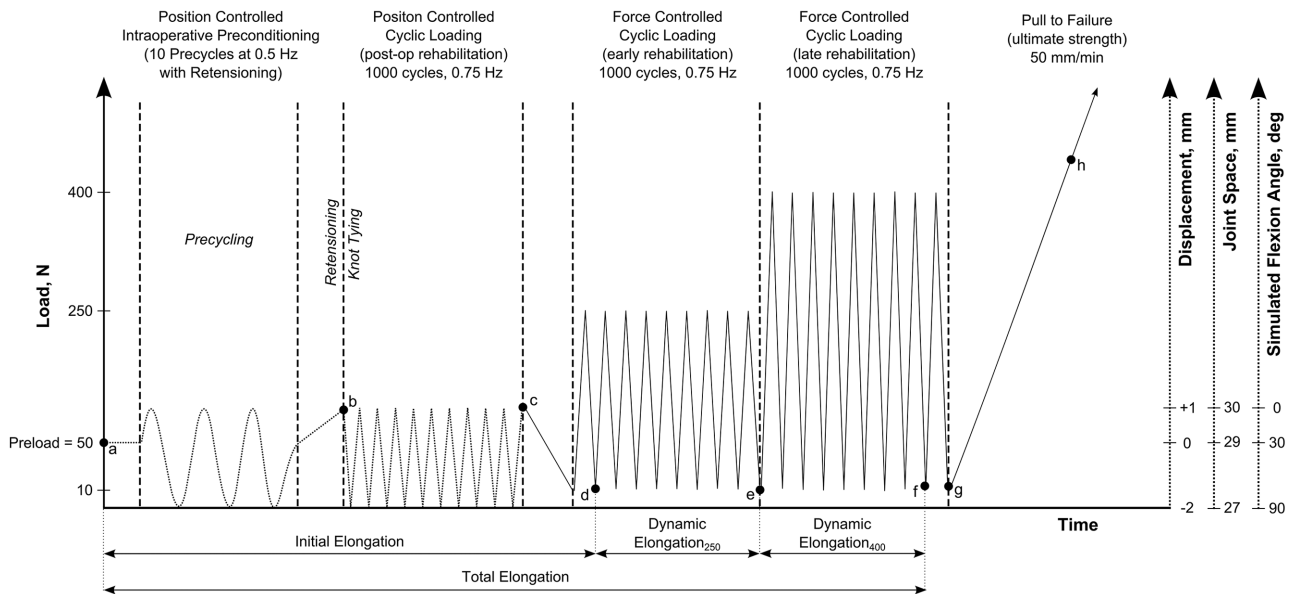
0 FiberWire (Arthrex), and tied with a surgeon’s knot. The free graft end was then passed through the loop of the femoral ALD, wrapped back toward the tibial ALD and looped through, and finally passed for the second time through the femoral ALD. Next, the free graft end was wrapped back toward the tibial ALD, resulting in a quadrupled construct. The graft end was brought inside the construct and the tip folded over the tibial ALD. Last, the graft was sutured for fixation back onto the first pulley as done for the other end. Under a tension of 20 N, the first cerclage stitch was made on the tibial side, and to complete the preparation the 2 remaining cerclage stitches were secured at a tension of 80 N (Figure 2).

**Graft Construct C.** The bovine graft was sectioned at a length of 280 mm, and the tibial and femoral ALDs were placed within the graft preparation station. The graft was passed through the loops of both ALDs, leaving 1 short end and 1 longer end. The longer end was then pulled through both loops again in the opposite direction until the graft ends met.<sup>4</sup> They were stitched together with a No. 0 FiberWire by overlapping the ends for a distance of 1 cm. After the knots were tied and the sutures cut, the sutured portion of the graft was pulled on the inside of the graft and rotated so that it was located within the loop of the tibial ALD.<sup>4</sup> The construct was placed under a tension of 20 N for the first cerclage stitch on the tibial side. Upon completion, a tension of 80 N was applied and the remaining 2 cerclage stitches were added.

**Graft Construct D.** The bovine tendons were sectioned at a length of 325 mm and then cut in half. The grafts were doubled and both ends were whipstitched together for the last 20 mm with a No. 2 FiberLoop. Then, the doubled graft was hung into the suture loop of the fixed loop device. The 2 looped whipstitched sutures were knotted to create 1 looped suture. Equal tensioning of the 4-strand construct during screw insertion was ensured by pulling on this created suture loop.

## Biomechanical Testing

Testing was performed by use of a materials testing machine (ElectroPuls E10000; Instron) with a 2-kN load cell installed within the crosshead. All constructs were tested with a cyclic loading protocol before a pull to failure. The native ACL is both force and motion controlled and shows a slack behavior at midflexion angles during passive or weightbearing knee flexion, which indicates an unloaded state of the ligament.<sup>3,17,26,35</sup> Therefore, the test protocol includes a position control block, allowing an unloaded graft situation, and a load control block



**Figure 3.** Test protocol for biomechanical testing including points of interest for data evaluation: initial elongation ( $\Delta ad$ ); force maintenance during position controlled loading ( $\Delta bc$ ); dynamic elongation<sub>250</sub> ( $\Delta de$ ) and dynamic elongation<sub>400</sub> ( $\Delta df$ ); total elongation ( $\Delta af$ ); and ultimate failure load and stiffness during pull to failure ( $\Delta gh$ ). Dotted lines indicate position controlled cycling.

(Figure 3). The position control block occurs at the beginning of testing and represents the postoperative phase of early range of motion exercise. Li et al<sup>17</sup> demonstrated that the ACL elongates with increasing knee extension, its maximum length of 30 mm occurs at full knee extension, and it decreases by approximately  $-1$  mm and  $-3$  mm at  $30^\circ$  and  $90^\circ$  of knee flexion, respectively. Each construct was initially fixed at 29 mm of joint space corresponding to a displacement of 0 mm and a  $30^\circ$  flexion angle, which is a commonly used fixation angle in ACL reconstruction surgery.<sup>10</sup> Then, the constructs were cycled in position control between  $+1$  mm and  $-2$  mm of displacement simulating range of motion up to  $90^\circ$  of knee flexion. The load control blocks with both a lower and a higher load level represent the early and late rehabilitation phases, respectively.<sup>14,23,25,28,32,33</sup> Hence, the evolved test protocol replicates in vivo loads acting on the ligament after an ACL reconstruction.

Before biomechanical testing, each prepared graft was preloaded with 80 N for 5 minutes. Subsequently, quadrupled tendon constructs (graft constructs A, B, and C) were inserted in the acrylic block representing the femur by pulling the femoral ALD through the socket until the button flipped. Then, the sutures of the tibial ALD were pulled through the porcine tibia, and a concave button was attached. The tibial tunnel was in line with the load axis to create a worst-case testing scenario. A preload of around 50 N was achieved by tensioning the shortening sutures on both the femoral and tibial sides. Then, the shortening strands on the tibial side were knotted over the button with a surgeon's knot secured by 3 alternate half hitch knots. Screw fixation constructs were threaded through the tibia and subsequently through the acrylic block. A

load of 50 N was applied on the graft while the interference screw was inserted until it was flush with the tibial plateau.

The test began with a preload of exactly 50 N, and this position was then used as a new baseline by adjusting the elongation to zero. For all-inside graft constructs, 10 pre-cycles at 0.5 Hz in position control mode were performed. Subsequently, the femoral ALD was retensioned to approximately 200 N and also knotted like the tibial side. For grafts prepared according to technique A, the whipstitching sutures were also tied over the button. Graft constructs D were not precycled as it is technically not possible to retension a fixed loop device after primary fixation. The final test setup can be seen in Figure 4.

Cyclic loading at 0.75 Hz began in a position control mode with movements between  $+1$  and  $-2$  mm. This was followed by 2 force control blocks, one proceeding between 10 and 250 N and the other between 10 and 400 N. Each block consisted of 1000 cycles, and testing was completed with a pull to failure at 50 mm/min. Data were recorded at a sampling rate of 500 Hz. The mode of failure was noted as the load and displacement curves were recorded. Based on these data, elongation behavior, force level in position control mode, stiffness during pull to failure, and ultimate load were determined.

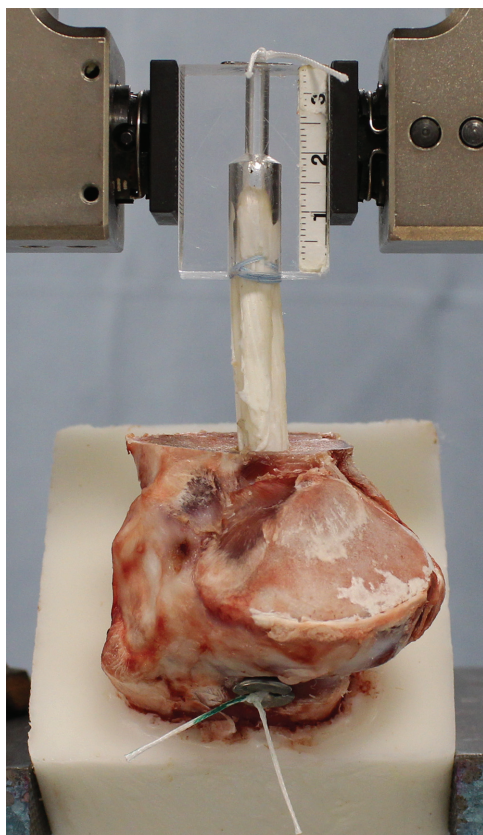
By definition, the initial elongation was the elongation occurring between the preload of 50 N and the first completed cycle of the first load control block ( $\Delta ad$ ) (Figure 3). The dynamic elongation<sub>250</sub> ( $\Delta de$ ) and dynamic elongation<sub>400</sub> ( $\Delta df$ ) were defined as the elongation occurring between the first and the last cycles of the 250-N load block and the 400-N load block, respectively. The sum of initial and dynamic elongation<sub>400</sub> was the total elongation ( $\Delta af$ ).



TABLE 1  
Results for Each Graft Construct<sup>a</sup>

	Graft Construct A	Graft Construct B	Graft Construct C	Graft Construct D
Initial elongation, mm	$-0.51 \pm 0.08$	$-0.46 \pm 0.22$	$-0.29 \pm 0.08$	$0.51 \pm 0.29$
Dynamic elongation <sub>250</sub> , mm	$1.24 \pm 0.15$	$1.07 \pm 0.12$	$1.22 \pm 0.12$	$1.46 \pm 0.23$
Dynamic elongation <sub>400</sub> , mm	$2.91 \pm 0.33$	$2.64 \pm 0.27$	$2.82 \pm 0.25$	$3.00 \pm 0.80$
Total elongation, mm	$2.40 \pm 0.30$	$2.13 \pm 0.31$	$2.53 \pm 0.21$	$3.53 \pm 0.98$
Initial force, N	$204.8 \pm 11.3$	$208.4 \pm 17.3$	$195.7 \pm 10.8$	$127.3 \pm 27.9$
Force maintenance, %	80.0	85.1	81.1	78.9
Stiffness, N/mm	$202.1 \pm 12.9$	$214.8 \pm 11.9$	$215.7 \pm 10.6$	$236.0 \pm 17.6$
Ultimate failure load, N	$988 \pm 112$	$973 \pm 137$	$1097 \pm 79$	$767 \pm 182$
Method of failure	Femoral suture rupture (100%)	Femoral suture rupture (82%), tibial suture rupture (9%), combination (9%)	Femoral suture rupture (91%), combination of femoral and tibial suture rupture (9%)	Graft slippage (100%)

<sup>a</sup>Values are expressed as mean  $\pm$  SD.



**Figure 4.** Construct testing setup. A custom-made acrylic block serves as a femur, whereas a tibia of porcine origin was used. The tibia was cut at 40 mm to ensure a constant tunnel length, and the space between acrylic block and bone was initially set to 29 mm representing a knee flexion angle of 30°.

Moreover, force maintenance ( $\Delta bc$ ) was specified as the proportion of residual force after the position controlled cyclic loading was completed. Stiffness was measured between 250 and 400 N during pull to failure ( $\Delta gh$ ).

## Statistical Analysis

Statistical analysis using SigmaPlot 13.0 (Systat Software) was performed to compare elongation, stiffness, and ultimate failure load between the 3 quadrupled tendon constructs and the 4-strand control group. The evaluation was completed by use of a 1-way analysis of variance before a pairwise Student-Newman-Keuls post hoc test. The level of significance was set at  $P < .05$ , and post hoc power analysis revealed a mean power of 0.9988, which is greater than the desired power of 0.8, leading us to conclude that our sample size was sufficient.

To determine that each data set followed normal distribution and equal variances, a Shapiro-Wilk test and a Brown-Forsythe test were performed, respectively. For data sets that failed either of these tests, the Kruskal-Wallis test, a nonparametric test, was used followed by a Student-Newman-Keuls post hoc test for pairwise analysis.

## RESULTS

Results (mean  $\pm$  SD) for elongation, stiffness, and ultimate failure load are presented in Table 1.  $P$  values for all tests are reported in Table 2.

## Cyclic Loading

**[AQ:3]** After the position control block was completed, it was observed that graft construct D showed the highest initial elongation ( $0.51 \pm 0.29$  mm) and the lowest initial force level ( $127.3 \pm 27.9$  N), which was a statistically significant difference compared with all quadrupled tendon graft constructs (graft constructs A, B, and C) ( $P < .001$  each). Force maintenance at the end of the position control block was within the range of 78.9% and 85.1% for all constructs tested.

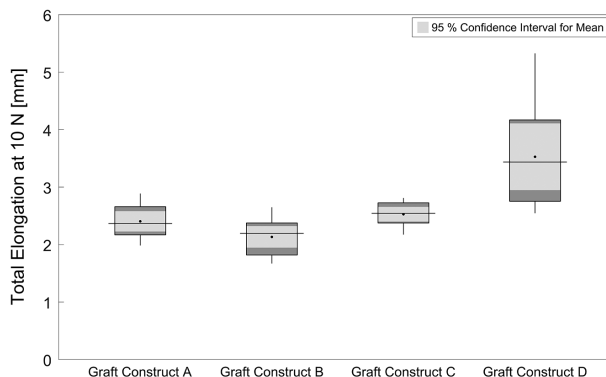
For dynamic elongation during the first load control block (250 N), a significant difference was found between

TABLE 2  
P Values for Student-Newman-Keuls Post Hoc Analysis<sup>a</sup>

	Graft Construct A	Graft Construct B	Graft Construct C	Graft Construct D
Initial elongation, mm				
Graft construct A	—	.536 (.071 <sup>b</sup> )	.025 (.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct B	.536 (.071 <sup>b</sup> )	—	.041 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct C	.025 (.001 <sup>b</sup> )	.041 (<.001 <sup>b</sup> )	—	<.001 (<.001 <sup>b</sup> )
Graft construct D	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	—
Dynamic elongation <sub>250</sub> , mm				
Graft construct A	—	.033	.672	.003
Graft construct B	.033	—	.035	<.001
Graft construct C	.672	.035	—	.002
Graft construct D	.003	<.001	.002	—
Dynamic elongation <sub>400</sub> , mm				
Graft construct A	—	.358 (.253 <sup>b</sup> )	.623 (.834 <sup>b</sup> )	.667 (.412 <sup>b</sup> )
Graft construct B	.358 (.253 <sup>b</sup> )	—	.379 (.055 <sup>b</sup> )	.280 (.140 <sup>b</sup> )
Graft construct C	.623 (.834 <sup>b</sup> )	.379 (.055 <sup>b</sup> )	—	.626 (.974 <sup>b</sup> )
Graft construct D	.667 (.412 <sup>b</sup> )	.280 (.140 <sup>b</sup> )	.626 (.974 <sup>b</sup> )	—
Total elongation, mm				
Graft construct A	—	.248 (.004 <sup>b</sup> )	.598 (.111 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct B	.248 (.004 <sup>b</sup> )	—	.216 (.007 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct C	.598 (.111 <sup>b</sup> )	.216 (.007 <sup>b</sup> )	—	<.001 (<.001 <sup>b</sup> )
Graft construct D	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	—
Initial force, N				
Graft construct A	—	.642 (.393 <sup>b</sup> )	.244 (.054 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct B	.642 (.393 <sup>b</sup> )	—	.237 (.067 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct C	.244 (.054 <sup>b</sup> )	.237 (.067 <sup>b</sup> )	—	<.001 (<.001 <sup>b</sup> )
Graft construct D	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	—
Stiffness, N/mm				
Graft construct A	—	.034	.059	<.001
Graft construct B	.034	—	.877	.002
Graft construct C	.059	.877	—	.001
Graft construct D	<.001	.002	.001	—
Ultimate failure load, N				
Graft construct A	—	.795 (.470 <sup>b</sup> )	.061 (.001 <sup>b</sup> )	.001 (.002 <sup>b</sup> )
Graft construct B	.795 (.470 <sup>b</sup> )	—	.085 (.022 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )
Graft construct C	.061 (.001 <sup>b</sup> )	.085 (.022 <sup>b</sup> )	—	<.001 (<.001 <sup>b</sup> )
Graft construct D	.001 (.002 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	<.001 (<.001 <sup>b</sup> )	—

<sup>a</sup>P values less than .05 were considered statistically significant.

<sup>b</sup>P values correspond to post hoc analysis after Kruskal-Wallis test.



**Figure 5.** Box and whisker plots including 95% CIs for the mean depicting total elongation values for tested graft constructs.

graft constructs A and B ( $P = .033$ ) and between graft constructs C and B ( $P = .035$ ). The dynamic elongation of graft construct D was greater than that of graft constructs A, B, and C ( $P = .003$ ,  $P < .001$ , and  $P = .002$ , respectively). No statistically significant difference was found between any of the groups during the second load control block (400 N).

Regarding the total elongation, graft construct D exceeded the threshold of 3 mm for clinical failure<sup>9</sup> and showed a total elongation of  $3.53 \pm 0.98$  mm. This was significantly higher than the total elongation of graft constructs A, B, and C ( $P < .001$  each). None of the quadrupled graft constructs tested exceeded a total elongation value of 3 mm within the applied loading cycles. Box and whisker plots depicting the total elongation results are illustrated in Figure 5.

## Pull to Failure

During pull to failure, the highest stiffness was found for graft construct D ( $236.0 \pm 17.6$  N/mm) and this was significantly stiffer than graft constructs A, B, and C ( $P < .001$ ,  $P = .002$ , and  $P = .001$ , respectively). Graft construct A showed the least stiffness ( $202.1 \pm 12.9$  N/mm). The highest ultimate failure load was  $1097 \pm 79$  N for graft construct C, and the lowest was  $82$  N for graft construct D. This was significantly less than failure load for constructs A, B, and C ( $P = .002$ ,  $P < .001$ , and  $P = .001$ , respectively). Moreover, a difference in failure load between constructs B and C ( $P = .022$ ) and between constructs A and C ( $P = .001$ ).

Change "TR" to "TightRope"; revised text should read "The methods of failure were similar for all 3 all-inside constructs, all specimens failed due to suture rupture of the TightRope, on either the femoral or tibial side, or due to a combination."

## Mechanism of Failure

The methods of failure were similar for all 3 all-inside constructs; all specimens failed due to suture rupture of the TR, on either the femoral or tibial side, or due to a combination. [AQ:5] Graft construct D exhibited graft slippage as the failure mode for all constructs tested.

Change the comma to "and"; revised text should read "The aim of an ACL reconstruction is to restore normal knee stability without altering knee kinematics and articular loading, or overconstraining the joint."

## DISCUSSION

The aim of an ACL reconstruction is to restore normal knee stability without altering knee kinematics, articular loading, or overconstraining the joint. [AQ:6] Avoidance of graft elongation plays an essential role in achieving knee stability, because changes in graft stiffness and temporal postimplantation changes in graft length affect knee laxity.<sup>6</sup> [AQ:7] In this analysis, 3 alternative quadrupled suspensory graft constructs were biomechanically tested in relation to a benchmark 4-strand construct fixed with tibial screw and femoral fixed loop device in order to compare elongation, stiffness, and load-to-failure properties. Compared with the benchmark construct, all quadrupled graft constructs exhibited less total elongation and higher ultimate failure load; among each other, the quadrupled graft constructs displayed small but statistically significant differences.

Graft construct B showed significantly less total elongation after completing the second force control block compared with graft constructs A and C. Whether these differences are clinically meaningful remains uncertain, particularly given that later graft-bone union will alter elongation.<sup>29</sup> However, graft construct D, with a tibial screw and femoral fixed loop device, exhibited a total elongation above the threshold of clinical failure of 3 mm.<sup>9</sup> Because no significant difference was found regarding dynamic elongation after completion of the second force block, the reason for a higher total elongation is a higher initial elongation, as it is technically not possible to retension fixed loop devices or screw fixation after primary fixation. Therefore, constructs using these types of fixation have a lower initial and final force level when undergoing position controlled cyclic loading, given that force

maintenance levels were comparable among all groups tested. Consequently, more elongation is needed within the first load cycle to reach the peak load of 250 N. In contrast, for ACL reconstructions fixed with ALDs, intraoperative preconditioning such as retensioning can eliminate initial elongation due to settling effects. This conclusion is supported by a study by Noonan et al,<sup>22</sup> who emphasized the importance of preconditioning by showing that both retensioning and knotting of the ALD could decrease the final cyclic elongation by around 50% compared with an ALD that underwent neither knotting nor retensioning.

Graft construct stiffness is also an important consideration in ACL reconstruction, as knee laxity depends not only on graft lengthening but also on stiffness of both the graft and the fixation method.<sup>6</sup> In this analysis, graft construct D had a statistically significant, higher stiffness compared with the 3 quadrupled constructs. The tibial screw for graft construct D was inserted until it was flush with the tibial tunnel; therefore, the graft portion that was inserted was smaller and hence stiffer. However, the range of reported stiffness values for ACL grafts between 111 and 397 N/mm.<sup>34</sup> Moreover, the greater or lesser construct stiffness relative to native ACLs remain uncertain, as construct stiffness may actually be somewhat different due to normalization of articular cartilage

properties, particularly in regard to loading allowances during rehabilitation, is the ultimate failure load. Graft construct C had a failure load of  $1097 \pm 79$  N, which was significantly higher compared with failure loads for graft constructs A and B. Moreover, the lowest failure load was observed for graft construct D, which was statistically significantly different than the quadrupled constructs. This difference potentially can be explained by the fixation method, as tibial screw fixation is considered to be the weakest point of the whole construct since the screw is in line with the load axis.<sup>2,16</sup> Nonetheless, all ACL reconstruction constructs reportedly have ultimate failure loads greater than the peak forces acting on the graft construct during the postoperative rehabilitation phase.<sup>14,28,32</sup>

This study has some inherent limitations. Porcine bones and bovine grafts were used for partial construct biomechanical testing. The advantages of using animal material over cadaveric material include better comparability and reproducibility as the animals are of similar age and body conditions. Although animal tissue often serves as substitute material,<sup>11,16</sup> some authors discourage using animal material due to noteworthy differences.<sup>7,24</sup> Although our bovine-porcine model of the human graft-femur constitutes a limitation, we believe that this limitation is acceptable considering that it allowed consistent and comparable biomechanical testing. Moreover, we chose a test setup with the graft construct aligned with the load axis. This simulates a worst-case testing scenario for ACL reconstruction testing but does not replicate common in vivo loading situations. Furthermore, our testing protocol exposed the graft constructs to a total of 3000 loading cycles, which may not fully represent the number of cycles



a graft experiences during the postoperative rehabilitation phase until final incorporation. Finally, this biomechanical study does not account for in vivo factors such as biological healing, which may also affect device elongation.

## CONCLUSION

In this study, the 3 quadrupled suspensory graft constructs exhibited small yet significant biomechanical differences. Graft construct A was the least stiff, graft construct B exhibited the smallest total elongation, and graft construct C had the highest load to failure.

However, stiffness values and ultimate failure loads of all constructs can be considered comparable in regard to clinical relevance, given that all were within the range of the native ligament and higher than the forces acting on the ACL during rehabilitation, respectively. Graft construct D with a tibial screw and femoral fixed loop device also revealed a clinically comparable stiffness and sufficient ultimate failure load, despite a decrease of approximately 30% compared with graft construct C. However, the total elongation of graft construct D exceeded the 3-mm threshold of clinical failure, suggesting that intraoperative preconditioning including precycling and retensioning enabled by use of ALDs is beneficial in ACL reconstruction.

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