

A biomechanical comparison of 4-strand and 5-strand anterior cruciate ligament graft constructs

Matthew L. Broadhead,^{1,2}
Animesh A. Singla,¹ Nicky Bertollo,^{1,3}
David Broe,^{1,3} William R. Walsh^{1,3}

¹University of New South Wales, Kensington; ²University of Newcastle, Newcastle; ³Surgical and Orthopaedic Research Laboratories, Prince of Wales Hospital, Randwick, Australia

Abstract

Hamstring tendon autografts are used for reconstruction of the anterior cruciate ligament. This study tested the hypothesis that a 5-strand hamstring autograft construct is superior in strength to a 4-strand construct. Four-strand and 5-strand tendon grafts constructs were prepared from ovine flexor tendons and then tested in a uniaxial electromechanical load system with suspensory fixation. The 4-strand and 5-strand constructs were pre-conditioned, stress-relaxed and loaded to ultimate failure. Stress-relaxation, stiffness and ultimate load were compared using a one-way ANOVA. There were no statistical differences in stress-relaxation, initial stiffness, secondary stiffness or ultimate load between 4-strand and 5-strand split tendon graft constructs. Inconsistent failure patterns for both 4-strand and 5-strand constructs were observed. The additional strand in the 5-strand construct may be shielded from stress with additional weakness secondary to the use of suspensory fixation. The potential biological benefit of religamentization and bony integration, with more autologous tissue in the intra-articular space and bony tunnels remains unknown.

Introduction

The anterior cruciate ligament (ACL) is one of several structures of the knee that provide stability during knee kinematics. It is important in preventing excessive anterior translation of tibia and internal rotation of tibia on the femur. Rupture of the ACL occurs most commonly during sporting activities with the most common mechanism being a non-contact pivoting injury. Chronic ACL deficiency may lead to recur-

rent episodes of knee instability with resultant chondral injuries and unreparable meniscal tears. For this reason, many young and more active patients choose to undergo ACL reconstruction. Between 2003-2008, 50,187 ACL reconstructions were performed in Australia. During this period 52 per 100,000 person years was the population-based incidence of ACL reconstruction. The time-estimated cost associated with ACL reconstruction surgery was over AUS\$ 75 million per year.¹

There are several different surgical techniques described for reconstruction of a torn ACL. Clinical trials, however, have been unable to determine the superiority of a single technique with significant heterogeneity in recommendations regarding the ideal construct. Keyhole, or arthroscopic-assisted surgery is widely regarded as the gold standard, and reconstruction most commonly involves harvesting an autograft from the patient and using it to reconstruct the native ACL anatomy. Options for graft selection include hamstring autograft, bone patella bone autograft, quadriceps tendon autograft and allograft. While there are various perceived advantages and disadvantages for the various graft options, choice of graft is largely influenced by surgical experience, surgeons' preference, and patients' circumstances. At the host institution for this study, primary reconstruction of the ACL is performed using either a 4-strand or 5-strand hamstring autograft technique. Briefly, this technique involves harvesting the semitendinosus and gracilis tendons from the patient and formation of a graft consisting of either four or five strands. The length and size of the tendons obtained from the patient largely determines whether a 4-strand or 5-strand construct is possible. There is a paucity of data comparing the clinical outcomes of 4-strand and 5-strand hamstring autograft techniques.

This study was conducted to test the hypothesis that 5-strand hamstring autograft constructs are superior in strength to 4-strand hamstring autograft constructs. This hypothesis follows from the finding that the strength of an ACL reconstruction is linearly proportional to the cross-sectional area of the graft.²⁻⁴ The primary aim of this study was to make a comparison of the biomechanical properties of 4-strand and 5-strand graft constructs, particularly evaluating stress-relaxation, stiffness and ultimate load. To make this comparison an *in vitro* model was used.

The *in vitro* ovine split tendon graft model described aims to replicate the human reconstructive suspensory technique using autologous hamstring tendon graft.

Correspondence: Matthew L. Broadhead, University of New South Wales, Kensington, High St, Kensington, Australia.
E-mail: matthewbroadhead@me.com
Tel: +61.411093758.

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Materials and Methods

Animals and harvesting of tendon grafts

An *in vitro* model using ovine flexor tendon split grafts was utilized to test the hypothesis that a 5-strand hamstring autograft constructs have superior strength to 4-strand constructs. This model was previously described by Hunt *et al.*⁵ The model has since been validated and extended to the *in vivo* setting for the evaluation of ACL endoligamentous and tunnel remodeling.⁶⁻⁹

The fresh frozen hind limbs of skeletal-mature sheep were obtained from a local supplier. All tissue was obtained and handled in accordance with animal rights protection laws. A longitudinal incision was made along the posterior aspect of the limb and the skin and fat was removed to display muscles and tendons. The Achilles tendon is formed by the superficial flexor tendon surrounded by the gastrocnemius tendon in sheep. The semitendinosus tendon in sheep is thin and transparent and not suitable for use as a graft.⁵ The tendon fascia was carefully dissected from the surface of the tendons. The superficial flexor digitorum and gastrocnemius tendons were separated and divided proximally and distally from insertion sites. Tendons were observed for defects and measured for length and width.

Small tendons with defects were discarded from the study.

Tendons were then fixed with clamps at either end and marked with a pen along the center of the tendon at 1cm intervals. The tendons were split along this line with a scalpel producing two parts for use as graft. Hence four flexor tendon grafts were harvested from each hind limb. The length and width of the split flexor tendon grafts were measured with digital calipers and recorded to ensure reproducibility in the model.

Graft preparation

Split flexor tendon grafts were marked at 3cm from each end prior to whip-stitching with #2 ethibond excel polyester suture (Ethicon, Somerville, NJ, USA). Stitches were placed 0.5 cm apart with a total of 4 passes of the suture up and down, with moderate tension between passes. Excess tendon was trimmed from the ends of the grafts. A mayo needle was used to pass the ethibond suture from one end of long graft through the middle of a 5 mm mersilene tape (Ethicon). This was then secured with 5 surgical square knots. All steps of graft harvesting and preparation were performed with careful consideration for reproducibility. The 5-strand construct was made by folding the superficial flexor tendon grafts back and forth three times. Graft obtained from the gastrocnemius tendon was folded in half. Hence the 5-strand graft consisted of three passes of superficial flexor tendon and two passes of gastrocnemius tendon. This was possible due to the consistently longer grafts obtained from the superficial flexor tendon (Figure 1). The 4-strand construct was made by folding both the superficial flexor and gastrocnemius grafts in half. Hence each tendon produced two passes in the graft (Figure 1). Suspensory fixation (EndoButton CL Ultra; Smith and Nephew, Andover, MA, USA) was used to test both 4-strand and 5-strand constructs (each n=6).

Grafting testing

4-strand and 5-strand constructs were tested using a uniaxial electromechanical load system (MTS, Eden Prairie, MN, USA). Constructs were fixed to the load system with cryogrips and pre-conditioned. A constant gauge length of 45 mm was used. Care was taken to pretension and precondition the individual strands of the construct (10 to 100 N for 50 cycles). Following stress relaxation for 90 seconds, the graft constructs were then tested to failure (20 mm.min⁻¹). Stress-relaxation, stiffness and ultimate load were recorded. Cyclic creep was not reported as this phenomenon was potentially masked by the preparation (Figure 2).

Statistical analysis

All statistical analysis was performed with SPSS software (IBM). Stress-relaxation, stiffness and ultimate load of the constructs were compared using a one-way ANOVA.

Results

Graft characteristics

The length and width of the harvested ovine superficial flexor and gastrocnemius tendons were measured using digital calipers after longitudinal splitting. This allowed for an equal distribution of graft material between 4-strand and 5-strand grafts constructs. The mean length and width of the 5-strand split tendon graft components were 215 mm (+/- 49.45 SD) and 5.33 mm (+/- 0.78 mm), respectively. The

mean length and width of the 4-strand split tendon graft components were 172.17 mm (+/-26.53) and 5.17 mm (+/- 0.72 SD), respectively. A greater length of split tendon graft component is necessary for formation of a 5-strand construct as the graft is tripled back on itself, rather than doubled back as for the 4-strand construct.

Stress-relaxation

Stress is defined as the force applied to the test construct per unit of area applied. Strain is the change in length of a material with respect to the material's original length. Stress relaxation is an observed decrease in stress while the test construct is held at constant strain. The mean stress relaxation was measured using the uniaxial electromechanical load system (MTS) with stress relaxation for 90 seconds. There was no observed difference in mean stress-relaxation force between the 4- and 5-strand constructs (P=0.59) (Figure 3A).

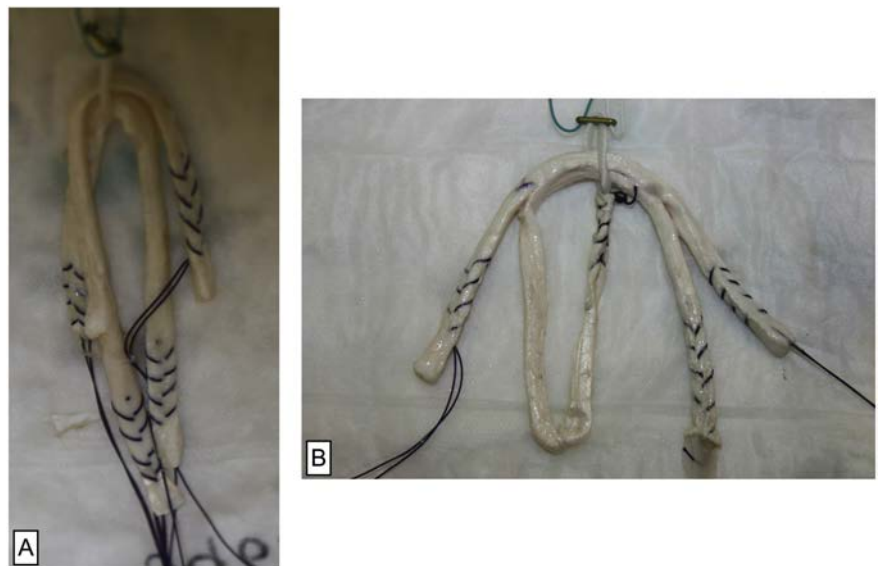


Figure 1. A) 4-strand and B) 5-strand split tendon graft constructs prior to testing.

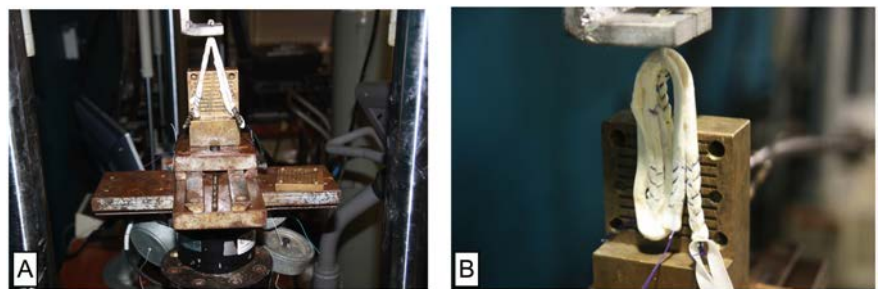


Figure 2. Testing a 4-strand (A) and 5-strand (B) construct in the uniaxial loading machine.

Ultimate failure load

Four-strand and 5-strand constructs were all tested to ultimate failure and were examined macroscopically after failure. Inconsistent ultimate failure patterns were observed. Some constructs failed at the EndoButton fixation, while others resulted in graft rupture, while still others appeared to fail secondary to a combined graft and fixation weakness. Half of all grafts tested failed at the EndoButton fixation, irrespective of whether the test construct was 4- or 5-stranded. Mean failure load was measured with the uniaxial electromechanical load system (MTS). There was no statistical difference in mean failure load between the 4-strand and 5-strand split tendon graft constructs ($P=0.46$) (Figure 3B).

Energy

The mean energy applied to the 4-strand and 5-strand constructs in joules prior to ultimate failure was measured using the uniaxial electromechanical load system (MTS). There was no statistical difference in the energy absorbed between the 4-strand and 5-strand constructs (Figure 3C).

Stiffness

Stiffness is the deflection of the tested construct while subjected to a given load. The steeper the gradient of the stress-strain curve, the stiffer the construct. Stiffness was measured using the uniaxial electromechanical load system (MTS). Both 4-strand and 5-strand split tendon graft constructs were found to have a bimodal pattern of stiffness (initial and secondary stiffness) (Figure 4). There was no significant difference between the initial ($P=0.30$) or secondary stiffness ($P=0.80$) between the two constructs (Figure 5).

Discussion

This study provides a biomechanical comparison of 4-strand and 5-strand split tendon graft constructs harvested from fresh frozen sheep hind limbs. The model has been used to replicate human ACL reconstruction techniques that utilize 4-strand and 5-strand hamstring tendon autografts. Hunt *et al.*⁵ have previously described this

model that has been validated and used for ovine *in vivo* studies of ACL reconstruction. The biomechanical comparison presented here measured stress relaxation, ultimate failure load and stiffness for the 4-strand and 5-strand constructs. Results across all parameters were unable to show superiority of the 5-strand construct, with inconsistent modes to failure evident. The lack of additional benefit when a fifth strand is added to the graft construct is surprising given that a linear correlation between cross-sectional area and ultimate failure load and stiffness has clearly been shown.¹⁰ Closer consideration of the 5-strand construct may explain these findings. The 5-strand construct is prepared with a strand that is tied to the EndoButton before being passed through the EndoButton loop to create a total of three strands. The forces acting on this construct may not be shared equally through all three strands, leading to stress shielding. There is a paucity of data describing the *in vitro* testing of 4-strand and 5-strand ACL graft constructs. Two clinical studies have examined the potential benefit of additional strands in hamstring tendon autograft con-

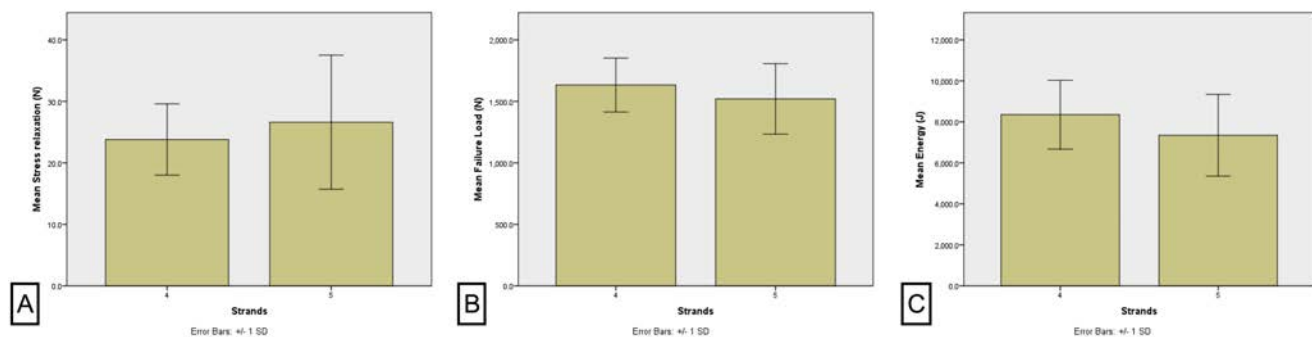


Figure 3. A) Stress relaxation for 4-strand and 5-strand constructs; B) ultimate failure load of for 4-strand and 5-strand constructs; C) energy absorbed prior to failure for 4-strand and 5-strand constructs.

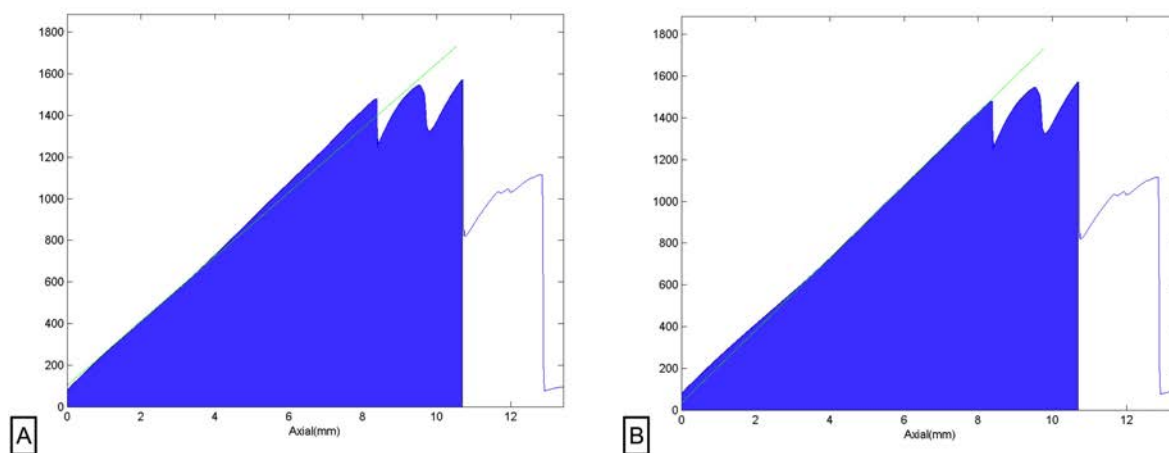


Figure 4. Stress-strain curve for 4-strand (A) and 5-strand (B) constructs.

structs. Prodomos *et al.*¹¹ compared 4- vs 5-strand ACL reconstructions in 40 patients using hamstring tendons. The 5-strand graft demonstrated increased stability as measured with a KT-1000 scores. The authors suggested that the increased stability offered with the 5-strand constructs would be of benefit to high risk patients. Zhao *et al.*¹² compared 4-strand and 8-strand hamstring tendon autografts. KT-1000 evaluation and clinical measures supported a double bundle ACL reconstruction with 8-strand vs 4-strand. Additionally, subjective measures, the IKDC and Lysholm score, were also superior for the 8-strand group.

Most published literature compares the biomechanical properties of the native ACL to bone-patella tendon-bone and double bundle hamstring autograft constructs. In these studies, the ultimate failure load ranges from 2400-4600 N, while the stiffness for double bundle constructs range from 200 to 900 N/mm.^{10,13-16} There is no published biomechanical data on 5-strand constructs, however the ultimate failure load in the current study was significantly lower (approximately 1500 N) than these published results. The suspensory technique of fixation (EndoButton) used may have contributed to the lower ultimate failure loads demonstrated in this study when compared to those values demonstrated for bone-patella tendon-bone and double bundle hamstring autograft constructs. Inconsistent patterns of failure were observed in this study. Half or all grafts tested failed at the EndoButton fixation, irrespective of whether the test construct was 4- or 5-stranded. *In vivo* results of these constructs may differ significantly from these results, especially after some time is allowed for ACL endoligamentous and tunnel remodeling with bony integration. Importantly, once the construct is fully integrated into the bony tunnel the weakness of the EndoButton, as well as the stress shielding of the additional strand already described, may no longer be problematic. Functionally the force would be shared

across all five strands of the construct with potential benefits in ultimate load.

The potential biological benefit of bony integration of a 5-strand hamstring tendon autograft construct remains unknown. The effect of having additional autologous graft both with the knee and bony tunnels represents an avenue for future investigation. This is significant when one considers the effect of graft-tunnel disparity. Graft-tunnel disparity is the space between the inserted graft material and the bony tunnel walls. When suspensory fixation is used with an undersized graft there is no compressive effect on the intraosseous section of the graft. At a cellular level, better graft-tunnel fit is believed to lead to early cross-linking of collagen fibers between the graft tendon and cancellous bone in the tunnel^{3,17} compared gap disparity of 0, 0.3 and 0.5 mm using an *in vivo* rabbit model. Reduced gap disparity was associated with denser and more organized healing tissue. Maximal tensile strength was also increased by reducing gap disparity. Yamazaki *et al.*¹⁸ showed that graft-tunnel disparity up to 2mm did not affect intraosseous healing of a flexor tendon graft using a canine model.

Conclusions

This study has provided a biomechanical comparison of 4-strand and 5-strand split tendon graft constructs using suspensory fixation. The results presented for stress relaxation, ultimate failure load and stiffness did not show a benefit in the use of a 5-strand construct. In fact, the biomechanical studies presented here suggest that, in its current form, the additional strand in the 5-strand construct may be shielded from stress with additional weakness secondary to the use of suspensory fixation. The potential biological benefit of religamentization and bony integration, with more autologous tissue in the intra-articular space and bony tunnels remains unknown. A similar study using a modified construct and an

alternative fixation device, such as an interference screw, is warranted. Although the results did not show a biomechanical difference between the 4- and 5-strand constructs, reduced graft tunnel disparity and the associated increase in pull-out strength, may still provide additional benefit to a modified 5-strand construct.

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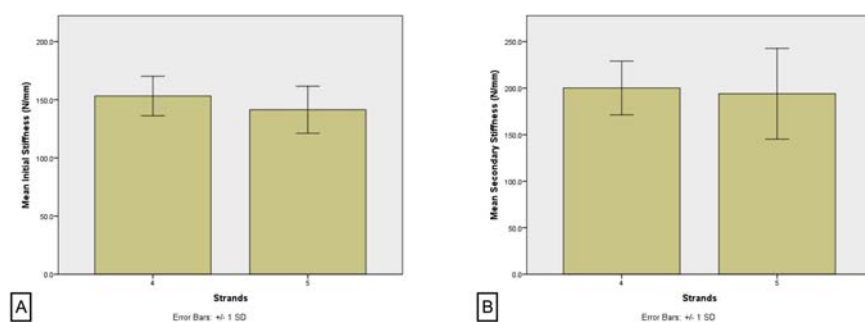


Figure 5. Initial (A) and secondary (B) stiffness of 4-strand and 5-strand constructs.

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