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Biomechanical comparisons of anterior cruciate ligament: reconstruction procedures with flexor tendon graft

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Abstract An experimental study was conducted to compare the biomechanical characteristics of six currently available anterior cruciate ligament (ACL) reconstruction procedures with flexor digitorum profundus tendons. Forty porcine knees were divided into eight groups, of 5 knees each. In groups A, B, C, and D, the flexor tendon graft was fixed with sutures and an Endobutton, with 20-mm-wide polyester tapes and staples, with 10-mm-wide polyester tapes and an Endobutton, and with bone plugs and interference screws, respectively. In group E, the graft was fixed using a combined procedure of those in groups B and D. In group F, the graft was directly fixed with interference screws. In groups G and H, the bone-patellar tendon-bone graft was fixed with interference screws, and sutures, respectively. Each femur-graft-tibia complex specimen was tested with a tensile tester by anteriorly translating the tibia until failure. This study demonstrated that the biomechanical properties of the femur-graft-tibia complex reconstructed with the flexor tendon graft were significantly affected by synthetic fixation devices. Regarding the average maximal load of the groups with the flexor tendon graft, group B had the highest (893 N) and group C had the second highest (770 N). Groups E and A were in the third rank. Group F had the second lowest (312 N), and Group D had the lowest (230 N). The maximal load of group B was significantly greater ($P < 0.01$) than that of group G (656 N) with the bone-patellar tendon-bone grafts. As to clinical relevance, this study indicated that the flexor tendon graft can be an alternative substitute for the bone-patellar tendon-bone graft for ACL reconstruction, if we understand the biomechanical characteristics of each reconstruction procedure.

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Key words Anterior cruciate ligament reconstruction · Flexor tendon graft · Biomechanics · Fixation strength

Introduction

Accelerated rehabilitation is usually recommended after anterior cruciate ligament (ACL) reconstruction.[17,21,24] Therefore, the tendon graft must be rigidly fixed to the femur and the tibia in ACL reconstruction. The bone-patellar tendon-bone graft has commonly been used as a substitute for ACL reconstruction, based on many biomechanical studies of initial fixation strength.[4,11,13] Recently, the flexor tendon has commonly been used as a substitute for ACL reconstruction because of its minimal graft site morbidity.[25] However, a number of studies have reported that the initial strength of the femur-flexor tendon graft-tibia complex was extremely low,[2,5,7,8,18,19] although Steiner et al.[22] reported that the doubled flexor tendon graft tethered to screw-posts with the multiple suture technique was comparable in initial fixation strength to the bone-patellar tendon-bone graft secured with interference screws. Currently, various fixation procedures with different synthetic devices have been developed for the flexor tendon graft: Yasuda et al.[24–26] fixed the doubled or tripled flexor tendons with polyester tapes and spiked staples; Rosenberg and Graf[20] developed the Endobutton technique for doubled flexor tendons; and Morgan[15] reported looped flexor tendons with bone plugs and interference screws. A few studies have been conducted to evaluate the biomechanical properties of each procedure.[12,22] However, no studies have simultaneously compared the biomechanical properties of the femur-graft-tibia complexes reconstructed with these various new procedures under the same mechanical conditions.

The purpose of this experimental study was to compare the biomechanical characteristics of currently available ACL reconstruction procedures using the flexor tendon graft in tensile testing.

Materials and methods

Study design

Porcine tendon and bone tissues harvested from fully mature LWD (Landrace, Large White, Duroc) pigs, weighing approximately 100kg, were used in this study to simulate ACL reconstruction. The porcine model had been established by determining the mechanical properties of the tendons in our previous biomechanical studies.[9,14] Forty fresh knees, 10 bone-patellar tendon-bone preparations, and 30 pairs of flexor digitorum profundus tendons harvested from the lower limbs of fully mature LWD pigs (body weight, approximately 100kg) were stored at −40°C until testing, and then thawed overnight at 4°C prior to the experiment. Two flexor tendons were sharply trimmed parallel to the fiber orientation so that their cross-sectional areas were 7mm^2 and 14mm^2, which corresponded to the average diameter of the human gracilis and semitendinosus tendons, respectively, according to the data reported by Noyes et al.[16] The knees were randomly divided into eight groups (groups A–H) of five knees each. For each group, the ACL was reconstructed, using one of eight different procedures (Fig. 1), after the ACL was resected. Femoral and tibial bone tunnels were drilled, using commercially available drill guide systems for human patients, through the anatomical insertions of the ACL. The diameters of the femoral and tibial drill holes were matched for the graft diameter in each knee. In six of the eight groups, the flexor tendon graft was fixed using six different procedures. In the two remaining groups, the ACL was reconstructed with a 10-mm-wide bone-patellar tendon-bone graft, which was fixed using two standard techniques (Fig. 1). These two groups were used to obtain control data from knees reconstructed with the "gold standard" techniques.

Pilot study

A pilot study was conducted to determine the mechanical properties of porcine tendons. We performed tensile tests using ten bone-patellar tendon-bone preparations and ten flexor digitorum profundus tendons harvested from fresh frozen porcine hind limbs. The specimens were thawed overnight at 4°C prior to testing. The length of the patellar tendon was approximately 50mm. For tensile tests of the bone-patellar tendon-bone

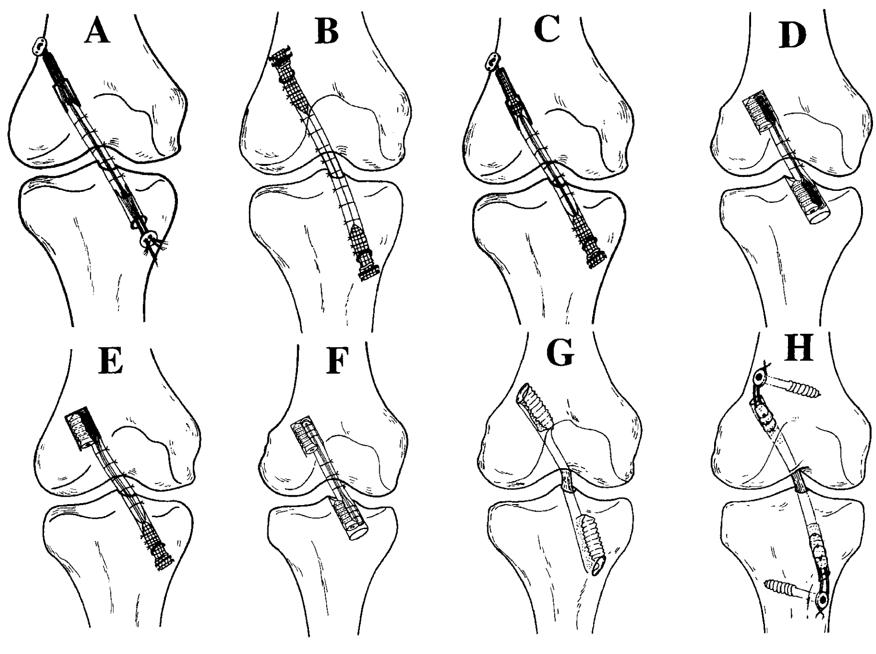

Fig. 1A–H. Anterior cruciate ligament (ACL) reconstruction procedures. **A** Doubled flexor tendons were fixed with sutures and buttons (an Endobutton at the femoral end). **B** Looped flexor tendons were fixed with 20-mm-wide polyester tapes and staples at each side. **C** Looped flexor tendons were fixed with 10-mm-wide polyester tapes and an Endobutton at the femoral side, and with 20-mm-wide polyester tapes and staples at the tibial side. **D** Doubled flexor tendons were fixed with bone plugs and interference screws at both sides. **E** Doubled flexor tendons were fixed with a bone plug and an interference screw at the femoral side, and with a 20-mm-wide polyester tape and staples at the femoral side. **F** Doubled flexor tendons were fixed with interference screws. **G** Bone-patellar tendon-bone graft was fixed with interference screws. **H** Bone-patellar tendon-bone graft was fixed with sutures and screws inserted into the bone preparation, the bone blocks were embedded in an aluminum pot with resin to firmly grip them. For tensile tests of the flexor tendons, we created a pair of specially designed cryo-grips[3] that could be attached to a tensile tester (Shimazu-Seisakusho, Tokyo, Japan). Inside the cryo-grip, there was a small space in which dry ice could be placed, so that each end of the flexor tendon was firmly gripped due to freezing. The length between the grips was set at 50 mm, which corresponded to the approximate length of the patellar tendon. The cross-sectional area of each tendon specimen was determined with an area micrometer,[23] similar to that reported by Butler et al.[5] The area micrometer consisted of an 8.0-mm-wide and 5.0-mm-thick rectangular slot attached to a micrometer and a plunger for insertion into the slot. After a tendon specimen was inserted into the micrometer slot, the plunger was placed on the tendon and pressed with a constant pressure of 0.12 MPa to fill the slot with tissue. The cross-sectional area of the tendon was calculated by multiplying the slot width by the measured thickness. After five cycles of 5% strain were loaded as preconditioning, tensile tests were performed at a cross-head speed of 50 mm/min. Two parallel lines were drawn transversely on the tendon surface with a stain (nigrosin) and were used as gauge-length markers for strain measurement. The distance between the two lines was approximately 30 mm. Elongation was determined with a video-dimension analyzer. Strain was calculated by dividing the tendon elongation by the initial distance between the two gauge-length markers. Stress was calculated by dividing the tensile load by the initial cross-sectional area of the tendon. The tangent modulus was defined by the linear slope of the stress-strain curve. Results of the pilot study are shown in Table 1. These data allowed us to conduct this biomechanical study using the porcine model.

Fixation procedures

In group A, a pair of the 7-mm^2 and 14-mm^2-thick flexor tendons was doubled, and three Tevdeck (Deknatel, Brooklyn, NY, USA) threads (number 2 thickness) were attached at both ends, using the baseball glove suture technique.[10] Drill holes in the femur and the tibia were created using a drill system for the arthroscopically assisted Endobutton (Acufex; Smith and Nephew Endoscopy, Andover, MA, USA) technique.[20] After the tendons were grafted across the knee joint through the tunnels, the femoral and tibial ends of the sutures were secured with an Endobutton, and with a conventional plastic button with a diameter of 14 mm (Acufex; Smith and Nephew Endoscopy), respectively.

In group B, a "hybrid" substitute for the two-incision procedure was created according to Yasuda et al.[24–26] Briefly, a pair of the flexor tendons was looped and both ends of each tendon were firmly sutured side by side, using three 2-0 polyester threads. Then, two 20-mm-wide meshed polyester tapes (Leeds-Keio Artificial Ligament; Neoligament Leeds, United Kingdom) were passed through the two loops of the flexor tendons. The chain-like junctions composed of the tendons and the tape were circumferentially ligated with three 2-0 polyester threads. Bone tunnels in the femur and the tibia were made using a conventional drill. The hybrid substitute was passed through the bone tunnels, so that the flexor tendon portion was located across the knee joint. Each end was then fixed to the bone with two spiked staples (Richards; Smith and Nephew Endoscopy).

In group C, the hybrid substitute for the one-incision procedure was created in the same manner as that used in group B. At the femoral end of the looped flexor tendons, however, a 10-mm-wide tape was attached instead of a 20-mm-wide tape. This thin tape was passed through holes of the Endobutton. A tibial drill hole was made in the same manner as that used in group B, but the femoral drill hole was created specially for the Endobutton technique, using the same technique as that used in group A. Thus, the flexor tendon graft was secured with a 10-mm-wide tape and an Endobutton at the femoral end, and with a 20-mm-wide tape and two spiked staples at the tibial end.

In group D, a bone-flexor tendon-bone graft was made and fixed in the bone tunnels according to Morgan.[15] Briefly, two cylindrical bone plugs, of 8-mm diameter and 20-mm length, were harvested from the femur and tibia with a hollow reamer. Three drill holes, of 1.5-mm diameter, were made in each bone plug so that they penetrated the plug. A pair of flexor tendons

Table 1. Mechanical properties of porcine tendons

Species	Tendon	n	Cross-sectional area (mm^2)	Tensile strength (MPa)	Tangent modulus (MPa)	Strain at failure (%)
Porcine	FDP	10	24.7 ± 0.9	94.9 ± 14.4	850.4 ± 34.3	11.3 ± 2.1
Porcine	BTB	10	33.9 ± 7.0	64.1 ± 11.1	332.4 ± 26.9	21.6 ± 3.6

Values are averages ± SD
FDP, Flexor digitorum profundus; BTB, bone-patellar tendon-bone preparation; n, number of specimens

was circumferentially wrapped around two bone plugs. The tendons were sutured and circumferentially ligated to each bone plug with 2-0 polyester threads. Thus, we obtained a quadruple stranded bone-flexor tendon-bone construct. Each bone plug was secured in the bone tunnel with an interference screw (Kurosaka screw, 9.0-mm diameter, 25-mm length; DePuy, Warsaw, IN, USA) according to the one-incision procedure.[15]

In group E, Morgan's technique, described above, was modified, using the hybrid technique described above. After a cylindrical bone plug was harvested from the femur, a pair of the flexor tendons was doubled and circumferentially wrapped around the bone plug, and sutured with the circumferential ligation technique. The free ends of the tendons were connected in series with a 20-mm-wide polyester tape, using the same technique as that used in group B. The bone plug was secured with a Kurosaka screw in the femoral tunnel according to the one-incision procedure,[15] and the polyester tape was fixed with two spike staples on the tibial cortex outside the tibial tunnel.

In group F, a pair of the flexor tendons was doubled, and each end of the tendons was sutured side by side, and circumferentially ligated with three 2-0 polyester threads. Each end of the tendon strand was fixed in a bone tunnel (9.0-mm diameter) with an interference screw (RCI screw, 8.0-mm diameter, 25-mm length; Smith and Nephew Endoscopy) inserted by the inside-out technique.

In group G, the ACL was reconstructed using the bone-patellar tendon-bone graft and a Kurosaka screw, according to the standard two-incision procedure. A 10-mm-wide bone-patellar tendon-bone graft was harvested with 25-mm-long bone blocks at both ends. The diameters of the femoral and tibial tunnels were both 10 mm. After the graft was placed through the tunnels, each end of the bone-patellar tendon-bone graft was secured with a Kurosaka screw.

In group H, the ACL was reconstructed with a bone-patellar tendon-bone graft and a suture-post technique for the two-incision procedure. We made three small drill holes in each bone block of the graft with a Kirschner wire 1.8-mm diameter. Three Tevdeck sutures, number 2 diameter, were passed through the holes. Femoral and tibial tunnels of 10-mm diameter were made. After the graft was placed through these tunnels, the sutures at both ends were firmly tethered to A–O cancellous screws (6.5-mm diameter, 25-mm length) (Symthes, Paoli, PA, USA) inserted into the femur and the tibia.

Biomechanical testing

The femur and the tibia were separately potted into steel cylinders, using polymethyl methacrylate resin.

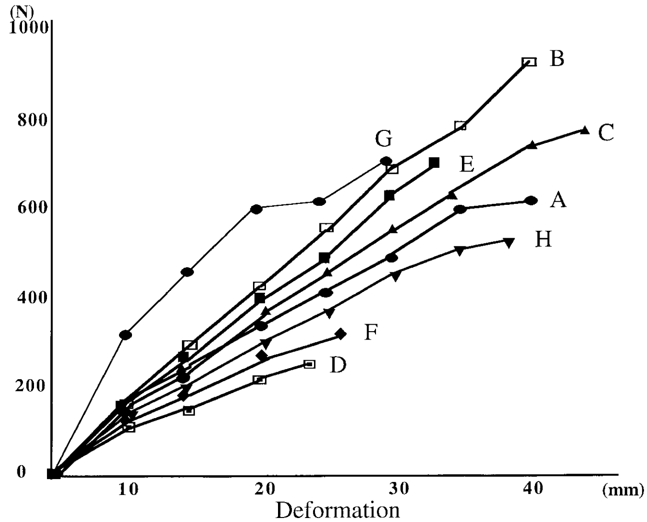

Fig. 2. Averaged load-deformation curves obtained from the eight types of femur-graft-tibia complexes

Soft tissues, except for the graft, were excised after the femur-graft-tibia complex was mounted onto a tensile tester with the specially designed grips at 30° of flexion, neutral rotation, and anatomical vertical alignment. Biomechanical tests were performed by anteriorly translating the tibia to the femur. Each specimen was kept moist with sprayed saline solution throughout the testing. After ten cycles of 50-N loads had been applied for preconditioning, an anterior drawer force was applied at a cross-head speed of 50 mm/min until graft failure. The applied force and the translation of the cross-head were recorded with an X-Y recorder (Yokogawa-Denki, Tokyo, Japan) to obtain a load-displacement curve (Fig. 2). From this curve, we obtained the maximal load, the stiffness, and the translation at failure. In addition, modes of failure in each specimen were recorded. The stiffness was defined as the slope of the linear region in the load-displacement curve.

Statistical analysis was performed in the eight groups for determining differences in each parameter, using one-way analysis of variation (ANOVA) with the Bonferroni/Dunn test for multiple comparisons. Significance level was set at $P < 0.01$.

Results

The average load-deformation curves obtained from the eight types of femur-graft-tibia complexes are shown in Fig. 2. Failure modes are shown in Table 2.

Regarding the average maximal load of the groups with the flexor tendon graft, group B had the highest and group C had the second highest. Groups E and A were in the third rank. Group F had the second lowest, and group D had the lowest (Fig. 3). Statistical analyses (Table 3) showed that the maximal load of group B was significantly greater ($P < 0.01$) than not only that of the other groups, except for group C, but also that of group G with the bone-patellar tendon-bone graft and interference screws. The maximal load of group C was significantly greater ($P < 0.01$) not only than that of groups F and D but also than that of group H with the bone-patellar tendon-bone graft and sutures. There were no significant differences among groups A, C, E, and G. The maximal load of groups D and F was significantly less ($P < 0.01$) than that of the other groups.

Concerning the average stiffness of the groups with the flexor tendon graft, group B had the highest and group E had the second highest. Groups C and A were in the third rank. Groups F and D were in the lowest rank (Fig. 4). Statistical analyses (Table 4) showed that

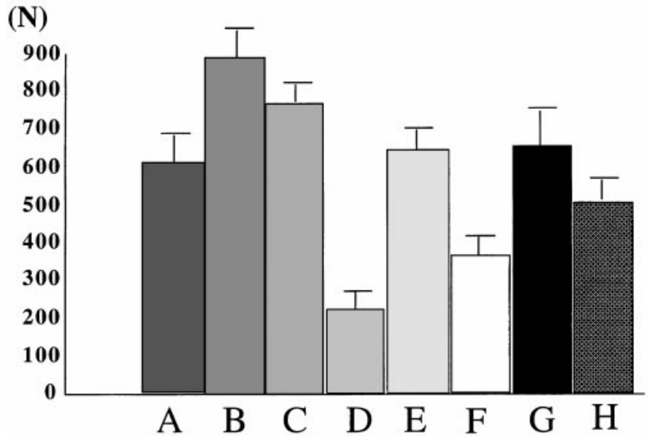

Fig. 3. Maximal load of femur-graft-tibia complexes reconstructed by each procedure. Statistical comparisons between two groups are shown below the bar graph

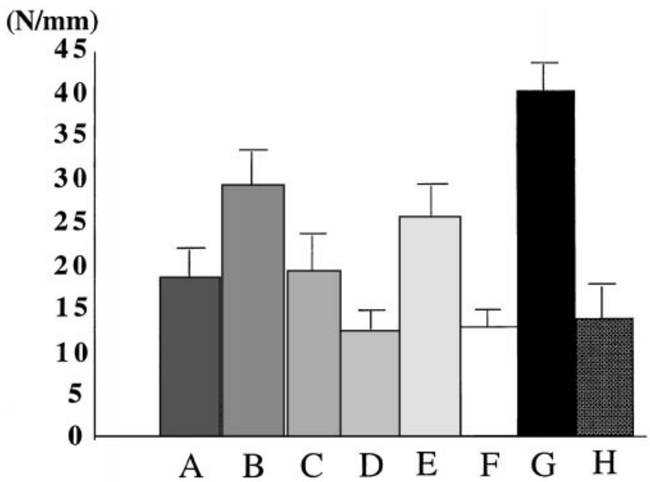

Fig. 4. Stiffness of femur-graft-tibia complexes reconstructed by each procedure. Statistical comparisons between two groups are shown below the bar graph

Table 2. Failure modes in the tensile tests

Group[a]	Mode of failure	Number of knees
A	Thread-tendon junction was disrupted	3
	Threads were torn	2
B	Tendon-tape junction failed	3
	Tape slipped out from the staples	2
C	Endobutton was pulled out	3
	Tendon-tape junction failed	2
D	Suture-tendon junction failed	5
E	Tendon-tape junction failed	3
	Tendons were torn by the screw or bone edge	2
F	Slippage of the tendons at the femoral side	3
	Slippage of the tendons at the tibial side	2
G	Bone-tendon junction was disrupted	3
	Bone plug slippage	2
H	Fracture of the bone plug	5

[a] See text for details of groups A–H

Table 3. Statistical comparisons of maximum load (N)

Group	A 608 ± 118	B 893 ± 100	C 770 ± 87	D 230 ± 26	E 647 ± 74	F 312 ± 43	G 656 ± 96	H 508 ± 130
A	—	$P < 0.0001$	$P = 0.0012$	$P < 0.0001$	NS	$P < 0.0001$	NS	NS
B	$P < 0.0001$	—	NS	$P < 0.0001$	$P = 0.0005$	$P < 0.0001$	$P = 0.0004$	$P < 0.0001$
C	$P = 0.0012$	NS	—	$P < 0.0001$	NS	$P < 0.0001$	NS	$P < 0.0001$
D	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	—	$P < 0.0001$	NS	$P < 0.0001$	$P < 0.0001$
E	NS	$P = 0.0005$	NS	$P < 0.0001$	—	$P < 0.0001$	NS	NS
F	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	NS	$P < 0.0001$	—	$P < 0.0001$	$P < 0.0001$
G	NS	$P = 0.0004$	NS	$P < 0.0001$	NS	$P < 0.0001$	—	$P = 0.0022$
H	NS	$P < 0.0001$	$P < 0.0001$	$p < 0.0001$	NS	$P < 0.0001$	$P = 0.0022$	—

Values are means ± SD. Each P value shows significance level of differences between the two groups
NS, Not significant

Table 4. Statistical comparisons of stiffness (N/mm)

Group	A 18.0 ± 3.8	B 29.4 ± 4.1	C 19.5 ± 3.5	D 12.5 ± 1.6	E 25.7 ± 2.5	F 12.8 ± 0.7	G 40.3 ± 7.1	H 13.8 ± 2.5
A	—	$P < 0.0001$	NS	NS	$P = 0.0050$	NS	$P < 0.0001$	NS
B	$P < 0.0001$	—	$P = 0.0003$	$P < 0.0001$	NS	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
C	NS	$P = 0.0003$	—	NS	NS	NS	$P < 0.0001$	NS
D	NS	$P < 0.0001$	NS	—	$P < 0.0001$	NS	$P < 0.0001$	NS
E	$P = 0.0050$	NS	NS	$P < 0.0001$	—	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
F	NS	$P < 0.0001$	NS	NS	$P < 0.0001$	—	$P < 0.0001$	NS
G	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	—	$P < 0.0001$
H	NS	$P < 0.0001$	NS	NS	$P < 0.0001$	NS	$P < 0.0001$	—

Values are means ± SD. Each *P* value shows the significance level of differences between the two groups
NS, Not significant

the stiffness of each group with the flexor tendon graft was significantly lower ($P < 0.01$) than that of group G with the bone-patellar tendon-bone graft and screws, although that of groups B and E was significantly higher ($P < 0.01$) not only than that of the other groups with the flexor tendon graft but also than that of group H with the bone-patellar tendon-bone graft and sutures. There were no significant differences among groups C, A, F, and D with the flexor tendon graft, and group H with the bone-patellar tendon-bone graft.

Discussion

This study demonstrated that the biomechanical properties of the femur-graft-tibia complex reconstructed with the flexor tendon graft were significantly affected by the synthetic devices used to fix the autograft. We used a porcine model in this study. Because the biomechanical properties of human specimens which are actually available are extremely varied, due to a wide array of factors,[1,27] there is a distinct possibility that, statistically, we may have false-negative results for significance in comparisons of biomechanical properties among the various fixation techniques. On the other hand, the biomechanical properties of porcine specimens are relatively uniform, as shown in the pilot measurements in this study. This is an advantage of the porcine model when the intention is to compare ligament reconstruction procedures. This was the reason why we used the porcine model in this study.

Regarding the maximal load, interestingly, group B, in which each end of the flexor tendon graft was fixed with a 20-mm-wide polyester tape, had a significantly higher value than group G, with the bone-patellar tendon-bone graft and interference screws. It has commonly been believed that the initial fixation strength of the flexor tendon graft is inferior to that of the bone-patellar tendon-bone graft.[2,5,7,8,18,19] However, this study has indicated that it is rather easier to strongly fix the flexor tendons to the bone, if we apply artificial materials such as meshed polyester tapes for fixation. The maximal load of group C was between that of groups B and H. This finding showed that the use of a 10-mm-wide tape and an Endobutton, instead of a 20-mm-wide tape and staples, respectively, slightly, but not significantly, reduced the fixation strength of the flexor tendon graft. This result supported the clinical utility of the 10-mm-wide tape and the Endobutton. In addition, the maximal load of group A, in which the flexor tendons were fixed with sutures and buttons, was comparable to that of group G, with the bone-patellar tendon-bone graft. This result was similar to the results of the study of Steiner et al.,[22] using human cadaveric knees.

The failure modes showed that the structurally weakest site in the femur-graft-tibia complex differed greatly among the procedures, depending on the artificial fixation materials. The weakest site is located at the tendon-artificial material interface for the procedures with the flexor tendon graft, while it is located at the tendon-bone junction or the bone plug for the procedures with the bone-patellar tendon-bone graft. It is necessary to know the weakest site in each complex in order to improve the reconstruction procedure. For example, the maximal load of group D, in which the flexor tendon graft was fixed with bone plugs and interference screws, was significantly lower than that of the other groups. The weakest point in the flexor tendon graft was located at the side-by-side suture site between the tendons. Therefore, the improvement of this weak point in group D resulted in better biomechanical properties of the femur-graft-tibia complex in group E.

Regarding the stiffness, all the groups with the flexor tendon graft had significantly lower values than group G, with the bone-patellar tendon-bone graft and screws. However, group H, in which the ACL was reconstructed with the bone-patellar tendon-bone graft and sutures, had values as low as that in group A, with the flexor tendons and sutures. The decrease in stiffness is considered to be one of the disadvantages of the multiple suture technique. This study showed that the use of a 20-mm-wide polyester tape instead of threads can significantly improve the stiffness, although it cannot completely compensate for this disadvantage. This improvement may be explained by changes in the biomechanical properties of the tape-tendon junction. On the other hand, the use of a 10-mm-wide tape and an Endobutton did not increase the stiffness of the femur-graft-tibia complex, compared with the use of sutures. This may be a weak point of the procedure with a 10-mm-wide tape and an Endobutton. Although the total length of the flexor tendon graft was reduced by using interference screws in groups D and F, the stiffness in these groups was significantly lower than that in group G, with the bone-patellar tendon-bone graft and interference screws. The occurrence of this phenomenon may be explained by slippage of the tendons at the side-by-side suture site in group D and by slippage at the tendon-metal interface in group F. However, the slippage in group D was reduced, by application of the 20-mm-wide polyester tape, in group E.

A weak point of this study was that we could not measure the mechanical properties of the porcine bones. However, the maximal failure load in our group G, with the bone-patellar tendon-bone graft and interference screws, was similar to the results in previous studies with human specimens (476N according to Kurosaka et al.[11] and 396–674N according to Steiner et al.[22]). Therefore, we believe that the porcine model is acceptable for biomechanical comparisons of ACL reconstruction procedures. However, the main limitation of this study is that the mechanical properties of porcine specimens are not precisely the same as those of human specimens.[5,6,9,14] Therefore, we must recognize that the absolute values obtained in this study cannot be extrapolated to ACL reconstruction procedures in human patients. Another limitation is that this study is an in-vitro study. Therefore, we should recognize that this study has dealt with only the initial biomechanical properties after surgery.

As to clinical relevance, first, the flexor tendon graft can be an alternative substitute for the bone-patellar tendon-bone graft for ACL reconstruction, if we understand the biomechanical characteristics of each reconstruction procedure. Specifically, we believe that the hybrid substitute is useful for cruciate ligament reconstruction, because we can achieve a wide safety range in the strength of the femur-graft-tibia complex against high loads accidentally applied in the early phase after surgery, and because we can easily and firmly fix the substitute onto the cortex of the bone with staples, applying a quantitative load.[25] Secondly, the initial strength of the tape-Endobutton procedure with the flexor tendon graft may be comparable to that of the bone-patellar tendon-bone graft procedure. This finding is useful when we apply postoperative rehabilitation for patients who have undergone surgery with this procedure. Thirdly, because the biomechanical properties of the flexor tendon graft fixed with interference screws were relatively inferior to the procedures which were used polyester tapes, whether or not bone plugs were used, postoperative management should be conducted carefully for knees reconstructed with flexor tendons and interterence screws.

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