Evaluation of Graft Ligamentization by MRI After Anterior Cruciate Ligament Reconstruction

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Background: The tendon graft used in anterior cruciate ligament reconstruction (ACLR) undergoes "ligamentization" after implantation, and the reported length of this process varies from 6 to 48 months. Some grafts have ruptured at subsequent follow-up evaluations. Although the progress of graft ligamentization can be followed with postoperative magnetic resonance imaging (MRI) for reassessment, it is not known whether a delay in ligamentization (as reflected by a higher signal of the graft) is associated with an increased chance of subsequent graft rupture.

Hypothesis: Signal intensity of the graft on reassessment MRI (signal-noise quotient [SNQ]) would be associated with the incidence of future graft rupture at subsequent follow-up.

Study Design: Case-control study; Level of evidence, 3.

Methods: A total of 565 ACLRs with intact graft on first-time reassessment MRI after surgery were followed for a mean period of 67 months. The rates of 1-year and 2-year follow-up were 99.5% and 84.5%, respectively. The signal intensity of the intact graft on the first-time reassessment MRI was evaluated (1) quantitatively by the SNQ and (2) qualitatively with the modified Ahn classification. Among the 565 ACLRs, 23 additional graft ruptures developed during a time interval of 7 months to 9 years after the surgery.

Results: Higher SNQ was associated with increased chance of subsequent graft rupture (SNQ 7.3 $\pm$ 6 for subsequent graft rupture vs 4.4 $\pm$ 4 for grafts without subsequent rupture; P = .004, Mann-Whitney U test). The other important confounders that were associated with increased chance of graft rupture were younger age at the time of ACLR ($P < .001$) and longer follow-up time (P = .002). Multiple linear regression showed that all 3 factors (higher SNQ, younger age, and longer follow-up) were independent predictors of graft rupture (SNQ, P = .03; age, $P < .001$; follow-up, P = .012). When the reassessment MRI was performed in the second year after ACLR, the odds ratio of future graft rupture of a heterogeneous hyperintense graft when compared with a homogeneous hypointense graft was 12.1 (95% CI = 2.8 to 52.6) $P < .001$, Fisher exact test).

Conclusion: Higher signal intensity of the intact graft on reassessment MRI (higher SNQ and heterogeneous hyperintense graft) was associated with increased chance of subsequent graft rupture.

Keywords: anterior cruciate ligament reconstruction; ligamentization; graft rupture; MRI

During anterior cruciate ligament (ACL) reconstruction (ACLR), a tendon graft is implanted into a patient's knee to replace the torn ACL. When healing of the graft occurs, the histological appearance and biomechanical properties of the implanted graft gradually change from a tendon-like structure to a ligament-like tissue. This remodeling process is called *ligamentization*.[3] Although tendons and ligaments share many structural similarities, their precise biochemical composition, histological structure,[2] and biomechanical properties[33] are different. The mechanical strength of the graft is transiently reduced at the time of ligamentization.[19] Additionally, the process of ligamentization is lengthy and can last for >2 years, as shown in a human biopsy study.[36]

The implanted graft is notorious for its tendency to rupture even after a successful ACLR. Studies have shown that the rate of graft rupture and revision surgery range

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from 3% to 5% in short-term to midterm follow-up.[12,39,42] The rate of revision surgery within the first 2 years can be as high as 5%.[39] The graft rupture rate reported in a meta-analysis of 47,613 ACLRs at a mean follow-up of 5 years was close to 3%.[42] In the Scandinavian national registry of 45,998 ACLRs, it was reported that the expected proportion of revision was around 2% at 2 years and 4% at 5 years after surgery.[12] The incidence of graft rupture increases with time. In midterm to long-term follow-up, the cumulative rate of graft rupture increased to 7.9% with follow-up of up to 10 years.[31] The rate of graft rupture was around 8% in patients with ACLR with bone–patellar tendon–bone (BPTB) autograft and 13% in those with hamstring (HS) autograft.[37] In studies with very long-term follow-up, the risk of graft rupture of BPTB autograft was 8% at 15-year follow-up[17] and that of HS autograft was 21% at 20-year follow-up.[41]

At the time of ligamentization, the signal intensity of the graft on magnetic resonance imaging (MRI) is increased.[15] Muramatsu et al[32] reported that the signal-noise quotient (SNQ) value increased at 1 month, peaked at 4 or 6 months, and then decreased continuously from 12 to 48 months. The increase in graft signal is secondary to an increase in vascularity and graft edema during ligamentization.[16] Hence, the signal intensity of the graft measured on postoperative MRI scans has been used extensively as a tool to follow the progress of graft maturation in ACLR.[§] The SNQ is one of the most commonly used MRI techniques to assess graft maturity.[32] Large-animal studies (sheep model) demonstrated the association of SNQ with the progress of histological changes and biomechanical strength of the graft during ligamentization.[46]

Although SNQ is extensively used as a research tool to study the effect of different risk factors on graft ligamentization in ACLR, the literature provides a paucity of data about the time to completion of ligamentization with reference to the SNQ of the graft.[32,50] It was historically believed that ligamentization was completed radiologically by the end of 12 months after ACLR.[15] However, recent longitudinal MRI studies suggest that the time for completed ligamentization is likely much longer. SNQ peaked at 4 to 6 months after ACLR, then gradually decreased, but only started to plateau at 18 to 24 months after the index ACLR with autograft. However, for allogenic tendons, SNQ continued to increase until 18 to 24 months after surgery (Table 1).[‖] Moreover, the level of SNQ representing the completion of ligamentization is not known. In most MRI studies, the reported SNQ at the final follow-up was much larger than that of the native ACL (Table 2).[¶] Furthermore, limited data are available regarding the SNQ of intact graft at midterm follow-up, 3 to 5 years after ACLR.[32,50]

The rate of graft ligamentization in ACLR varies greatly, ranging from 1 year to 48 months in human studies.[36] Some grafts demonstrate prolonged ligamentization in terms of increased SNQ when compared with others.[#] It is likely that the maturation process of the graft is affected by many factors, including the type of graft used (eg, autograft vs allograft,[10,24,32] BPTB vs HS[11]), the surgical technique used (eg, transportal vs transtibial femoral tunnel drilling,[40] concomitant anterolateral ligament reconstruction[38]), the quality of the operation (eg, sagittal graft angle,[35] graft bending angle[26]), the use of biological augmentation (eg, ACL remnant preservation,[1] platelet-rich plasma application[13]), and host factors (eg, age,[39] activity level,[39] smoking status[18]). Although it has been observed that the cumulative graft rupture rate continues to increase in the first 2 years after ACLR,[31] it is not known whether the grafts that experience prolonged ligamentization are those at risk of future graft rupture. It is possible that the risk factors leading to a delay in ligamentization contribute to an increased risk of graft rupture in longer follow-up. However, there are currently no studies investigating the possibility of using SNQ to predict the chance of subsequent graft rupture.

The current study was conducted to answer the following research questions:

1. How much time is required for complete ligamentization of the ACLR graft with reference to the SNQ of the graft measured on postoperative MRI?
2. Will signal intensity of the intact graft on the reassessment MRI be able to predict the risk of future graft rupture at subsequent follow-up?
3. If the signal intensity of the graft is a predictor of future graft rupture, what is the best time to perform reassessment MRI to predict this?

We hypothesized that the signal intensity of the graft on reassessment MRI scans (the SNQ) would be associated with future graft rupture. The hypothesis was tested in a cohort of 565 ACLRs with intact graft found on postoperative reassessment MRI, with a mean follow-up of >5 years. The primary outcome assessed was SNQ of the intact graft on reassessment MRI. The secondary outcome was the qualitative assessment of graft maturation using a modified version of the Ahn classification.[1]

METHODS

The current study was a retrospective review of 765 ACLRs performed by 2 fellowship-trained sports medicine surgeons (including W.P.Y.) in the authors' institution between July 2007 and December 2019. The study was approved by the ethics committee of the authors' institution (approval document No. UW 22-250). Postoperative MRIs were routinely arranged for all patients receiving ACLR as part of the research protocol. A majority of patients recruited in this study underwent only 1 postoperative reassessment MRI scan, which was performed at a mean of 18 months after the index operation. Sequential MRI at fixed time points was not performed. Patients were assessed in a designated preoperative assessment clinic 1

[§]References 5-7, 10, 11, 13, 14, 22, 23, 26-28, 32, 34, 35, 38, 40, 43, 47-49.

[‖]References 10, 14, 24, 28, 32, 40, 47, 49, 50.

[¶]References 5-7, 10, 11, 13, 14, 22, 23, 26-28, 32, 34, 35, 38, 40, 43, 47-49.

[#]References 5-7, 10, 11, 13, 14, 22, 23, 26-28, 32, 34, 35, 38, 40, 43, 47-49.

TABLE 1
Change of Mean SNQ of the ACLR Graft Over Time[a]

Lead Author (Year)	No. of Knees	Type of Graft	SNQ at 6 mo	SNQ at 12 mo	SNQ at 18 mo	SNQ at 24 mo	SNQ at 36 mo	SNQ at 48 mo	
Muramatsu (2008)[32]	20	Autograft	2.5	2	—	1.6	1.7	0.6	
	24	Allograft	0.5	1.1	—	1.4	0.7	0.8	
Wen (2020)[47]	108	Autograft	4.4	3.6	—	3.3	—	—	
Zhang (2020)[49]	22	Autograft	8.7	6.6	—	4.5	—	—	
Liu (2018)[28]	37	Autograft	20	16	—	15	—	—	
Grassi (2021)[14]	20	Autograft	2.8	—	2.3	—	—	—	
Cusumano (2022)[10]	23	Autograft	13	9.8	—	—	—	—	
	24	Allograft	7.9	10.4	—	—	—	—	
Li (2017)[24]	21	Autograft	25	18	—	—	—	—	
	17	Allograft	29	20					
Saito (2019)[40]	87	Autograft	6.9	4.8	—	—	—	—	
			SNR at 6 mo	SNR at 12 mo		SNR at 24 mo			SNR at 60 mo
Zhang (2020)[50]	37	Autograft	17	14		11			11

[a]ACLR, anterior cruciate ligament reconstruction; SNQ, signal-noise quotient; SNR, signal-noise ratio. Dashes indicate data not available.

TABLE 2
Report of Signal-Noise Quotient in the Literature[a]

Lead Author (Year)	Native ACL or ACLR Graft	Type of Graft	Type of ACLR	No. of Knees	Serial MRI	Time of MRI After Surgery, mo	SNQ, Mean or Mean ± SD
Muramatsu (2008)[32]	Native ACL	NA	NA	10	NA	NA	0.76 ± 0.67
	BPTB	Autograft	SB	20	Yes	36	1.7
	BPTB	Allograft	SB	24	Yes	36	0.7
Fukuda (2022)[11]	BPTB	Autograft	SB	30	No	24	2.3 ± 0.5
	HS	Autograft	DB	45	No	24	2.9 ± 0.9 (AM)
							0.9 ± 1.1 (PL)
Bouguennec (2021)[5]	HS	Autograft	—	139	No	24	2 ± 3.5
Wen (2020)[47]	HS	Autograft	SB	108	Yes	24	3.3 ± 1.1
Zhang (2020)[49]	HS	Autograft	SB	22	Yes	24	4.5
Lee (2018)[23]	HS	Autograft	SB	100	No	24	3.3 ± 0.7
Liu (2018)[28]	HS	Autograft	SB	37	Yes	24	15 ± 43
Grassi (2021)[14]	HS	Autograft	SB	20	Yes	18	2.3
Zhang (2021)[48]	HS	Autograft	SB	69	No	18	1.7 ± 0.8
Gong (2022)[13]	HS	Autograft	SB	60	No	12	1.7 ± 0.7
Cusumano (2022)[10]	HS	Autograft	SB	23	Yes	12	9.8 ± 7.1
	Peroneus	Allograft	SB	24	Yes	12	10.4 ± 8
Okutan (2022)[34]	HS	Autograft	SB	94	No	12	4.5 ± 2.3
Cavaignac (2020)[7]	HS	Autograft	SB	62	No	12	3.2 ± 4
Oshima (2020)[35]	HS	Autograft	SB	98	No	12	2.6 ± 1.5
Li (2019)[26]	HS	Autograft	—	43	No	12	19.4
Li (2019)[27]	HS	Autograft	SB	42	Yes	12	8.1 ± 4.6
Saito (2019)[40]	HS	Autograft	DB	87	Yes	12	4.8 ± 3.5
Takahashi (2019)[43]	HS	Autograft	DB	65	No	12	21.7 ± 14.6
Cavaignac (2018)[6]	HS	Autograft	SB	62	No	12	5.6 ± 4.1
Lee (2016)[22]	HS	Autograft	—	23	No	12	3.8
Rojas (2021)[38]	HS	Autograft	SB	52	No	10	5.9 ± 4.6
Current study				565	No	18	4.5 ± 4.2

[a]ACL, anterior cruciate ligament; ACLR, anterior cruciate ligament reconstruction; AM, anteromedial bundle; BPTB, bone–patellar tendon–bone; DB, double bundle; HS, hamstring; MRI, magnetic resonance imaging; NA, not applicable; PL, posterolateral bundle; SB, single bundle; SNQ, signal-noise quotient. Dashes indicate data not provided.

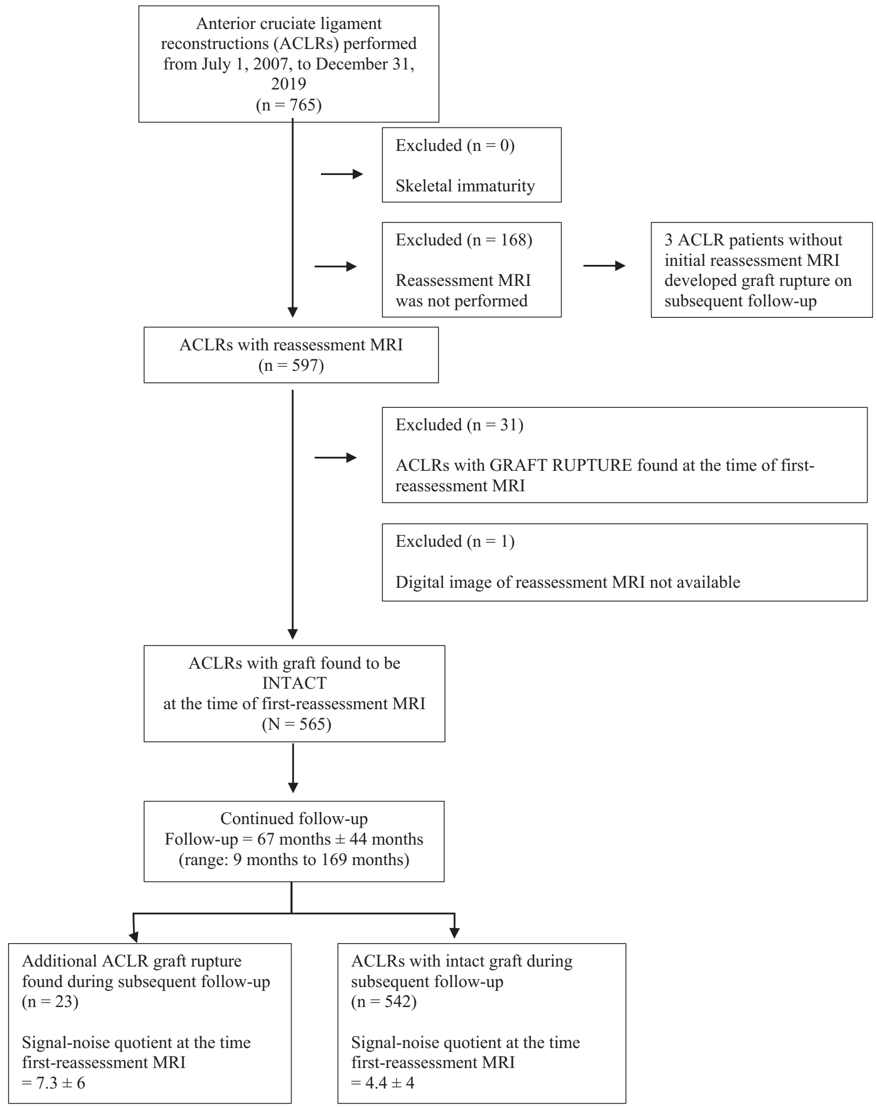

Figure 1. Enrollment of participants. ACLR, anterior cruciate ligament reconstruction; MRI, magnetic resonance imaging.

week before the surgery and were followed up every 3 months in the first year after ACLR and then annually. During the preoperative assessment clinic, the smoking status of the patients was recorded. Patients were classified as smokers (those who, prior to surgery, had the habit of smoking) and nonsmokers (those who, prior to the operation, never smoked). The rehabilitation protocol was standardized, and patients were advised to refrain from pivoting sports for 9 months. All patients received closed kinetic chain training in the first 3 months after surgery, followed by open kinetic chain training for the next 3 months and agility training in the last 3 months. The following clinical information was documented at the preoperative evaluation and each of the postoperative follow-up evaluations: Tegner activity score, International Knee Documentation Committee subjective score, history of repeated injury, and physical examination findings. All data were prospectively collected between 2007 and 2021. Cases were included if a graft was implanted into the patient's knee to reconstruct the injured ACL at the time of operation. Cases were excluded if (1) the patient was skeletally immature with open physis found on preoperative knee radiographs; (2) reassessment MRI was not performed after surgery; (3) graft rupture was found at the first-time reassessment MRI; or (4) digital image files of the reassessment MRI were not available.

Among the initial cohort of 765 ACLRs, 597 patients underwent postoperative reassessment MRI (78%). A total of 31 graft ruptures (5%) were found at the first reassessment MRI. The digital image of 1 reassessment MRI was not available. All patients were skeletally mature. After the exclusion criteria were applied, a total of 565 ACLRs with intact graft on first reassessment MRI were available as the sample of the current study (Figure 1).

TABLE 3
Patient Characteristics of the Whole Cohort and the Subgroup Analysis[a]

	Whole Cohort of the Study[b]	Subgroup Analysis[c]
Patients, n	565	167
Follow-up, mo	67 ± 44	68 ± 42
Time between reassessment MRI and surgery, mo	18 ± 9	19 ± 8
Age, y	27 ± 8	28 ± 9
Sex, male/female, n	462/103	128/39
Side, right/left, n	289/276	94/73
Body mass index	24 ± 3.5	24.8 ± 4.1
Generalized ligamentous laxity, n (%)	19 (3.4)	0 (0)
Smoker, n (%)	116 (21)	27 (16)
Preinjury Tegner activity scale score	6.5 ± 1.3	6.3 ± 1.3
Time lag between injury and date of surgery, mo	19 ± 36	24 ± 47
Multiligament injury, n (%)	33 (6)	0 (0)
EUA pivot shift 3, n (%)	50 (8.8)	8 (4.8)
Revision surgery, n (%)	35 (6)	0 (0)
Single bundle/double bundle, n	388/177	91/76
Concomitant ALLR, n (%)	109 (19)	0 (0)
Allograft, n (%)	8 (1.4)	0 (0)
HS/BPTB/QT/Other, n	501/44/15/5	167/0/0/0
Mode of fixation of femoral graft: suspensory/aperture, n	372/193	149/18
Mode of fixation of tibial graft: suspensory/aperture, n	57/508	1/166
Quantitative assessment of graft signal: SNQ	4.5 ± 4.2	4.3 ± 3.9
Qualitative assessment of graft signal: Ahn I/Ahn II/Ahn III, n	322/220/0	106/61/0

[a]Data are expressed as mean ± SD unless otherwise noted. ACLR, anterior cruciate ligament reconstruction; Ahn I, homogeneous hypointense graft; Ahn II, heterogeneous hyperintense graft; Ahn III, graft rupture; ALLR, anterolateral ligament reconstruction; BPTB, bone–patellar tendon–bone graft; EUA, examination under anesthesia; HS, hamstring graft; MRI, magnetic resonance imaging; Other, other soft tissue graft (eg, Achilles tendon, tibialis posterior, etc); QT, quadriceps tendon graft; SNQ, signal-noise quotient.

[b]Included both primary and revision ACLRs, all types of graft implanted, patients with known risk factors for graft rupture, and concomitant ALLR.

[c]Included primary ACLRs operated with hamstring autograft, in the absence of concomitant ALLR, multiple ligament injury, and generalized ligamentous laxity.

The mean follow-up period for the 565 ACLRs was 67 months (range, 9-169 months). The rates of 1-year and 2-year follow-ups were 99.5% and 84.5%, respectively.

The characteristics of the whole cohort are summarized in Table 3. The mean ± SD age of the patients at the time of operation was 27 ± 8 years. The male to female ratio was 4.5 to 1. The body mass index was 24 ± 3.5. The mean preinjury Tegner activity score was 6.5 ± 1.3. A total of 19 patients had generalized ligamentous laxity, and 21% of the patients were smokers. In total, 530 ACLRs were primary ACLRs and 35 were revision surgeries. The incidence of multiligament injury was 5.8%. All ACLRs except for 8 entailed autograft. HS autograft was the most common graft used (88.5%). BPTB graft was used in 44 cases (8%) and quadriceps tendon graft in 15 cases (2.7%).

All first-time reassessment MRI scans performed at the authors' institution used the same protocol with a 1.5-T MRI machine. The patient was instructed to lie in a supine position with the involved knee in full extension. The orientations of axial, sagittal, and coronal images were defined after acquisition of 2-dimensional scout images. MRI images were obtained with T1-weighted, T2-weighted, and fat-saturated proton density scanning sequences. The slice thickness of the sagittal image was 3 mm. Contrast was not applied.

Of all first-time reassessment MRI scans, 82.6% were performed in the second year and 8.7% were done in the third year after the index ACLR (Figure 2). The mean time of conducting the reassessment MRI was 18.3 ± 9 months. A total of 545 ACLRs underwent reassessment MRI within the first 3 years after surgery, and 20 ACLRs underwent reassessment MRI ≥4 years after the operation.

Among the 565 ACLRs with intact graft on first-time reassessment MRI, graft rupture occurred in 23 cases. The diagnosis of subsequent graft rupture was made by either diagnostic arthroscopy or repeated MRI. Among the 23 subsequent graft ruptures, 10 occurred within the first 2 years after the index operation and 5 occurred at 3 to 5 years of follow-up. An additional 8 ruptures occurred at follow-up of >5 years (Figure 3).

MRI Evaluation

The digital images of the first-time reassessment MRI were imported into Miele-LXIV Digital Imaging and Communications in Medicine workstation and viewer. The signal intensity of the intact graft in the T2-weighted sagittal images was assessed (1) quantitatively using the SNQ method and (2) qualitatively with the modified Ahn classification. The measurements were done in the absence of the knowledge of future graft rupture in subsequent follow-up.

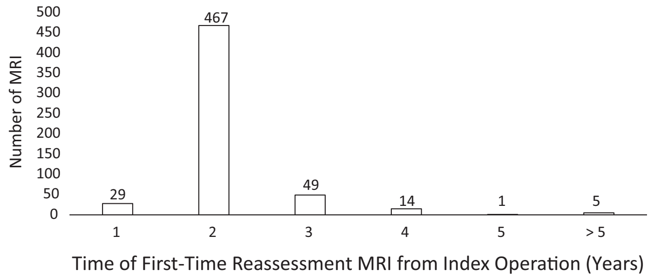

Figure 2. Time of first-time reassessment magnetic resonance imaging (MRI) after anterior cruciate ligament reconstruction.

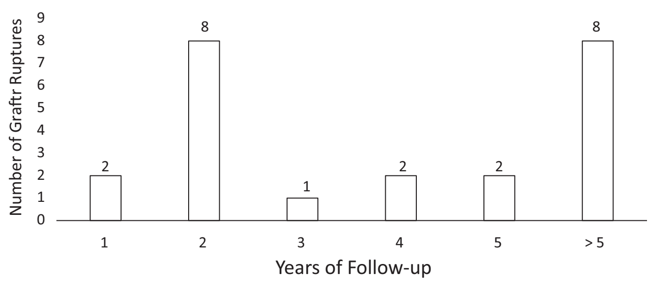

Figure 3. Number of additional graft ruptures during subsequent follow-up.

Signal-Noise Quotient

The contour of the intra-articular portion of the graft was outlined, and the mean signal per unit area of the whole graft was measured. The data were normalized with the mean signal of quadriceps tendon and the background (which was taken at about 2 cm anterior to the patellar tendon) (Figure 4). The normalized SNQ was calculated according to the following equation:

$$\mathrm{SNQ} = \frac{\text{Signal Of ACL graft}}{\text{Signal Of background}} - \frac{\text{Signal of quadriceps tendom}}{\text{Signal Of background}}.$$

Modified Ahn Classification

The signal of the graft was qualitatively assessed using the modified Ahn classification.[1] Grade 1 was a graft with homogeneous hypointensity comparable with the surrounding intact posterior cruciate ligament; grade 2 was a heterogeneous graft with areas of either focal or diffuse hyperintensity; grade 3 was either a homogeneous hyperintense graft with no normal appearing strands of ligament or complete absence of graft. Grade 1 represented completion of ligamentization, whereas grade 3 represented a ruptured graft (Figure 5).

Subgroup Analysis

A subgroup analysis was carried out in a homogeneous sample composed of primary ACLRs operated with HS

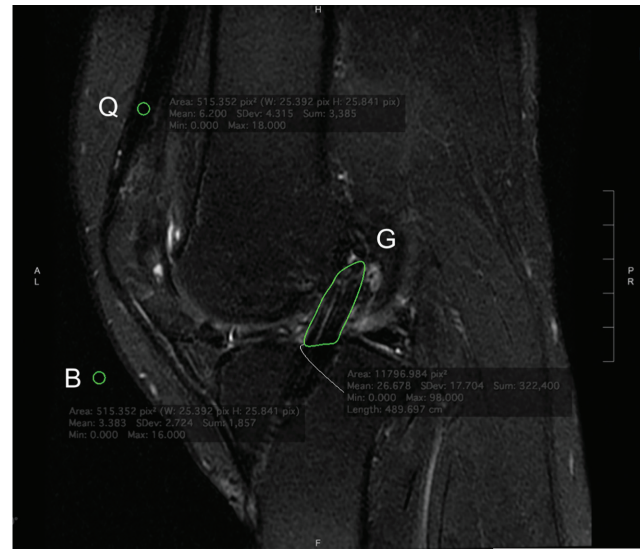

Figure 4. Measurement of signal-noise quotient of the whole graft.
B, mean signal of the background noise (mean signal per unit area of the background measured at 2 cm anterior to the patellar tendon)
G, mean signal of the whole graft (mean signal per unit area of the whole graft)
Q, mean signal of the quadriceps tendon (mean signal per unit area of the quadriceps tendon measured at 2 cm above its insertion at the upper pole of patella)

autograft. The inclusion criteria of the subgroup analysis were (1) primary ACLR and (2) HS autograft. The exclusion criteria of the subgroup were (1) patients having potential risk factors for graft rupture (ie, revision surgery, multiligament injury, generalized ligamentous laxity) and (2) patients with concomitant anterolateral ligament reconstruction. The characteristics of the subgroup analysis are reported in Table 3.

Interobserver and Intraobserver Repeatability

The interobserver and intraobserver repeatability tests were performed by the 2 authors. One author is a fellowship-trained sports medicine surgeon who has worked in the field of orthopaedic surgery for close to 30 years (W.P.Y.). The second author was a final year medical student (Y.C.C.). MRI assessment of signal-noise quotient and modified Ahn classification were performed by the first observer (W.P,Y,). To obtain interobserver repeatability, the measurements were repeated by a second independent observer (Y.C.C.) in 100 cases. Intraobserver repeatability was obtained by repeating the measurement at an interval of 1 week. The inter- and intraobserver errors were reported using intraclass correlation coefficient (ICC) for SNQ measurement and kappa coefficient for Ahn classification. The repeatability was considered to be excellent if the ICC was 0.75 to 1; good if 0.6 to 0.74; fair if 0.4 to 0.59; and poor if <0.4.[9] Concerning kappa coefficient,

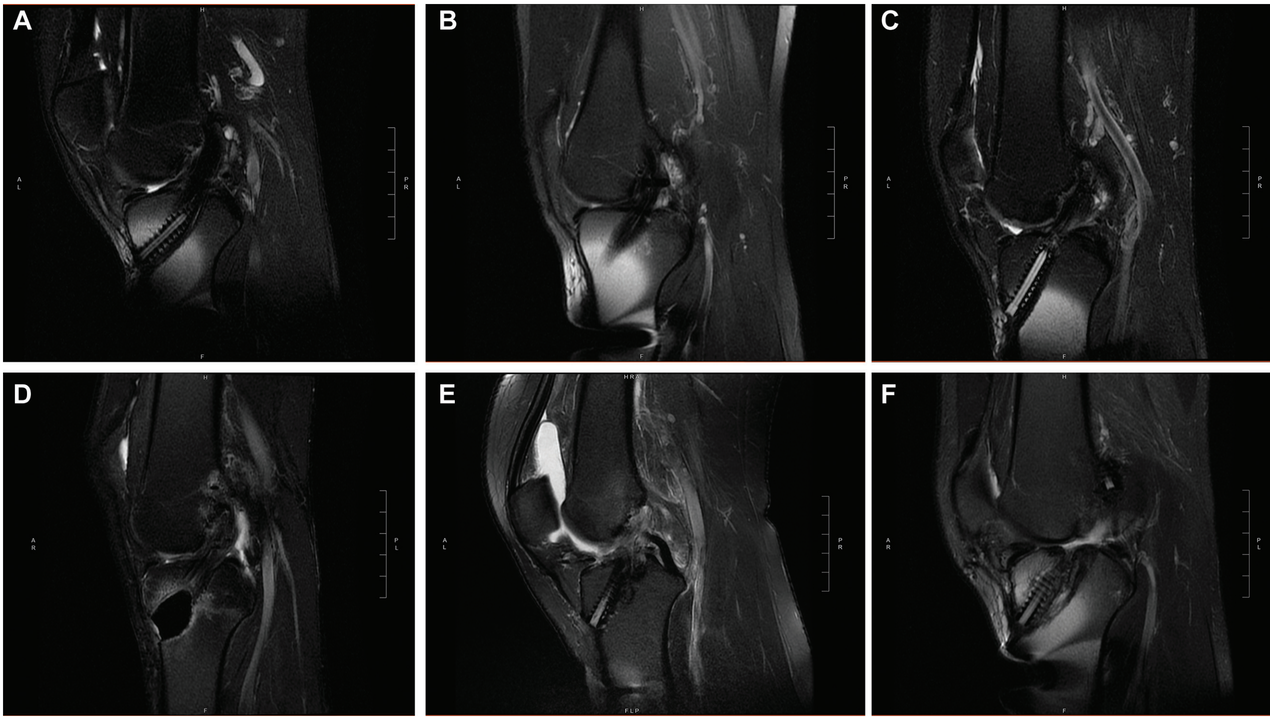

Figure 5. Qualitative assessment of graft maturation using the modified Ahn classification. (A) Grade 1A: homogeneous hypointensity comparable with the surrounding intact posterior cruciate ligament. (B) Grade 1B: near-homogeneous hypointensity with normal-appearing strands of ligaments. (C) Grade 2A: heterogeneous hyperintensity of graft with strands of ligaments visualized (focal increase in hyperintensity). (D) Grade 2B: heterogeneous hyperintensity of graft with strands of ligaments visualized (diffuse increase in hyperintensity). (E) Grade 3A: homogeneous hyperintensity of graft with absence of normal appearing strands of ligament. (F) Grade 3B: complete absence of graft. Grade 1 represented completion of ligamentization, whereas grade 3 represented a ruptured graft.

repeatability was considered to be almost perfect agreement if it was 0.81 to 1; substantial agreement if 0.61 to 0.8; moderate agreement if 0.41 to 0.6; fair agreement if 0.21 to 0.4; slight agreement if 0.01 to 0.2; and poor agreement if <0.[20]

Power Analysis

An interim analysis was carried out in a cohort of 183 ACLRs performed between 2011 and 2014. The mean SNQ in the intact graft on first-time reassessment MRI scans in the interim analysis was 6 ± 4.3. A total of 7 graft ruptures developed in subsequent follow-up of the interim analysis at a median follow-up of 7 years (3.8%). The mean SNQ measured during first-time reassessment MRI of the subsequent graft rupture group was 8.9. The power of the proposed study was calculated according to a type I error of .05, type II error of 0.8, and expected clinical difference of SNQ of 2.9. The enrollment ratio was set at 25. The minimal sample size required was 468.

Statistical Analysis

All statistical analyses were performed using SPSS (Version 28; IBM). Patient characteristics were reported using descriptive statistics. Possible statistical associations among variables were examined by chi-square test and independent t test, depending on whether the data were nonparametric or parametric. For parametric data with a sample size <25, repeated statistical examination was performed using Mann-Whitney U test. For comparison with reported data in the literature (eg, SNQ of native ACL), 1-sample Kolmogorov-Smirnov test was used. If >1 variable was found to be associated with subsequent graft rupture in further follow-up, multiple linear regression was conducted to determine the independent predictor of graft rupture. Statistical significance was assumed when $P < .05$.

RESULTS

Time to Complete Ligamentization of the ACLR Graft With Reference to the SNQ Measured on Postoperative MRI Scan

The mean signal-noise ratio of the whole graft measured on reassessment MRI was 4.5 ± 4.2 (range, 0-28.1). The SNQ of the graft differed significantly with reference to the years of reassessment MRI performed (P = .014; analysis of variance). The SNQs of the grafts were significantly

Figure 6. Signal-noise quotient of intact graft in reassessment magnetic resonance imaging (MRI) versus time of MRI.

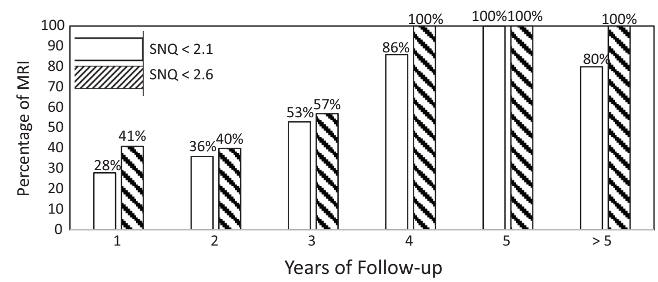

Figure 7. Percentage of reassessment magnetic resonance imaging (MRI) with graft signal-noise quotient (SNQ) within normal range.

higher when the reassessment MRI was done within the first 3 years compared with those performed from the fourth postoperative year onward (P < .001; Mann-Whitney U test) (Figure 6).

The reported SNQ of native ACL published in the literature was used as a reference value. Among 10 individuals with normal knees (age, 24-40 years), the mean SNQ of native ACL was 0.76 ± 0.67.[25] Significant difference was found between the SNQ of the ACLR graft and that of the normal ACL if the reassessment MRI was conducted within 4 years after ACLR. The SNQ of the graft was comparable with the SNQ of normal ACL only when the reassessment MRI was performed ≥4 years after the index ACLR (Appendix Table A1, available in the online version of this article).

The mean SNQ of the native ACL was 0.76 +/- 0.67. The value of the "mean SNQ + 2 SDs" was 2.1. The distribution of the SNQ of the intact graft below 2 SDs of the mean SNQ of native ACL (2.1) is shown in Figure 7. We noted a progressive increase in the percentage of grafts having an SNQ <2.1 with increasing number of years from the index operation, reflecting the increase in proportion of grafts reaching completed ligamentization at the time of reassessment MRI. We found a significant difference between the mean SNQ of the intact graft when the reassessment MRI was conducted within 3 years from the index operation (SNQ = 4.59 ± 4.3) and those that were performed at ≥4 years (SNQ = 1.25 ± 0.8) (P < .001; Mann-Whitney U test). The largest SNQ of the intact graft when the first-time reassessment MRI was performed ≥4 years after the index ACLR was 2.6. The distribution of the percentage of SNQ values ≤2.6 with reference to the year of the reassessment MRI is shown in Figure 7.

The mean time for performing the reassessment MRI was 18.3 ± 9 months. The Tegner activity score at the time MRI was 5.1 ± 1.9. We noted a weak negative association between activity level of the patient at the time of MRI and the SNQ (P = .011).

Ability of Signal Intensity of Intact Graft on Reassessment MRI to Predict Risk of Future Graft Rupture

Among the 565 ACLRs with intact graft on first-time reassessment MRI, 23 additional graft ruptures occurred in subsequent follow-up (at a mean of 44.7 months after the index operation; range, 7-107 months).

The SNQ of the grafts that developed subsequent graft rupture was significantly higher than the SNQ of those that did not (7.3 ± 6 vs 4.4 ± 4, respectively; P = .004, Mann-Whitney U test).

The comparisons of patient and surgical characteristics between the ACLRs that developed graft rupture in subsequent follow-up (n = 23) and those that did not (n = 542) are shown in Appendix Table A2 (available online). The group that developed subsequent graft rupture during further follow-up was younger at the time of ACLR (22 vs 27 years old, respectively; P < .001) and had longer follow-up (8.2 vs 5.5 years, respectively; P = .002). We found no difference between the 2 groups in terms of sex distribution, laterality of the operated limb, body mass index, presence of generalized ligamentous laxity, smoking status, preinjury activity level, time between injury and ACLR, concomitant multiple ligament injury, presence of significant pivot shift (grade 3) during examination under anesthesia, type of surgery performed (primary vs revision; single bundle vs double bundle; concomitant anterolateral ligament reconstruction or not), and type of graft used (autograft vs allograft; HS vs BPTB vs quadriceps tendon graft) (Appendix Table A2, available online).

Younger age (P < .001), longer follow-up (P = .002), and larger SNQ of graft at the time of reassessment MRI (P = .004) were associated with increased chance of graft rupture in further follow-up. Multiple regression analysis revealed that all 3 factors were significant independent predictors of subsequent graft rupture (age, P < .001; length of follow-up, P = .012; SNQ, P = .03).

Among the 565 ACLRs with intact graft found on first-time reassessment MRI, 325 grafts were classified by modified Ahn classification as having completed ligamentization and 240 grafts as having "hyperintensity," reflecting the presence of edema in the graft (Appendix Table A3, available online). Among the 240 grafts that showed hyperintensity on first-time reassessment MRI, 20 developed graft rupture in subsequent follow-up. In contrast, only 3 of the 325 grafts that showed complete ligamentization ruptured in subsequent follow-up. The odds ratio of graft rupture of the whole cohort (565 ACLRs) when hyperintensity was found in the graft on reassessment MRI compared

TABLE 4
Relationship of Graft Maturity of Intact Graft on First-Time Reassessment and Subsequent Graft Rupture in Further Follow-up[a]

Point When Reassessment MRI Was Conducted	Ahn Grade[b]	No. of ACLRs With Additional Graft Rupture on Subsequent Follow-up	No. of ACLRs With No Additional Graft Rupture on Subsequent Follow-up	*P* (Fisher exact test)
First year after index ACLR	Grade 1	0	18	NS
	Grade 2	1	10	
Second year after index ACLR	Grade 1	2	256	<.001
	Grade 2	18	191	
Third year after index ACLR	Grade 1	1	31	NS
	Grade 2	0	17	
≥4 years after index ACLR	Grade 1	0	17	NS
	Grade 2	1	2	

[a]ACLR, anterior cruciate ligament reconstruction; MRI, magnetic resonance imaging.
[b]Grade 1 indicates completed ligamentization. Grade 2 indicates a "hyperintense" graft.

with the graft that achieved complete ligamentization was 9.8 (95% CI, 2.9-33.2). We found no significant difference between the Tegner activity score of the 23 patients who developed additional graft rupture on subsequent follow-up and that of the 542 patients who did not (6.9 ± 1.1 vs 6.4 ± 1.4, respectively; *P* not significant, Mann-Whitney *U* test). Among the 23 grafts that developed additional graft rupture, no significant difference was found between the 20 grafts that had graft hyperintensity (ie, Ahn grade 2) and the 3 grafts that had normal homogeneous hypointensity (ie, Ahn grade 1) at the time of reassessment MRI in terms of the patient Tegner activity score before injury (7.1 ± 0.9 vs 5.7 ± 1.5, respectively; *P* = .092, Mann-Whitney *U* test), the Tegner score at the time of MRI (5.9 ± 1.7 vs 4.3 ± 1.5; *P* = .087, Mann-Whitney *U* test), and the Tegner score at the time of graft rupture (5.9 ± 1.6 vs 5 ± 1.7; *P* = .206, Mann-Whitney *U* test).

Optimal Time to Perform Reassessment MRI to Predict Risk of Future Graft Rupture in Subsequent Follow-up

The association of graft maturity on reassessment MRI and subsequent rupture with reference to the timing of first-time reassessment MRI is presented in Table 4. We found that the modified Ahn grade of the intact graft when the first reassessment MRI was conducted in the second year after the index operation was significantly associated with the incidence of future graft rupture ($P < .001$; Fisher exact test). This association was not observed when the MRI was done within the first postoperative year or from the third postoperative year onward. The odds ratio of graft rupture of the hyperintense graft when reassessment MRI was performed in the second year was 12 (95% CI = 2.8 to 52.6).

Concerning the 23 ACLRs that developed subsequent graft rupture on further follow-up, 3 were classified as grade 1 (ie, complete or near-complete ligamentization with homogeneous hypointense graft signal) and 20 as grade 2 (10 as heterogeneous hyperintensity with diffuse increase in signal intensity and 10 as heterogeneous hyperintensity with focal increase in signal intensity). The intra-articular portion of the graft was divided into the femoral one-third, the middle one-third, and the tibial one-third. Among the 10 grafts showing a focal increase in graft hyperintensity, 9 were found in the tibial one-third of the intra-articular portion of the graft (2 over the whole tibial region [Figure 8B], and 7 over the posterior aspect [Figure 8C]). In 1 case, the focal increase in hyperintensity was noted over the anterior aspect of the femoral one-third of the intra-articular portion of the graft. The focal hyperintensity over the anterior aspect of the graft was likely related to impingement of the graft by the intercondylar notch (Figure 8, A and B). The focal increase in signal intensity in the posterior aspect of the tibial one-third of the graft (Figure 8C) and the diffuse increase in hyperintensity of the graft (Figure 5D) were possibly signs representing ongoing ligamentization.

Subgroup Analysis

The mean SNQ of the whole graft measured on reassessment MRI in the subgroup (ie, 167 primary ACLRs operated with HS autograft) was 4.3 ± 3.9 (range, 0-22.6). Only 7 further graft ruptures were found at a mean follow-up of 56 months in this subgroup. Similar conclusions were reached in the subgroup analysis as in the whole cohort: (1) ligamentization of the ACLR grafts was completed within 36 months after the index operation; (2) higher SNQ on reassessment MRI was associated with increased chance of graft rupture; (3) hyperintense graft identified on reassessment MRI performed in the second year after surgery was associated with the risk of future graft rupture in subsequent follow-up.

The SNQs of the grafts were significantly higher when the reassessment MRI scans were obtained within the first 3 years compared with those obtained from the fourth postoperative year onward (*P* = .039; Mann-Whitney *U* test) (Appendix Figure A1, available online). A significant difference was found between the SNQ of the ACLR graft of the subgroup and that of the normal ACL (0.76 ± 0.67)[32] if the reassessment MRI scan was performed within the first 3 years after ACLR. The SNQ of the graft was comparable with the SNQ of normal

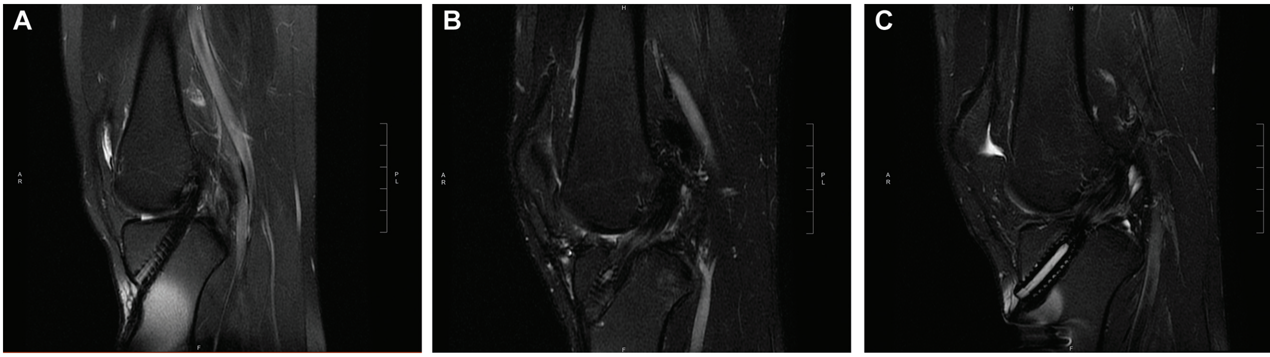

Figure 8. Region of focal increase in hyperintensity of the 10 grade 2B grafts that developed graft rupture in subsequent follow-up. (A) Focal increase in hyperintensity over the anterior aspect of femoral one-third of intra-articular portion of the graft (1 case). (B) Focal increase in hyperintensity over the tibial one-third of intra-articular portion of the graft (whole tibial insertion) (2 cases). (C) Focal increase in hyperintensity over the tibial one-third of the intra-articular portion of the graft (posterior aspect) (7 cases).

ACL only when the reassessment MRI scan was done ≥4 years after the index ACLR.

Will signal intensity of the intact graft on the reassessment MRI be able to predict the risk of future graft rupture? We found that the SNQ was significantly higher for the grafts that developed subsequent graft rupture compared with those that did not in the subgroup analysis (8.5 ± 6.4 vs 4.2 ± 3.7, respectively; P = .023, Mann-Whitney U test). The comparisons of patient and surgical characteristics between the ACLRs that developed graft rupture in subsequent follow-up (n = 7) and those that did not (n = 162) are shown in Appendix Table A4 (available online).

If the signal intensity of the graft is a predictor of future graft rupture, what is the best time to perform reassessment MRI to predict this? The subgroup analysis showed that modified Ahn grade of the intact graft when the first reassessment MRI was conducted in the second year after the index operation was significantly associated with the incidence of future graft rupture (P = .001; Fisher exact test). All the ACLRs that developed graft rupture at a mean follow-up of 56 months showed heterogeneous hyperintensity (ie, Ahn grade 2) when the reassessment MRI scan was performed in the second year after surgery. The relationship of graft maturity on reassessment MRI and subsequent rupture with reference to the timing of first-time reassessment MRI is presented in Appendix Table A5 (available online).

Interobserver and Intraobserver Repeatability

Concerning measurement of the mean SNQ of the intra-articular portion of the graft, both the interobserver repeatability and the intraobserver repeatability were excellent. The ICCs were 0.96 and 0.85, respectively.

Concerning qualitative assessment of the graft signal by modified Ahn classification, the kappa coefficients for interobserver and intraobserver agreement were 0.7 and 0.82, respectively. The interobserver agreement was considered to be substantial, and the intraobserver agreement was almost perfect.

DISCUSSION

In a cohort of 565 ACLRs with intact graft found on reassessment MRI, complete ligamentization of the graft was observed at 4 years after surgery, when the SNQ of the graft assessed by MRI was not different from that of the native ACL (Appendix Table A1, available online). If the normal range of the SNQ is defined as within 2 SDs from the mean SNQ of the native ACL, the proportion of ACLR grafts achieving an SNQ in the "normal" range increased with increasing time from the index surgery (Figure 7).

Significant differences were found between the SNQ of the grafts on MRI scans conducted within 3 years after surgery and those that were conducted >3 years after the operation (P < .001; Mann-Whitney U test). All of the ACLR grafts had an SNQ <2.6 when the reassessment MRI was performed ≥36 months after the index operation.

Higher SNQ of intact graft on reassessment MRI was significantly associated with increased incidence of graft rupture in subsequent follow-up (7.3 for subsequent graft rupture vs 4.4 for grafts without subsequent rupture; P = .004, Mann-Whitney U test). Graft hyperintensity measured by the modified Ahn classification was associated with future graft rupture when the reassessment MRI scan was conducted in the second year after ACLR (P < .001; Fisher exact test) (Table 4). The odds of rupture in a hyperintense graft (modified Ahn grade 2) was 12 times (95% CI = 2.8 to 52.6) that of a homogeneous hypointense graft (modified Ahn grade 1). The SNQ of an intact graft helped predict the chance of future graft rupture. For the purpose of predicting possible future graft ruptures, reassessment MRI should be performed in the second year after ACLR (ie, 12-24 months).

The data of the current study support the observation by Zhang et al[50] that the maturation of an ACLR graft was not completed until 24 months after ACLR. Our findings echo those of Muramatsu et al,[32] who reported that the SNQ of the graft became comparable with that of the native ACL after a period of 36 to 48 months from the index operation (Table 1). Li et al[25] observed a positive association between patient level of activity and SNQ in a cohort of 104 male patients receiving primary ACLR. The reassessment MRI in Li et al's[25] series was performed at a mean of 30.7 months after surgery, and the mean Tegner activity score of Li et al's[25] patients was 3.4 ± 0.8 at the time of MRI. We failed to identify a similar relationship between patient activity and SNQ. MRI was performed much earlier in the current cohort than in Li et al's series (18.3 months after surgery vs 30.7 months, respectively)[25], and our patients were much more active than Li et al's (Tegner activity score at the time of MRI, 5.1 ± 1.9 vs 3.4 ± 0.8, respectively).[25] However, we were not able to ascertain that these factors were the causes of the observed difference. Further study is required to clarify this.

In the current study, we found a statistically significant difference between the graft SNQ when the reassessment MRI was conducted within 36 months versus ≥4 years after surgery ($P < .001$; Mann-Whitney U test). It is reasonable to propose that the time of completion of ligamentization of the ACLR graft is 3 years (Figure 7).

A number of MRI techniques are available to study the signal intensity of the graft, namely SNQ, signal-noise ratio,[50] signal intensity ratio,[29] Positron emission tomography-MRI,[30] T1-weighted rho and T2-weighted mapping,[21] and 3-dimensional ultra–short echo T2-weighted MRI.[45] Among the various techniques, SNQ is the most extensively used technique[44] and has been correlated with histological and biomechanical findings.[46] Despite the presence of significant variations between the SNQ reported among different studies,[**] it remains a valuable research tool both for following patients in a single cohort and for comparing data with normal native ACL.

Although graft signal intensity is used extensively as a research tool to study the potential factors that may affect the rate of graft ligamentization, no studies of which we are aware have ascertained the association of SNQ with subsequent graft rupture in further follow-up. One of the most important findings in this study is that SNQ helped predict the chance of future graft rupture. The current study proposes a practical way to assess the risk of graft rupture in the postoperative period, which can be helpful in determining the optimal time of return to sports in selected patients. Patients with abnormally high SNQ found on the reassessment MRI conducted in the second year after surgery could be advised to refrain from pivoting sports until the graft ligamentization has been completed. Injection of a bioadjuvant agent (eg, platelet-rich plasma) is another potential option to hasten the progress of ligamentization.[4]

Limitations

The current study has a number of limitations. Although all of the clinical data were prospectively collected, the current research was a retrospective study, where selection bias, transfer bias, and assessment bias are expected to be more common. The adoption of well-defined exclusion criteria in the subgroup analysis helped reduce potential selection bias. The blinding of the observers during MRI assessment helped minimize possible assessment bias. This was reflected by the relatively high interobserver and intraobserver repeatability in the measurement of SNQ (ICCs were 0.96 and 0.85, respectively). However, due to the retrospective nature of the current study, the data were incomplete. For example, although smoking status was documented prospectively, there was no detailed information regarding whether the smokers had transiently stopped smoking at the time of operation and whether the nonsmokers started smoking in the postoperative period. Given that smoking is an important confounder of graft rupture after ACLR,[8] bias in this aspect cannot be ruled out.

Although SNQ is commonly used as a research tool to assess the effects of various surgical techniques on graft ligamentization after ACLR, there is large variation in the SNQ values reported by different studies.[††] The reported SNQ of HS autograft in the literature ranges from 1.7 to 22. It is not known whether the observed variations were related to the variable that was under study, were due to the presence of other confounders, or reflected the intrinsic limitation of this measurement method. In addition, possible differences in the scanning parameters of the MRI among different studies (eg, strength of magnet, field of view, slice thickness, voxel size, flip angle) may contribute to the observed variation. The lack of standard reporting of MRI scanning parameters (including those in the current study) makes comparison of SNQ among different studies difficult. Despite the significant variation between the SNQ reported among different studies, SNQ seems to be a valuable research tool in following patients of the same cohort and in comparing data with a normal value in native ACL. According to the equation of signal-noise-quotient, a low SNQ (<1) indicates that the signal intensity of the graft is comparable with that of a normal quadriceps tendon. This is a definite sign of complete ligamentization.[32]

The incidence of subsequent graft rupture increased with the time of follow-up. In the current study, 35% of subsequent graft ruptures occurred after a follow-up of ≥5 years after ACLR. The cohort of the current study consisted of patients with ACLRs performed between 2007 and 2019. Around 20% of the ACLRs were performed from 2017 onward. The maximum duration of possible follow-up in this group was <5 years at the time of the drafting of the article. Hence, some grafts that are "intact" at the present moment may develop graft rupture with longer follow-up.

The current cohort consisted of a heterogeneous sample of patients undergoing ACLR. The influence of potential confounders might not be completely controlled. However, it is not uncommon in our daily practice to encounter patients who have multiple risk factors for graft rupture (eg, a young elite athlete with grade 3 pivot-shift test scheduled for revision ACLR with allograft). Thus, the

**References 5-7, 10, 11, 13, 14, 22, 23, 26-28, 32, 34, 35, 38, 40, 43, 47-49.

††References 5-7, 10, 11, 13, 14, 22, 23, 26-28, 32, 34, 35, 38, 40, 43, 47-49.

current study sample provides a good representation of the usual patients that a sports surgeon may encounter in clinical practice. Graft signal intensity in terms of SNQ provides sports surgeons with a convenient method to assess the degree of graft maturation even in patients who have multiple risk factors for graft rupture. However, this study investigated graft ligamentization of only the intra-articular portion of the graft. The status of bone-tendon healing and the presence of postoperative tunnel widening were not studied. Only results concerning the intra-articular graft signal were reported. Information on intra-tunnel graft signal was not evaluated. In addition, no serial MRI scans were available to document the longitudinal changes of graft hyperintensity with time.

The results of the current study were reproduced in a subgroup analysis of primary ACLRs performed using HS autograft. However, similar subgroup analysis was not performed in the other 2 subgroups (BPTB autograft and quadriceps tendon autograft) because the sample sizes of these 2 subgroups were relatively small. It is possible that the timeline for ligamentization might be different for ACLRs performed using BPTB autograft or quadriceps tendon autograft with bone plug. Readers should be aware of this limitation when extrapolating the results of the current study to their clinical practice.

In the current study, patients with a hyperintense graft (ie, Ahn grade 2) identified on reassessment MRI performed in the second year after ACLR were at greater risk of subsequent graft rupture than those with a homogeneous hypointense graft (ie, Ahn grade 1). Similar statistical significance was not observed when reassessment MRI was conducted at other time points (Table 4); however, the samples for 1-year, 3-year, and 4-year reassessment MRI did not have enough power. It is not known whether the *P* values would be significant for the other periods if the sample size was larger. The number of graft reruptures in patients undergoing first-time reassessment MRI at $\geq$4 years was limited, adding fragility to the statistical analysis.

Most of the patients in the current cohort underwent only 1 set of postoperative MRI scans. Longitudinal data showing the progress of SNQ with time were not available. Thus, the method of the current study was not the most appropriate way to determine the time of completion of ligamentization. Only 6 MRI scans (ie, 1% of the whole cohort) were obtained $\geq$5 years after the index surgery. The current study could not provide good data on possible changes in graft signal in midterm follow-up, as in some studies in the literature.[50] However, due to the amount of resources required to perform repeated MRI scans, the sample size of studies involving multiple postoperative MRI scans is typically small, especially if the time of follow-up is beyond 2 years (Table 1). This may be the reason why the relationship between SNQ and subsequent graft rupture was not investigated in these studies. The large sample size of the current cohort (565 ACLRs) allowed us to study this important relationship in an adequately powered manner. Our research showed that increased SNQ was associated with subsequent graft rupture.

The interobserver repeatability and intraobserver repeatability were excellent in the SNQ measurement (ICC = 0.96 and 0.85, respectively). Concerning the Ahn classification, the interobserver agreement was substantial (kappa coefficient = 0.7) and the intraobserver agreement was almost perfect (kappa coefficient = 0.85). However, the second observer performed repeated measurement in only 100 sets of MRI scans. Although this is a substantial number of repeated measurements, there is still a chance that the reliability coefficients would change if the repeatability test had been completed in all 565 sets of MRI scans. Errors in SNQ measurement and Ahn classification can arise from selection of images for measurement and correct identification and outlining of the graft.

In the current study we had no detailed record of the activity level of patients in the postoperative period, other than the Tegner activity scale. Although the patients' activity was reported using the Tegner activity scale, detailed information about the frequency and intensity of activity (including rehabilitation and sports) was not available. This might be the reason why we did not observe a positive association between patient activity level and graft SNQ.

Despite demonstrating the association of high SNQ with subsequent graft rupture, the results of the current study may not change the clinical practice of orthopaedic surgeons. It is difficult to ask most patients to refrain from returning to preinjury sports on the basis of MRI graft hyperintensity alone. However, in selected groups of patients, there is a possibility of adjusting the frequency and intensity of training and competitive sports participation according to the results of reassessment MRI conducted in the second year after surgery.

CONCLUSION

In a cohort of 565 patients with ACLRs and a mean follow-up of 67 months, it was shown that ligamentization of ACLR grafts was likely completed within 36 months after the index operation. Higher SNQ on reassessment MRI was associated with an increased chance of graft rupture on subsequent follow-up. Patients with a hyperintense graft identified on reassessment MRI scans performed in the second year after surgery were at 12 times greater risk of subsequent graft rupture than those with a homogeneous hypointense graft.

REFERENCES

1. Ahn JH, Lee SH, Choi SH, Lim TK. Magnetic resonance imaging evaluation of anterior cruciate ligament reconstruction using quadrupled hamstring tendon autografts: comparison of remnant bundle preservation and standard technique. *Am J Sports Med*. 2010;38(9):1768-1777.
2. Amiel D, Frank C, Harwood F, Fronek J, Akeson W. Tendons and ligaments: a morphological and biochemical comparison. *J Orthop Res*. 1983;1(3):257-265.
3. Amiel D, Kleiner B, Roux RD, Harwood FL, Akeson WH. The phenomenon of ''ligamentization'': anterior cruciate ligament reconstruction with autogenous patellar tendon. *J Orthop Res*. 1986;4:162-172.
4. Beyzadeoglu T, Pehlivanoglu T, Yildirim K, Buldu H, Tandogan R, Tuzun U. Does the application of platelet-rich fibrin in anterior cruciate ligament reconstruction enhance graft healing and maturation? *Orthop J Sports Med*. 2020;8(2):2325967120902013.

5. Bouguennec N, Robinson J, Douiri A, Graveleau N, Colombet PD. Two-year postoperative MRI appearances of anterior cruciate ligament hamstrings autografts are not correlated with functional outcomes, anterior laxity, or patient age. *Bone Joint Open*. 2021;2(8):569-575.
6. Cavaignac E, Marot V, Faruch M, et al. Hamstring graft incorporation according to the length of the graft inside tunnels. *Am J Sports Med*. 2018;46(2):348-356.
7. Cavaignac E, Mesnier T, Marot V, et al. Effect of lateral extra-articular tenodesis on anterior cruciate ligament graft incorporation. *Orthop J Sports Med*. 2020;8(11):2325967120960097.
8. Chan YC, Yau WP. Association of smoking with graft rupture after anterior cruciate ligament reconstruction. *Orthop J Sports Med*. 2022;10(10):23259671221127244.
9. Cicchetti DV. Guidelines, criteria, and rules of thumb for evaluating normed and standardized assessment instruments in psychology. *Psychol Assess*. 1994;6(4):284-290.
10. Cusumano A, Capitani P, Messina C, et al. Different timing in allograft and autograft maturation after primary anterior cruciate ligament reconstruction does not influence the clinical outcome at mid-long-term follow-up. *Knee Surg Sports Traumatol Arthrosc*. 2022;30(7):2281-2290.
11. Fukuda H, Ogura T, Asai S, et al. Bone-patellar tendon-bone autograft maturation is superior to double-bundle hamstring tendon autograft maturation following anatomic anterior cruciate ligament reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2022;30(5):1661-1671.
12. Gifstad T, Foss OA, Engebretsen L, et al. Lower risk of revision with patellar tendon allografts compared with hamstring autografts: a registry study based on 45,998 primary ACL reconstructions in Scandinavia. *Am J Sports Med*. 2014;42(10):2319-2328.
13. Gong H, Huang B, Zheng Z, Fu L, Chen L. Clinical use of platelet-rich plasma to promote tendon-bone healing and graft maturation in anterior cruciate ligament reconstruction—a randomized controlled study. *Indian J Orthop*. 2022;56:805-811.
14. Grassi A, Casali M, Macchiarola L, et al. Hamstring grafts for anterior cruciate ligament reconstruction show better magnetic resonance features when tibial insertion is preserved. *Knee Surg Sports Traumatol Arthrosc*. 2021;29:507-518.
15. Howell SM, Clark JA, Blasier RD. Serial magnetic resonance imaging of hamstring anterior cruciate ligament autografts during the first year of implantation: a preliminary study. *Am J Sports Med*. 1991;19(1):42-47.
16. Howell SM, Knox KE, Farley TE, Taylor MA. Revascularization of a human anterior cruciate ligament graft during the first two years of implantation. *Am J Sports Med*. 1995;23(1):42-49.
17. Hui C, Salmon LJ, Kok A, Maeno S, Linklater J, Pinczewski LA. Fifteen-year outcome of endoscopic anterior cruciate ligament reconstruction with patellar tendon autograft for ''isolated'' anterior cruciate ligament tear. *Am J Sports Med*. 2011;39(1):89-98.
18. Kim SJ, Lee SK, Kim SH, Kim SH, Ryu SW, Jung M. Effect of cigarette smoking on the clinical outcomes of ACL reconstruction. *J Bone Joint Surg Am*. 2014;96(12):1007-1013.
19. Kondo E, Yasuda K, Katsura T, Hayashi R, Kotani Y, Tohyama H. Biomechanical and histological evaluations of the doubled semitendinous tendon autograft after anterior cruciate ligament reconstruction in sheep. *Am J Sports Med*. 2012;40(2):615-624.
20. Landis JR, Koch GG. The measurement of observer agreement for categorical data. *Biometrics*. 1977;33(1):159-174.
21. Lansdown DA, Xiao W, Zhang AL, et al. Quantitative imaging of anterior cruciate ligament (ACL) graft demonstrates longitudinal compositional changes and relationships with clinical outcomes at 2 years after ACL reconstruction. *J Orthop Res*. 2020;38(6):1289-1295.
22. Lee BI, Kim BM, Dho DH, Dwon SW, Kim HJ, Hwang HR. Does the tibial remnant of the anterior cruciate ligament promote ligamentization? *Knee*. 2016;23(6):1133-1142.
23. Lee DW, Kim JG, Lee JH, Park JH, Kim DH. Comparison of modified transtibial and outside-in techniques in anatomical single-bundle anterior cruciate ligament reconstruction. *Arthroscopy*. 2018;34(10):2857-2870.
24. Li H, Chen J, Li H, Wu Z, Chen S. MRI-based ACL graft maturity does not predict clinical and functional outcomes during the first year after ACL reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2017;25:3171-3178.
25. Li H, Chen S, Tao H, Li H, Chen S. Correlation analysis of potential factors influencing graft maturity after anterior cruciate ligament reconstruction. *Orthop J Sports Med*. 2014;2(10):2325967114553552.
26. Li H, Liu S, Sun Y, Li H, Chen S, Chen J. Influence of graft bending angle on graft maturation, the femoral tunnel, and functional outcomes by 12 months after anterior cruciate ligament reconstruction. *Orthop J Sports Med*. 2019;7(11):2325967119882663.
27. Li Q, Zhang Y, Zhan L, Han Q, Wu M, Zhang N. Correlation analysis of magnetic resonance imaging-based graft maturity and outcomes after anterior cruciate ligament reconstruction using International Knee Documentation Committee score. *Am J Phys Med Rehab*. 2019;98(5):387-391.
28. Liu S, Li H, Tao H, Sun Y, Chen S, Chen J. A randomized clinical trial to evaluate attached hamstring anterior cruciate ligament graft maturity with magnetic resonance imaging. *Am J Sports Med*. 2018;46(5):1143-1149.
29. Lutz PM, Achtnich A, Schutte V, Woertler K, Imhoff AB, Willinger L. Anterior cruciate ligament autograft maturation on sequential postoperative MRI is not correlated with clinical outcome and anterior knee stability. *Knee Surg Sports Traumatol Arthrosc*. 2022;30(10):3258-3267.
30. Magnussen RA, Binzel K, Zhang J, et al. ACL graft metabolic activity assessed by ^{18}FDG PET-MRI. *Knee*. 2017;24(4):792-797.
31. Magnussen RA, Meschbach NT, Kaeding CC, Wright RW, Spindler KP. ACL graft and contralateral ACL tear risk within ten years following reconstruction: a systematic review. *JBJS Rev*. 2015;3(1).
32. Muramatsu K, Hachiya Y, Izawa H. Serial evaluation of human anterior cruciate ligament grafts by contrast-enhanced magnetic resonance imaging: comparison of allografts and autografts. *Arthroscopy*. 2008;24(9):1038-1044.
33. Nathaniel AD, Barret JG, Award H, Bautista CA, Banes A, Butler DL. A brief history of tendon and ligament bioreactors: impact and future prospects. *J Orthop Res*. 2020;38(11):2318-2330.
34. Okutan AE, Kalkisim M, Gurun E, Ayas MS, Aynaci O. Tibial slope, remnant preservation, and graft size are the most important factors affecting graft healing after ACL reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2022;30(5):1584-1593.
35. Oshima T, Putnis S, Grasso S, Klasan A, Parker DA. Graft size and orientation within the femoral notch affect graft healing at 1 year after anterior cruciate ligament reconstruction. *Am J Sports Med*. 2020;48(1):99-108.
36. Pauzenberger L, Syre S, Schurz M. ''Ligamentization'' in hamstring tendon grafts after anterior cruciate ligament reconstruction: a systematic review of the literature and a glimpse into the future. *Arthroscopy*. 2013;29(10):1712-1721.
37. Pinczewski LA, Lyman J, Salmon LJ, Russell VJ, Roe J, Linklater J. A 10-year comparison of anterior cruciate ligament reconstructions with hamstring tendon and patellar tendon autograft: a controlled, prospective trial. *Am J Sports Med*. 2007;35(4):564-574.
38. Rojas G, Perelli S, Ibanez M, Formagnana M, Ormazabal I, Monllau C. Effect of modified Lemaire anterolateral extra-articular tenodesis on the magnetic resonance imaging maturity signal of anterior cruciate ligament hamstring graft. *Am J Sports Med*. 2021;49(9):2379-2386.
39. Runer A, Csapo R, Hepperger C, Herbort M, Hoser C, Fink C. Anterior cruciate ligament reconstructions with quadriceps tendon autograft result in lower graft rupture rates but similar patient-reported outcomes as compared with hamstring tendon autograft: a comparison of 875 patients. *Am J Sports Med*. 2020;48(9):2195-2204.
40. Saito M, Nakajima A, Sonobe M, et al. Superior graft maturation after anatomical double-bundle anterior cruciate ligament reconstruction using the transtibial drilling technique compared to the transportal technique. *Knee Surg Sports Traumatol Arthrosc*. 2019;27(8):2468-2477.
41. Salmon LJ, Health E, Akrawi H, Roe JP, Linklater J, Pinczewski LA. 20-year outcomes of anterior cruciate ligament reconstruction with

hamstring tendon autograft: the catastrophic effect of age and posterior tibial slope. *Am J Sports Med*. 2018;46(3):531-543.
42. Samuelsen BT, Webster KE, Johnson NR, Hewett TE, Krych AJ. Hamstring autograft versus patellar tendon autograft for ACL reconstruction: is there a difference in graft failure rate? A meta-analysis of 47,163 patients. *Clin Orthop Relat Res*. 2017;475:2459-2468.
43. Takahashi T, Kimura M, Hagiwara K, Ohsawa T, Takeshita K. The effect of remnant tissue preservation in anatomical double-bundle ACL reconstruction on knee stability and graft maturation. *J Knee Surg*. 2019;32(6):565-576.
44. Van Dyck P, Zazulia K, Smekens C, Heusdens CHW, Janssens T, Sijbers J. Assessment of anterior cruciate ligament maturity with conventional magnetic resonance imaging: a systematic literature review. *Orthop J Sports Med*. 2019;7(6):2325967119849012.
45. Warth RJ, Zandiyeh P, Rao M, et al. Quantitative assessment of in vivo human anterior cruciate ligament autograft remodeling: a 3-dimensional UTE-T2 imaging study. *Am J Sports Med*. 2020; 48(12):2939-2947.
46. Weiler A, Peters G, Maurer J, Unterhauser FN, Sudkamp NP. Biomechanical properties and vascularity of an anterior cruciate ligament graft can be predicted by contrast-enhanced magnetic resonance imaging: a two-year study in sheep. *Am J Sports Med*. 2001; 29(6):751-761.
47. Wen Z, Zhang H, Yan W, et al. Oval femoral tunnel technique is superior to the conventional round femoral tunnel technique using the hamstring tendon in anatomical anterior cruciate ligament reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2020;28(7):2245-2254.
48. Zhang J, Ma Y, Pang C, Wang H, Jiang Y, Ao Y. No differences in clinical outcomes and graft healing between anteromedial and central femoral tunnel placement after single bundle ACL reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2021;29(6):1734-1741.
49. Zhang S, Liu S, Yang L, Chen S, Chen S, Chen J. Morphological changes of the femoral tunnel and their correlation with hamstring tendon autograft maturation up to 2 years after anterior cruciate ligament reconstruction using cortical suspension. *Am J Sports Med*. 2020;48(3):554-564.
50. Zhang Y, Liu S, Chen Q, Hu Y, Sun Y, Chen J. Maturity progression of the entire anterior cruciate ligament graft of insertion-preserved hamstring tendons by 5 years: a prospective randomized controlled study based on magnetic resonance imaging evaluation. *Am J Sports Med*. 2020;48(12):2970-2977.