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Am J Sports Med published online July 29, 2016
DOI: 10.1177/0363546516656181

AJSM PreView, published on July 29, 2016 as doi:10.1177/0363546516656181

Incorporation of Hamstring Grafts Within the Tibial Tunnel After Anterior Cruciate Ligament Reconstruction

Magnetic Resonance Imaging of Suspensory Fixation Versus Interference Screws

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Background: The success of anterior cruciate ligament (ACL) reconstruction requires solid graft incorporation within the tunnels to enable graft remodeling. Resorbable interference screws (RIS) provide limited tendon-bone contact because much of the tunnel circumference is occupied by the screw itself, while adjustable suspensory fixation (ASF) systems provide larger contact zones, which favor ligamentization.

Purpose: To evaluate ligamentization of a 4-strand semitendinosus (4ST) graft fixed with ASF compared with RIS within the tibial bone tunnel at 6 months postoperatively using magnetic resonance imaging (MRI).

Study Design: Cohort study; Level of evidence, 2.

Methods: We prospectively enrolled 121 consecutive patients undergoing primary ACL reconstruction using a single-bundle 4ST graft. The femoral end of the graft was fixed using suspensory fixation in all knees. The tibial end of the graft was fixed using ASF in 67 knees and RIS in 54 knees. Six months postoperatively, knee laxity measurements were taken, and MRI was performed to assess graft incorporation within the tibial tunnel.

Results: At 6-month follow-up, MRI scans of 109 knees were available for analysis. The mean tibial tunnel enlargement in the ASF group was 2.3 ± 1.1 mm (range, 0.5-6.0 mm), while in the RIS group, it was 4.7 ± 2.8 mm (range, 0.5-19.0 mm) ($P < .001$). The Howell graft signal assessment findings were excellent in 97% of knees in the ASF group and in 25% of knees in the RIS group ($P < .001$). The mean signal-to-noise quotient (SNQ) was 0.078 ± 0.62 in the ASF group and 0.671 ± 0.83 in the RIS group ($P < .001$).

Conclusion: ASF provides more favorable conditions than RIS for the incorporation and ligamentization of 4ST grafts within the tibial tunnel. The ASF system used showed very little tunnel widening, which suggests that it grants stabilization. The SNQ was also considerably better in the ASF group.

Keywords: ACL repair; anterior cruciate ligament; interference screws; suspensory fixation

The advent of bioabsorbable interference screws in anterior cruciate ligament (ACL) reconstruction resolved several drawbacks of metal screws,[4] including graft damage, postoperative image distortion, and need for surgical removal.[14,42,51] Despite their widespread use, bioabsorbable screws remain associated with migration,[42] cyst formation,[55] and tunnel widening,[17,32,36,46] even with the incorporation of osteoconductive agents such as beta-tricalcium phosphate (β-TCP) or hydroxyapatite.[1-3,23,39]

To mitigate such adverse effects, suspensory graft fixation devices were introduced and are gradually replacing interference screws.[43] While the initial EndoButton continuous-loop design was associated with frequent tunnel widening,[7,9,17,27,36,46] likely caused by increased graft micromotion and thence a "bungee cord" effect or "windshield wiper"

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One or more of the authors has declared the following potential conflict of interest or source of funding: P.C. received royalties from SBM, which manufactures the 2 devices described in this article. N.G.: Amplitude, Sanofi-Aventis (other financial or material support, paid consultant), Arthrex Inc, DePuy (paid consultant, paid presenter or speaker), SBM (unpaid consultant).

The American Journal of Sports Medicine, Vol. XX, No. X
DOI: 10.1177/0363546516656181

TABLE 1
Patient Demographics (N = 109 Patients)[a]

	ASF Group (n = 60; 55%)	RIS Group (n = 49; 45%)	*P* Value
Age at surgery, y	28.9 ± 9.5 (14.8-55.3)	27.6 ± 6.8 (15.8-46.7)	NS
Follow-up, mo	6.6 ± 1.2 (5.1-10.7)	6.6 ± 1.3 (4.0-10.0)	NS
Male sex, n (%)	46 (77)	41 (84)	NS
Tunnel diameter, mm	8.8 ± 0.7 (8-11)	8.2 ± 0.7 (7-10)	<.01

[a]Data are reported as mean ± SD (range) unless otherwise indicated. ASF, adjustable suspensory fixation; NS, nonsignificant; RIS, resorbable interference screws.

phenomenon, more recent designs were enhanced with adjustable tension loops and render promising results.[36]

The success of ACL reconstruction requires solid incorporation of the tendon graft within the bone tunnels to enable its histological remodeling into a structure similar to the native ACL.[11,22,50] The performance of a graft fixation device therefore depends both on its mechanical strength and on its ability to provide favorable conditions for rapid graft healing and ligamentization.[54] The fixation of soft collagenic tissue from the graft tendon within the bone tunnel is initiated by Sharpey fibers[35,52] and depends on the tendon-bone area of contact.[35] Interference screws provide a limited tendon-bone contact area because much of the tunnel circumference is occupied by the screw itself, while suspensory fixation systems provide larger contact zones that favor ligamentization.[28]

Numerous authors have attempted to evaluate graft ligamentization at 6 months postoperatively[16,30,40] because this is a common time frame for athletes to be cleared to resume sports and it is sufficient time for the graft remodeling process to render detectable changes.[20,21,37,47,54,56] Moreover, Howell et al[20] demonstrated that grafts can attain sufficient stability as early as 3 months postoperatively. Healing of the graft at the bone-graft interface, bony reaction around the tunnel, and homogeneity and density of the graft can be objectively and precisely assessed using magnetic resonance imaging (MRI). While most studies focused on the intra-articular graft portion, the authors are not aware of any published studies that compared the properties of the graft itself within the bone tunnel.

The purpose of this study was to evaluate the ligamentization (remodeling and maturation) of 4-strand semitendinosus (4ST) grafts within the tibial bone tunnel, using MRI at 6 months postoperatively, fixed with adjustable suspensory fixation (ASF) systems compared with resorbable interference screws (RIS). The hypotheses were that (1) the ASF system provides improved conditions for bony graft integration compared with RIS fixation because of better contact between the graft and bone and (2) the ASF system does not lead to tunnel widening as found for the continuous-loop EndoButton.[19]

METHODS

Study Design

The authors prospectively enrolled 121 consecutive patients undergoing primary ACL reconstruction between February and September 2014. All procedures were performed by 2 surgeons at the same center using single-bundle 4ST grafts. The graft was fixed within the femoral tunnel using a suspensory system in all 121 knees but within the tibial tunnel using ASF in 67 knees and using RIS in 54 knees. The knees that underwent ASF in the tibia were all consecutive cases operated by one surgeon (P.C.), and likewise, the knees that received RIS in the tibia were also consecutive cases operated by another surgeon (S.J.). All patients provided written informed consent for their participation in the study, which was approved by the institutional review board in advance (No. 3320150901).

The inclusion criteria were patients aged 18 to 64 years undergoing arthroscopic primary ACL reconstruction surgery for isolated ACL tears confirmed both clinically (laxity, pain, and instability) and on MRI. The exclusion criteria were concomitant lesions of the collateral ligaments or posterior cruciate ligament (PCL), revision procedures, and procedures that required meniscal repair.

From the 121 patients enrolled, 12 declined to continue participation in the study, which left data from 109 patients for analysis. There were no intraoperative or postoperative complications or ruptures. The RIS group consisted of 41 men (83.7%) and 8 women with a mean age of 27.6 years (range, 15.8-46.7 years), and the ASF group consisted of 46 men (76.7%) and 14 women with a mean age of 28.9 years (range, 14.8-55.3 years). The groups were equivalent in terms of age, sex, tibial tunnel diameter, and follow-up time (Table 1), but the tunnel diameter was slightly larger in the ASF group compared with the RIS group (mean, 0.6 mm; $P < .01$).

Surgical Technique

The patients all underwent surgery under general anesthesia with the same surgical technique.[12] An air tourniquet was applied to the limb with a pressure of 300 mm Hg before harvesting the semitendinosus using a tendon harvester (Conmed Linvatec). The tendon was then folded to form a 4-strand graft that was stitched along its entire length. The femoral and tibial tunnels were prepared using compactors rather than drills to maximize bone density at their boundaries. The femoral tunnel was created inside out from the anteromedial portal in an anatomic position (insertion of anteromedial fibers of the ACL),[13,48,49] and suspensory femoral fixation was used for all patients. The tibial tunnel was created outside in at the center of the anteromedial bundle footprint, and its diameter was matched size for size with the graft diameter. In 67 knees, the graft was fixed within the tibial tunnel using the PULLUP (SBM) ASF system (ASF group). In 54 knees, the graft was fixed within the tibial tunnel using LIGAFIX 60 (SBM) RIS (RIS group), which were always 1 mm larger in diameter than the tunnel created to improve fixation strength and were composed of poly-L-lactic acid (PLLA) combined with 60% β-TCP.

Postoperative Rehabilitation

All patients followed the same nonaggressive rehabilitation protocol with immediate weightbearing with crutches, a fully mobile brace, and complete range of motion. Cycling and swimming were permitted at 6 weeks, jogging at 3 months, and cutting or pivoting sports at 6 months.

Postoperative Assessment

At 6 months postoperatively, patients were assessed clinically and with MRI. Side-to-side laxity measurements were made using GNRB 150 N (GeNouRoB). Graft behavior in the tibial tunnel was analyzed using fat-saturated MRI sequences in sagittal and axial planes. The scans were obtained using a 1.5-T superconducting magnet (Signa; GE Healthcare) with a dedicated surface coil. Imaging was confined to a thickness of 2 mm. Proton density–weighted images were acquired with the standard spin echo technique (1000-ms repetition time and 20-ms echo time).

All graft measurements were taken with the RadiAnt DICOM Viewer 1.9.16 (Medixant). We used 2 windows: one for the sagittal view and another for the axial view. In the first window, the sagittal section was selected in which the tibial tunnel was cut longitudinally, showing the maximum section of the graft and device. The tibial tunnel was then divided in 3 equal parts using the measurement tools. The corresponding axial sections were selected in the second window in the middle of each sagittal view of the tunnel.

The following parameters were assessed:

1. Tibial tunnel length and diameter compared with the initial drilled diameter recorded in the surgical case reports.
2. Graft filling and tibial tunnel filling (cross-sectional area) and contact (cross-sectional perimeter), the volume occupied by the graft within the tunnel, and the extent of graft-bone contact (complete, three-quarters, half, a quarter, or none of the tunnel wall).
3. Length of the graft portion that is not in contact with the fixation device.
4. Cortical formation at the tunnel wall and closure of the distal tunnel aperture.
5. MRI graft signal to assess graft quality and homogeneity as graded by Howell et al,[20] who demonstrated that the signal intensity correlates with water content in the zone, which would indirectly and inversely correlate with graft maturity, although there are no published studies that demonstrate this indirect association. Grade I ("normal") designated a zone filled with a graft that had a homogeneous low-intensity signal indistinguishable from that of the PCL or patellar tendon, grade II designated a volume of the graft analyzed on multiple slices of at least 50% of the normal ligament signal intermixed with portions of the graft that had acquired increased signal intensity, grade III designated a graft within a zone that had less than 50% of its volume exhibiting a normal-appearing ligament signal, and grade IV designated a diffuse increase in the signal intensity with no normal-appearing strands of ligament (Figure 1).
6. The signal-to-noise quotient (SNQ = *graft signal − PCL signal / background signal (air)*)[30,56]: the signal intensity was measured by selecting a region of interest (ROI) centered around the graft in the sagittal view of the tibia at the level where the graft had best visibility (Figure 2). Circular ROIs with a diameter of 25 pixels were evaluated for measuring the signal of the PCL and background. The background ROI was placed approximately 1 cm anterior to the inferior part of the patella in the sagittal view. Intratunnel graft signal intensity was additionally measured independently on the third proximal part of the tibial tunnel (Figure 3). The SNQ was calculated with a measurement accuracy of 1 decimal digit. Lower SNQ ratios indicate lesser water content and theoretically better graft maturity and healing.

Statistical Analysis

Sample size analysis indicated that a minimum of 49 patients per group was required to detect a significant difference in the SNQ between the 2 groups, assuming an SD of 0.7 and a difference of 0.5 between the groups. For the power analysis, the α value was set at 0.05 and the β value at 0.8. Statistical analyses were performed using R version 3.2.2 (R Foundation for Statistical Computing). Descriptive statistics were used to summarize the data. Data were not normally distributed. Between-group differences were evaluated using the Wilcoxon rank-sum test (Mann-Whitney *U* test). Assumptions for individual tests were checked before the analyses were performed. P values <.05 were considered statistically significant.

RESULTS

The mean side-to-side difference in anteroposterior laxity was 0.62 ± 2.13 mm in the ASF group and 1.29 ± 1.48 mm in the RIS group (Table 2), but the difference was not statistically significant (P = .198). The mean graft length free of the fixation device within the tunnel was 3 times greater in the ASF group than in the RIS group (21.5 ± 4.52 mm and 6.9 ± 4.75 mm, respectively; P < .001). The mean tibial tunnel length was 37.7 ± 3.91 mm in the ASF group and 38.8 ± 3.77 mm in the RIS group (P = .06). The mean tunnel enlargement in the ASF group was 2.3 ± 1.13 mm (range, 0.5-6.0 mm), while in the RIS group, it was 4.7 ± 2.81 mm (range, 0.5-19.0 mm) (P < .001). Cyst formation was observed around

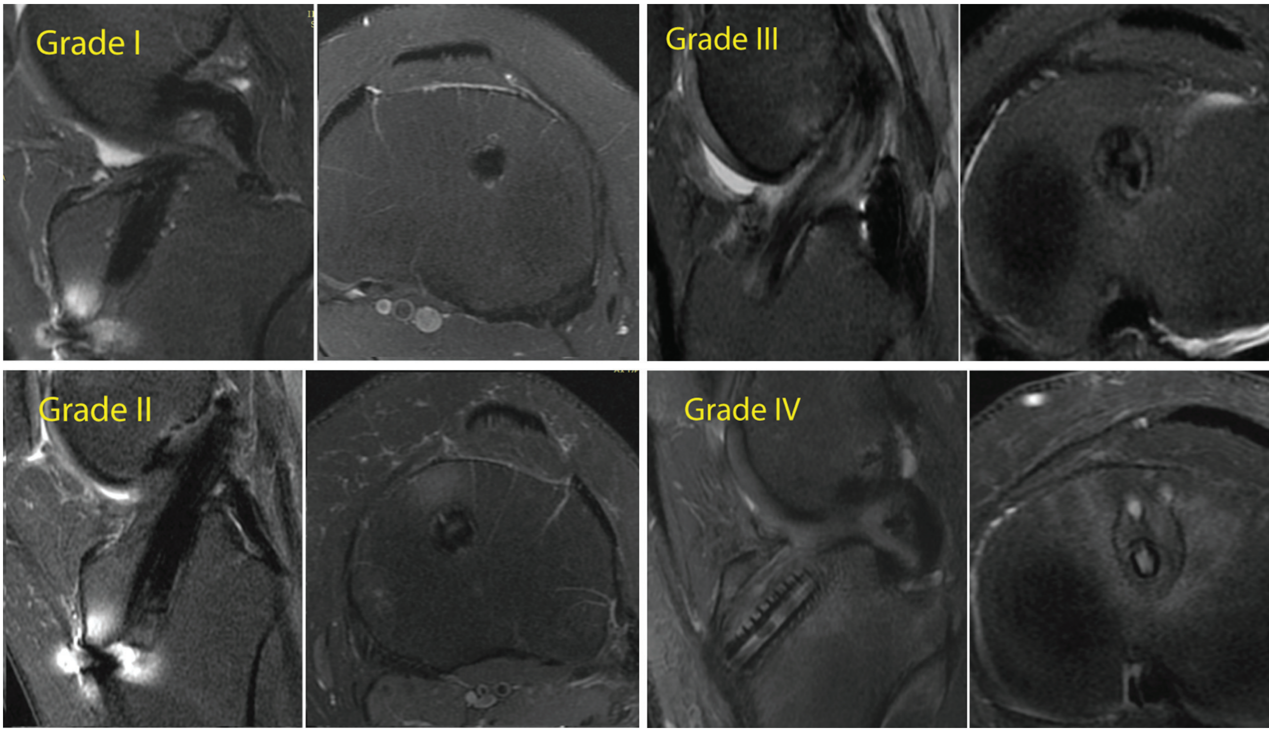

Figure 1. Graft signal examples on magnetic resonance imaging as graded by Howell et al[20] (grades I-IV).

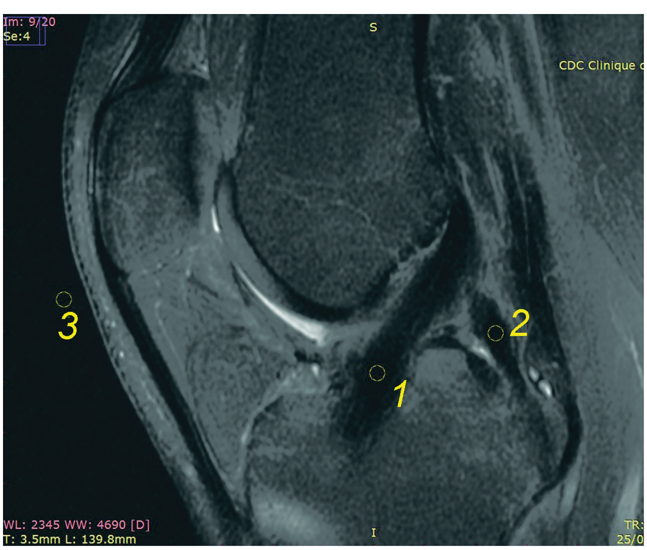

Figure 2. Method for determining the signal-to-noise quotient. Sagittal T2 image showing (1) a region of interest with a diameter of 25 pixels, (2) the posterior cruciate ligament, and (3) background signal.

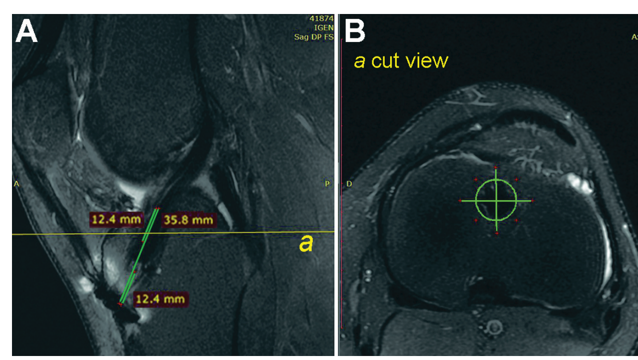

Figure 3. Different measurements achieved with RadiAnt software. (A) Sagittal T2 view: the 3 parts of the tibial tunnel, with line *a* showing the cut level for the axial view. (B) Axial T2 view: the tibial tunnel is divided in 4 parts to calculate graft tunnel filling and graft-bone contact.

the tibial tunnels in 7 knees of the RIS group in which the tunnel diameter was considerably larger than in the rest of the group (7.2 ± 5.6 mm).

The tibial tunnel aperture was completely closed by bone (Figure 4) in 33% of the ASF group and in 4% of the RIS group. Salient or partial cortical formation around the tunnel edge was observed in sagittal or frontal views (Figure 5) in significantly more knees of the RIS group (61.2%) than of the ASF group (21.7%) ($P < .001$).

Tibial tunnel filling by the graft, assessed in axial views, was over three-quarters of the cross-sectional area in 91.6% in the ASF group and 73.5% in the RIS group ($P = .029$). Tibial tunnel contact with the graft was over three-quarters of the tunnel perimeter in 79.7% in the ASF group versus 75.5% in the RIS group ($P = .068$).

The Howell graft signal assessment findings were excellent in 97% of knees in the ASF group and in 25% of knees

TABLE 2
Clinical and Radiographic Outcomes (N = 109 Patients)

	ASF Group (n = 60; 55%)	RIS Group (n = 49; 45%)	*P* Value
Side-to-side difference in laxity, mm	0.62 ± 2.13 (−6.3 to −3.9)	1.29 ± 1.48 (−2.3 to 5.4)	.198
Length of graft free of fixation device, mm	21.5 ± 4.52 (13.0 to 32.0)	6.9 ± 4.75 (0.0 to 19.0)	<.001[b]
Total tunnel length, mm	37.7 ± 3.91 (27.0 to 45.0)	38.8 ± 3.77 (32.0 to 48.0)	.060
Tunnel enlargement, mm	2.3 ± 1.13 (0.5 to 6.0)	4.7 ± 2.81 (0.5 to 19.0)	<.001[b]
Tunnel aperture closed, %	33.0	4.0	<.001[b]
Cortical formation, %			<.001[b]
Present	1.7	22.4	
Partial	20.0	38.8	
Absent	78.3	38.8	
Tunnel filling by graft, %			.029[b]
Complete	33.3	38.8	
Three-quarters	58.3	34.7	
Tunnel contact with graft, %			.068
Complete	33.9	28.6	
Three-quarters	45.8	46.9	
Howell grade I, %	97.0	25.0	<.001[b]
Signal-to-noise quotient	0.078 ± 0.62 (–0.5 to 4.0)	0.671 ± 0.83 (–0.2 to 4.2)	<.001[b]

[a]Data are reported as mean ± SD (range) unless otherwise indicated. ASF, adjustable suspensory fixation; RIS, resorbable interference screws.
[b]Statistically significant difference between groups ($P < .05$).

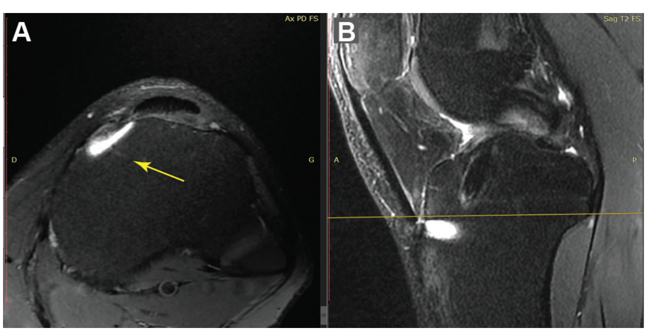

Figure 4. Tunnel bone filling of a knee in the adjustable suspensory fixation (ASF) group: (A) Arrow showing distal tibial bone tunnel vanishing. (B) Sagittal view with the horizontal line corresponding to the axial cut level. The white spot is caused by the metal platelet part of the ASF system.

in the RIS group ($P < .001$) (Figure 6). The mean SNQ was 0.078 ± 0.62 in the ASF group and 0.671 ± 0.83 in the RIS group ($P < .001$).

DISCUSSION

The most important finding of the present study was that MRI revealed better conditions for graft incorporation within the tibial tunnel using ASF than RIS. Both the Howell graft grade and SNQ were significantly better in the ASF group, indicating greater graft maturity and healing. The second hypothesis was also validated, as there was little tunnel enlargement observed using ASF, in contrast with the literature on distal fixation devices.[31] This is possibly because of adequate stabilization of the ASF

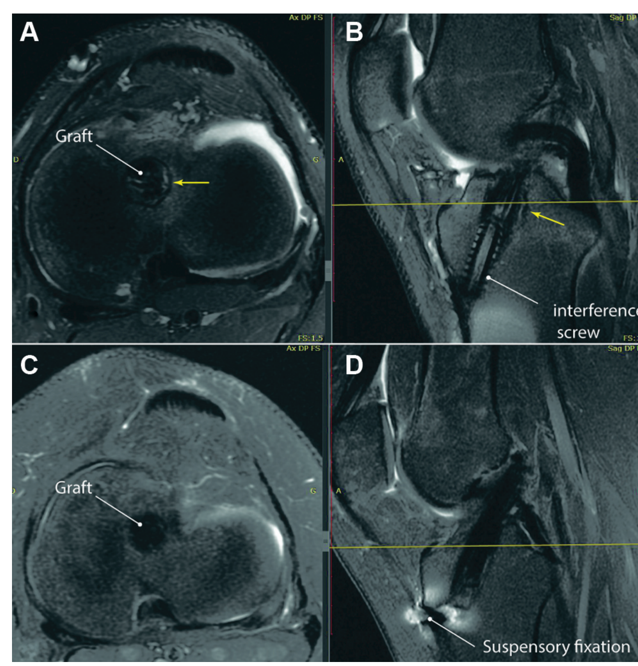

Figure 5. (A, B) Knee in the resorbable interference screws (RIS) group with tunnel edge corticalization (arrows); note the high signal between the graft and bone. (C, D) Knee in the adjustable suspensory fixation (ASF) group with complete bone graft filling and no edema in the surrounding bone.

system and nonaggressive rehabilitation, although direct correlations with the latter parameters were not specifically investigated in this study. Tunnel enlargement in the RIS group was likely caused by oversizing of screws, which were always 1 mm larger in diameter than the

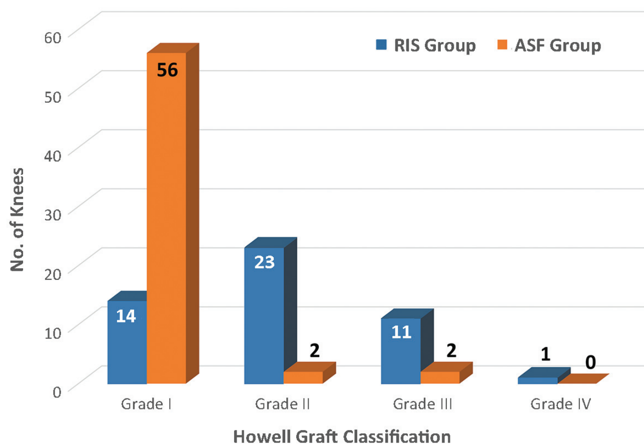

Figure 6. Howell graft classification on magnetic resonance imaging: almost all the adjustable suspensory fixation (ASF) group knees were graded I. In the resorbable interference screws (RIS) group, only 28.6% were grade I, 46.9% were grade II, 22.4% were grade III, and 2.0% (1 knee) were grade IV.

tunnel prepared to improve fixation strength, especially in women in whom the tibial cancellous bone is often relatively soft.[8,10] The second reason for enlargement may be the acidic degradation of the resorbable polymer, which caused the formation of cysts in 7 knees (14.3%), consistent with findings reported in the literature.[26,41]

In the majority of the ASF group, the graft had excellent contact with the bone tunnel edge without any adverse bony reactions, and graft behavior was better than in the RIS group. The SNQ in this study was rather good compared with that in other studies. In rare cases, the SNQ calculated was negative, albeit of a very small magnitude, because the graft signal was slightly stronger than the PCL signal. Muramatsu et al[38] reported SNQ values of 0.74 ± 0.67 in a group of 10 healthy ACLs, while Ma et al[29] stated that hamstring autografts fixed with interference screws in the tibial tunnel had intra-articular SNQ values at 6 months of 1.5 ± 0.6. Our results in the ASF group were significantly lower than these results, which may be caused by superior graft homogeneity and tight graft folding that avoided synovial fluid penetration in the graft-bone interface.[12] In the majority of hamstring grafts, the different strands are separated, which allows joint fluid to penetrate, thereby increasing the SNQ value. The MRI sagittal view of a healthy ACL frequently did not appear as homogeneous as the PCL signal perhaps because the inclination of the ligament made it difficult to obtain a strict 2 mm–thick sagittal view.

In 2001, Weiler et al[56] investigated the value of contrast-enhanced MRI for the prediction of ACL graft vascularity and biomechanical properties and found a significant negative linear correlation between biomechanical parameters and the SNQ. Our measurements were performed without contrast agents, and therefore, we cannot extrapolate our SNQ values to estimate biomechanical properties, as the increased MRI signal is probably related to an increase in water concentration, representing graft edema.[18,45] McFarland et al[34] showed that graft edema is a proven marker for following changes in graft strength in dogs. There is thus an inverse relationship in which an increase in the graft signal indicates a decrease in graft strength. Consequently, the low signal observed in the ASF group (low SNQ) indicates the absence of graft edema and thus improved graft remodeling at 6 months. MRI is frequently used to assess ACL grafts; however, there is still controversy about the applicability of MRI for detecting graft integrity.[5,24,33,44,53] It would be interesting to carry out an additional study using gadolinium-diethylenetriamine penta-acetic acid (Gd-DTPA)–enhanced imaging to evaluate vascularization of the graft. Regarding bone graft filling, the ASF group demonstrated better graft behavior than the RIS group. In the large majority of ASF knees, the graft filled completely or three-quarters of the tunnel, and the Howell grading was much better. Howell et al[20] reported in their study that graft grading decreased from 0 to 3 months and became stable after this period.

Another aspect that contributed to our conclusion that graft behavior is excellent in the ASF group was that we noticed very slight bony densification of the tunnel: 1.7% compared with 22.4% in the RIS group. This result indicated very good contact between the graft and bone, which reflected good bony integration of the graft. The less bony densification is also because of the absence of joint fluid in the tunnel. Mechanical studies have shown that the suspensory fixation system is the strongest one among all other systems.[6] Thus, these combined qualities place suspensory fixation in an excellent situation as the preferred fixation choice when using the hamstring tendon as a graft.

The main limitation of this study was the difficulty in obtaining a perfect MRI sagittal section because of the mediolateral inclination of some tibial tunnels. Another limitation was that interobserver and intraobserver agreement of MRI measurements were not calculated. However, our samples were large, the 2 groups were homogeneous, the operations were performed by experienced surgeons working at the same center, and all patients followed the same rehabilitation protocol. With this said, the study design with one surgeon performing all ASF procedures and another surgeon performing all RIS procedures could be a source of bias with the outcomes. Only tibial graft fixation differed. Our assessment was carried out at 6 months, and it is known that autograft maturation continues to progress afterwards and so could tunnel widening. Furthermore, the short follow-up did not permit the collection of reliable clinical scores in addition to laxity measurements, which could provide further evidence regarding the potential benefits of each fixation system. Thus, our study cannot determine whether there are clinical differences in outcomes that correspond to the differences seen in the imaging parameters. Lane et al[25] reported that the histological and biochemical properties of tendon grafts were similar to those of the healthy ACL at 4 years after surgery. Falconiero et al[15] investigated vascularity, cellularity, metaplasia, and the fiber pattern by arthroscopic biopsy and reported similarity between the graft and healthy ACL at 12 months.

CONCLUSION

Six-month MRI assessments revealed that ASF provides more favorable conditions than RIS for the incorporation and ligamentization of 4ST grafts within the tibial tunnel. The ASF system used showed very little tunnel widening in comparison to RIS, which suggests that the former grants stabilization. The SNQ was also considerably better in the ASF group possibly because of tight graft folding that prevented synovial fluid penetration in the graft-bone interface. Clinicians may find the results of this study interesting and clinically relevant in understanding how the choice of graft fixation influences graft behavior and the healing process in the tibial tunnel when using the hamstring tendon as a graft in ACL reconstruction.

ACKNOWLEDGMENT

The authors are grateful to Mo Saffarini (Alliance Scientifique SAS) for his assistance with article preparation and editing.

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