Original Article

Quadruple Hamstring Autograft Technique for Anterior Cruciate Ligament Reconstruction Reduces Allograft Augmentation

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Purpose: To assess the discrepancy in graft diameter between double- and quadruple-folded hamstring autografts and the need for allograft augmentation to obtain an adequate graft diameter during arthroscopic anterior cruciate ligament reconstruction. **Methods:** All patients undergoing anterior cruciate ligament reconstruction with hamstring autograft between 2017 and 2021 at a single institution by a single surgeon were identified. The surgeon changed from double-folded hamstring autograft to quadruple-folded hamstring autograft within the study period. **Results:** A total of 191 patients were identified, of whom 57 received double-folded autografts and 134 quadruple-folded autografts. Patient characteristics between cohorts were similar. Median double-folded graft size (7.5 mm; interquartile range, 7.0-8.0 mm) was significantly thinner than the quadruple-folded graft size (9 mm; interquartile range, 8.5-9.5 mm, $P = .001$). Quadruple-folded autograft was less likely to require an allograft augmentation than the double-folded autograft (0.7% vs 26.3%) (odds ratio 0.02; 95% confidence interval 0.00-0.16; $P < .001$). **Conclusions:** Quadruple-folded hamstring autograft provides a larger graft diameter and reduced need for allograft augmentation. **Level of Evidence:** Level III, retrospective comparative study.

The anterior cruciate ligament (ACL) is the most commonly injured ligament about the knee, with an increasing incidence of 22% between 2002 and 2014.[1] For anterior cruciate ligament reconstruction (ACLR), hamstring autograft has become increasingly popular as a graft option, with noninferior retear rates or outcomes reported compared with other graft options.[2,3] Hamstring diameters have been found to vary in mean folded graft diameter, with biomechanical studies showing that graft size affects ultimate failure load.[4,5]

Common techniques to increase graft diameter include allograft augmentation and quadruple-folding of the hamstring autograft, which form additional stranded grafts.[4] Studies assessing the use of allograft augmentation raise concerns for increased rates of graft rupture, particularly in adolescents; however, this is inconclusive in adults.[6-11] The purpose of this study was to assess the discrepancy in graft diameter between double- and quadruple-folded hamstring autografts and the need for allograft augmentation to obtain an adequate graft diameter during ACLR. It was hypothesized that quadruple-folded grafts would have a larger average diameter and reduced need for allograft augmentation.

Methods

Full approval was obtained from the institutional review board (The University of Tennessee College of Medicine Institutional Review Board #1782087). Data were collected retrospectively on patients undergoing ACLR between 2017 and 2021. Patient records were accessed by two fourth-year orthopaedic surgery residents (C.W.P. and C.D.N.) through an electronic medical record system for review of operative notes. Exclusion criteria included patients in whom hamstring autograft was not used or revision ACLR. Inclusion

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The authors report that they have no conflicts of interest in the authorship and publication of this article. Full ICMJE author disclosure forms are available for this article online, as supplementary material.

Received April 8, 2022; accepted September 13, 2022.

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2666-061X/22420

https://doi.org/10.1016/j.asmr.2022.09.006

criteria included all patients, adult and adolescent, undergoing ACLR with or without meniscal injury between 2017 and 2021 in whom hamstring autograft was used. All patients were assessed and surgeries performed by a single sports medicine fellowship-trained surgeon at the same hospital several years into practice. This surgeon underwent a change in technique in 2019, from previously using double-folded hamstring autograft to a quadruple-folded technique. The decision to augment with allograft was made when the harvested graft was less than 8.0 mm. The following variables were obtained for each cohort, double-folded or quadruple-folded: number of patients, patient characteristics including age, sex, smoking status as well as graft diameter, and potential allograft augmentation.

Surgical Technique

Once under anesthesia, the patient was positioned and draped in a standard fashion with an unsterile thigh tourniquet placed proximal to the isolation drapes. Once the leg is exsanguinated, an approximately 4.0-cm incision is placed over the pes insertion. After a transverse incision is made through the sartorial fascia, the semitendinosus and gracilis are harvested and transferred to a back table for graft preparation while the surgeon proceeds with ACL remnant debridement and drill tunnel preparation.

Suspensory, cortical fixation of the graft was used on both the femoral and tibial side. A femoral-side TightRope (Arthrex, Naples, FL) and tibial-side TightRope (Arthrex) were placed on posts on opposite sides of the

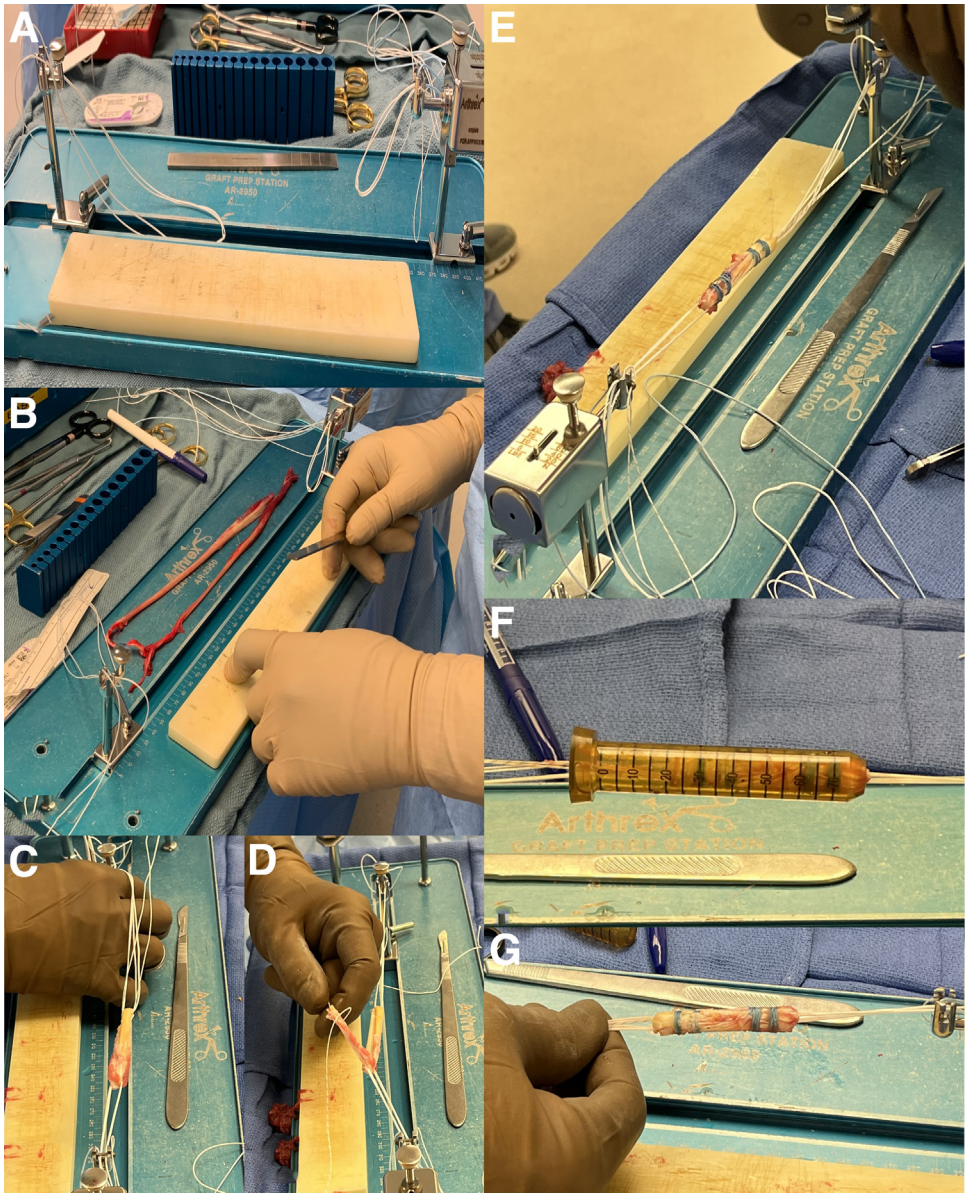

Fig 1. Technique for quadruple-folded hamstring autograft. (A) A femoral-side TightRope (Arthrex) and tibial-side TightRope (Arthrex) are placed on posts on opposite sides of the prep station. (B) The remaining muscular tissue is removed from the gracilis and semitendinosus, leaving only tendinous tissue. (C) The double-folded graft is passed through the loop formed by the tibial-side TightRope while the tail ends of the graft are passed through the loop formed by the femoral-side TightRope and folded down over the prior double-folded graft creating the quadruple-folded graft. (D) The tails are whip stitched together using 2-0 FiberWire, with these strands then being secured to the tibial post. (E) Circumferential stitches using 2-0 ETHIBOND are placed at 10 mm and 20 mm from each graft end. (F-G) The graft is then tubularized, finalizing graft preparation.

Table 1. Summary of Patient Characteristics and Preaugmentation Graft Diameters

	Double-Folded (n = 57)	Quadruple-Folded (n = 134)	*P* Value
Age, y	18 (17-31.5)	26 (18-38.25)	.022*
Sex			.811
Male	27 (47.4%)	66 (49.3%)	
Female	30 (52.6%)	68 (50.7%)	
Smoking	6 (10.5%)	14 (10.4%)	.987
Follow-up, mo	6 (3-7)	4 (2-6)	.008*
Graft size, mm	7.5 (7.0-8.0)	9.0 (8.5-9.5)	<.001*
6.0	3 (5.3%)	–	
6.5	7 (12.3%)	–	
7.0	11 (19.3%)	2 (1.5%)	
7.5	16 (28.1%)	5 (3.7%)	
8.0	12 (21.1%)	17 (12.7%)	
8.5	5 (8.8%)	20 (14.9%)	
9.0	1 (1.8%)	43 (32.1%)	
9.5	2 (3.5%)	19 (14.2%)	
10.0	–	22 (16.4%)	
10.5	–	3 (2.2%)	
11.0	–	2 (1.5%)	
11.5	–	1 (0.7%)	
Augmentation	15 (26.3%)	1 (0.7%)	<.001*

NOTE. Data are presented as number (%) and median (interquartile range).

*Statistically significant difference ($P < .05$).

prep station (Fig 1A).[12,13] The remaining muscular tissue was removed from the gracilis and semitendinosus leaving only tendinous tissue (Fig 1B). The semitendinosus graft was then folded over on itself, forming a double-folded graft with 2 free tail ends and 1 looped end. The looped end of the graft was then passed through the loop formed by the tibial-side suspensory cortical fixation whereas the tail ends of the graft were passed through the loop formed by the femoral-side suspensory cortical fixation. The looped and tail ends of the graft were then folded down over the previously double-folded graft, creating the quadruple-folded graft (Fig1C). The 2 tails were whip stitched together using 2-0 FiberWire, with these strands then being secured to the tibial post (Fig 1D).[14] The quadruple-folded 4-strand semitendinosus graft diameter and length were then assessed, ideally 9.0 mm in diameter and 65-70 mm in length. If the diameter was less than 8.0 mm, the gracilis previously harvested was incorporated into the graft alongside the semitendinosus graft in the manner described previously, forming an 8-strand graft.

Once adequately sized, the graft was then measured and marked at 10 and 20 mm from each end of the graft, providing 4 marks for circumferential stiches. Using 2-0 ETHIBOND, the needle was passed from the inner strands of the graft through the outer strands.[15] The suture was then wrapped around all 4 strands of the graft 2 to 3 times before passing back through the outside to inside graft strands. Once passed back through the graft, 3 half hitches were tied with the suture tails. The needleless tail of the knot was cut while the needled strand was passed back through the graft to bury the knot (Fig 1E). The graft was then tubularized, finalizing graft preparation (Fig 1 F and G).

Statistical Analysis

Statistical analyses were performed with IBM SPSS Statistics, version 27.0 (IBM Corp., Armonk, NY). Tests were conducted 2-tailed and an alpha level of 0.05 defined statistical significance. Power analysis confirmed the sample size was adequate based on a medium effect size, a power of 0.80, and an alpha level of 0.05. Patient characteristics are expressed as frequencies (percentage) for categorical variables and median (interquartile range) for continuous variables. Normality was assessed by the Shapiro–Wilk test ($P > .05$). A Mann–Whitney *U* test was conducted to determine significant differences in patient characteristics and graft sizes. The χ^2 test or Fisher exact test was conducted to determine significant differences in categorical patient characteristics and need for augmentation. To adjust for confounders, logistic regression was conducted to determine the association between the 2 procedures and the need for augmentation. Linearity was assessed by the Box–Tidwell procedure. Multicollinearity was absent.

Results

In total, 229 patients were identified to have undergone ACLR between 2017 and 2021. Of those, 38 patients were excluded due to use of allograft only (22), patellar tendon (2), and quadriceps tendon (13) grafts. Of the 191 patients included, 57 underwent the double-folded autograft technique whereas 134 received the quadruple-folded autograft technique (Table 1). Patient characteristics were similar in sex (male 49.3% vs 47.4%) and smoking status (10.4% vs 10.5%). The quadruple-folded cohort, however, was significantly older than the double-folded cohort (median 26 vs 18 years; $P = .022$). Median graft size of the double-folded technique (7.5 mm; interquartile range 7.0-8.0 mm) was significantly thinner than the quadruple-folded technique (9 mm; interquartile range 8.5-9.5 mm; $U = 6940.5$, $z = 9.031$, $P = .001$). Quadruple-folded autograft was significantly less likely to require an allograft augmentation than the double-folded autograft (odds ratio 0.02; 95% confidence interval 0.00-0.16; $P < .001$). When adjusted for age and sex, quadruple-folded autograft remained less likely to require an augmentation (odds ratio 0.02; 95% confidence interval 0.00-0.17; $P < .001$) (Table 2). The single patient who underwent allograft augmentation following quadruple-folded technique displayed an increase in graft diameter from 6.5 to 9.5 mm. The patient was 46 years old, with several years of knee pain and

Table 2. Unadjusted and Adjusted Associations Between Allograft Augmentation and Procedure Type

	Allograft Augmentation		
	OR (95% CI)	Adjusted OR (95% CI)	*P* Value
Procedure, quadruple-folded	0.02 (0.00-0.16)	0.02 (0.00-0.17)	<.001*
Age	—	0.98 (0.93-1.04)	.608
Sex, male	—	0.58 (0.18-1.85)	.361

CI, confidence interval; OR, odds ratio.
*Statistically significant ($P < .05$).

documented mucoid degeneration of the ACL on magnetic resonance imaging.

Discussion

Double-folded hamstring autografts, in comparison with quadruple-folded, are significantly smaller in diameter and require a greater frequency of augmentation, affirming the authors' hypotheses. The median graft diameters (quadruple-folded 9.0 mm vs double-folded 7.5 mm) fall on opposite sides of what has previously been shown to be the minimum graft diameter below which retear rates are significantly greater (8.0 mm).[16] Allograft augmentation is a technique used to increase graft diameter, as demonstrated in this study, with an increase in augmented graft diameter from a median 6.5 mm to 9.25 mm.

The effect of allograft augmentation on retear rates is currently debated. Perkins and colleagues[6] in 2019 performed a retrospective review on patients 19 years and younger. They found 2.6 times the odds of graft failure in allograft augmented grafts compared with comparably sized, isolated 4-strand hamstring autograft in this adolescent population. Pennock and colleagues[7] in 2017 also performed a retrospective review of allograft augmented ACLR on a pediatric population and found that allograft augmentation led to a statistically increased rate of graft failure. Burrus and colleagues[8] in 2015 came to a similar conclusion in an adult population with increased rates of failure in allograft augmented grafts with worse patient outcome scores.

In contrast, Jimenez and colleagues[9] in 2019 in a retrospective review showed no significant difference in failure rates between allograft augmented grafts and hamstring autograft in an adult population, although the failure rate in augmented group was 8.7% whereas the autograft alone failure rate was 4.3%. They did, however, find Knee Injury and Osteoarthritis Outcome Scores to be significantly greater in autograft alone than augmented grafts. Rao and colleagues[11] in 2020 in another retrospective review of adult and adolescents found augmenting a hamstring autograft that is 8 mm or less with allograft showed no difference in overall reoperation, revision of ACL failure, or patient reported outcomes. Lastly, Heyworth and colleagues[10] in 2021 showed no difference in retear rates and clinical outcomes with augmented grafts in an adolescent group in a retrospective review.

Because controversy within the literature exists about the impact of allograft augmentation, the study surgeon sought to eliminate any potential risk of worse retear rates or patient outcome scores. By augmenting the semitendinosus autograft with the gracilis autograft forming an 8-stranded graft, it was hypothesized and later supported in this study that an appropriate diameter could be achieved, thus eliminating the need for allograft augmentation. Suspensory fixation was required due to the length of the graft necessarily shortened due to quadruple folding. While this posed an additional change in the studied surgeon's technique, literature has come out in support of this suspensory type fixation.[17] A prospective randomized control trial comparing allograft augmented versus autograft alone during ACLR using suspensory fixation is needed.

Limitations

This study is not without limitations. The 2 cohorts are not entirely comparable. However, logistic regression was used to adjust odds ratios for confounders, particularly age, to determine the association between the 2 procedures and the need for augmentation. Additional limitation comes from the retrospective nature inherent in this form of study.

Conclusions

Quadruple-folded hamstring autograft provides a larger graft diameter and reduced need for allograft augmentation.

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