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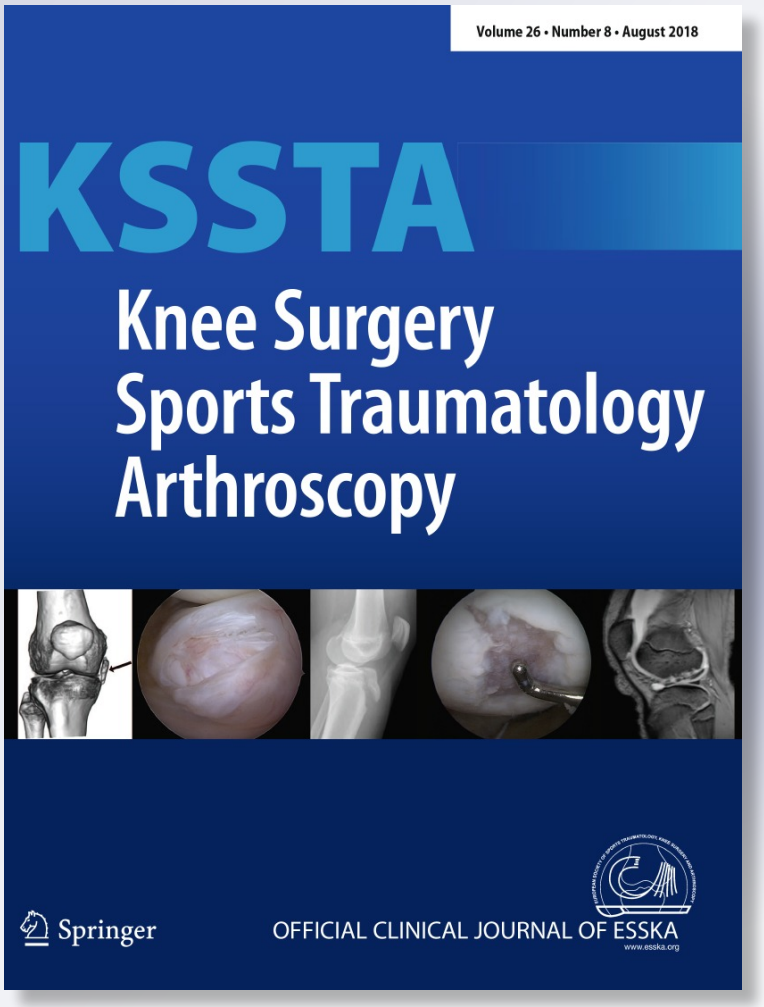

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ACL graft compression: a method to allow reduced tunnel sizes in ACL reconstruction

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Abstract

Purpose A common problem during ACL reconstruction is asymmetry of proximal–distal graft diameter leading to tunnel upsizing and graft–tunnel mismatch. Compression downsizing provides a graft of uniform size, allowing easy passage into a smaller tunnel. The purpose of this study was to quantify the graft compression technique and its effects on graft biomechanics and stability. It was hypothesised that compression downsizing would significantly reduce cross-sectional area (CSA); that no significant changes in graft biomechanics would occur; graft fixation stability would be improved.

Method Sixty-eight non-irradiated peroneus longus (PL) tendons were investigated. Twenty were halved and paired into ten four-strand grafts, 20 strands were compressed by 0.5–1 mm diameter and changes in CSA recorded using an alginate mould technique. The following properties were compared with 20 control strands: cyclic strain when loaded 70–220 N for 1000 cycles; stiffness; ultimate tensile load and stress; Young's modulus. 24 PL tendons were quadrupled into grafts, 12 were compressed and all 24 were submerged in Ringer's solution at 37 °C and the CSA recorded over 12 h. Twelve compressed and 12 control quadrupled grafts were mounted in porcine femurs, placed in Ringer's solution for 12 h at 37 °C and graft displacement at the bone tunnel aperture recorded under cyclic loading.

Results Mean decreases in CSA of 31% under a stress of 471 kPa and 21% under a stress of 447 kPa were observed for doubled and quadrupled grafts, respectively. Compressed grafts re-expanded by 19% over 12 h compared to 2% for controls. No significant differences were observed between compressed and control grafts in the biomechanical properties and graft stability; mean cyclic displacements were 0.3 mm for both groups.

Conclusions No detrimental biomechanical effects of graft compression on allograft PL tendons were observed. Following compression, the grafts significantly increased in size during in vitro joint simulation. No significant difference was observed in graft stability between groups. Graft compression did not cause adverse mechanical effects in vitro. Smaller tunnels for compressed grafts reduce bone loss and ease anatomical placement.

Keywords Anterior cruciate ligament · ACL reconstruction · Compression downsizing · Tendon graft biomechanics

Abbreviations

ACL	Anterior cruciate ligament
ACLR	Anterior cruciate ligament reconstruction
ANOVA	Analysis of variance
CSA	Cross-sectional area
ICC	Intraclass correlation coefficients
n.s.	Non-significant
PL	Peroneus longus
SD	Standard deviation
SPSS	Statistical Package for the Social Sciences
USA	United States of America
UFL	Ultimate failure load
UTS	Ultimate tensile stress

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Introduction

Bone tunnel enlargement after ACL reconstruction (ACLR) has been suggested as a factor contributing to graft failure [1]. The aetiology of this is unclear but is thought to be a combination of mechanical and biological factors [2, 3]. A common problem encountered during ACLR is asymmetry of proximal–distal graft diameter, and one way to deal with this is to drill the tunnel to match the larger end of the graft to avoid difficulty during graft passage. The resulting graft–tunnel mismatch in diameter creates effective tunnel widening before mechanical or biological factors compound the issue. The result is reduced bone stock, which may compromise revision surgery. It has been suggested that extravasation of synovial fluid can impair graft integration in the period following surgery and thereafter cause tunnel widening, especially when cortical suspensory fixation is used, which may allow cyclic micromotion across the graft–bone interface [4–8]. Compression downsizing, in which the graft is squeezed to a smaller uniform diameter, could provide an ACL graft of uniform size, allowing easy passage into a smaller tunnel. It could also be hypothesised that a compressed graft would have the further advantage that it could relax and expand within the bone tunnel, effectively blocking the passage of synovial fluid. In addition, studies with up to 15 years follow-up have reported ruptured ACL grafts in 4.1–11.2% of patients [9, 10]. Smaller tunnels would preserve bone stock to aid revision surgery, in addition to making it easier to keep the tunnel aperture within the native ACL attachment.

The effects of a clinically applicable graft compression technique on the biomechanical properties of grafts have not been reported. Much like the commonly accepted technique of pre-tensioning ACL grafts prior to insertion and fixation [11], compression downsizing is a simple concept that aids surgical technique and may benefit the patient. During surgery, graft downsizing is obtained easily by pulling the graft through progressively smaller diameter sizing tubes. When a larger tension than that needed to simply slide the graft through the tube is felt (typically when the diameter is 0.5 or 1 mm below the nominal graft size), the graft can be left inside the tube while the ACL procedure progresses, and then may be pulled into an undersized bone tunnel.

The purposes of this study were to measure changes in clinically available human allograft CSA during and following compression downsizing, in addition, to identify any effects on graft biomechanics and fixation stability.

It was hypothesised that compression downsizing would produce a significant reduction in CSA that would subsequently expand during in vitro joint simulation; that no significant changes in graft biomechanics would be observed; and that graft fixation stability would be improved. The clinical relevance is that smaller tunnels will reduce the volume of bone lost during ACL reconstruction, and compressed grafts—which are expected to swell when immersed in joint fluid—may also grip the wall of the bone tunnel and thereby reduce slippage under loading post-surgery and before healing occurs.

Materials and methods

Following ethical approval, 68 peroneus longus (PL) chemically treated and non-irradiated (BioClense, RTI Surgical, Alachua, FL, USA) allograft tendons were obtained and assigned to test groups (Fig. 1); they were stored at − 20 °C before use. These tendons were available for testing, and their mechanical properties do not differ from those of other hamstring tendons [12].

Quantification of graft compression

This assessment was performed for each of the two common methods of four-strand ACL graft preparation: two doubled tendons; one quadrupled tendon (Fig. 1). The allograft tendons were wrapped in a surgical swab immersed in isotonic Ringer's solution at room temperature for 2 h prior to testing to normalise any osmotic effect between specimens. Ten PL tendons were doubled over in pairs (that is, a four-strand graft structure) and placed in a custom-designed steel compression block which provided a 75-mm-long cylinder of a given diameter under a compressive load (Fig. 2). Starting with a 12.5-mm-diameter block, progressively smaller blocks were used until a 20 N compressive load was unable to close the block completely; this was taken as the pre-compression graft diameter.

Using a non-destructive alginate moulding method [13] (Blueprint cremix, Dentsply DeTrey, Germany), moulds were taken of the central 40 mm of each strand, then four 2 mm axial slices were taken at equal intervals and the mean pre-compression CSA (mm^2) was calculated (Image J, National Institute of Health, Bethesda, MD, USA) from a digital image (Canon EOS 100D). Twenty preliminary tests were carried out on a steel bar of known cross section on five separate days to confirm the accuracy and repeatability of this method. The graft was then placed in a compression block 1 mm less in diameter and a screw-driven Instron 5565 materials testing machine (Instron, High Wycombe, UK) used to apply a 10 N/s compressive load until a final force was recorded at closure, this was held for 10 min. A compression of 1 mm in diameter was chosen as this was sufficient to produce grafts of uniform diameter and the forces required to compress grafts beyond

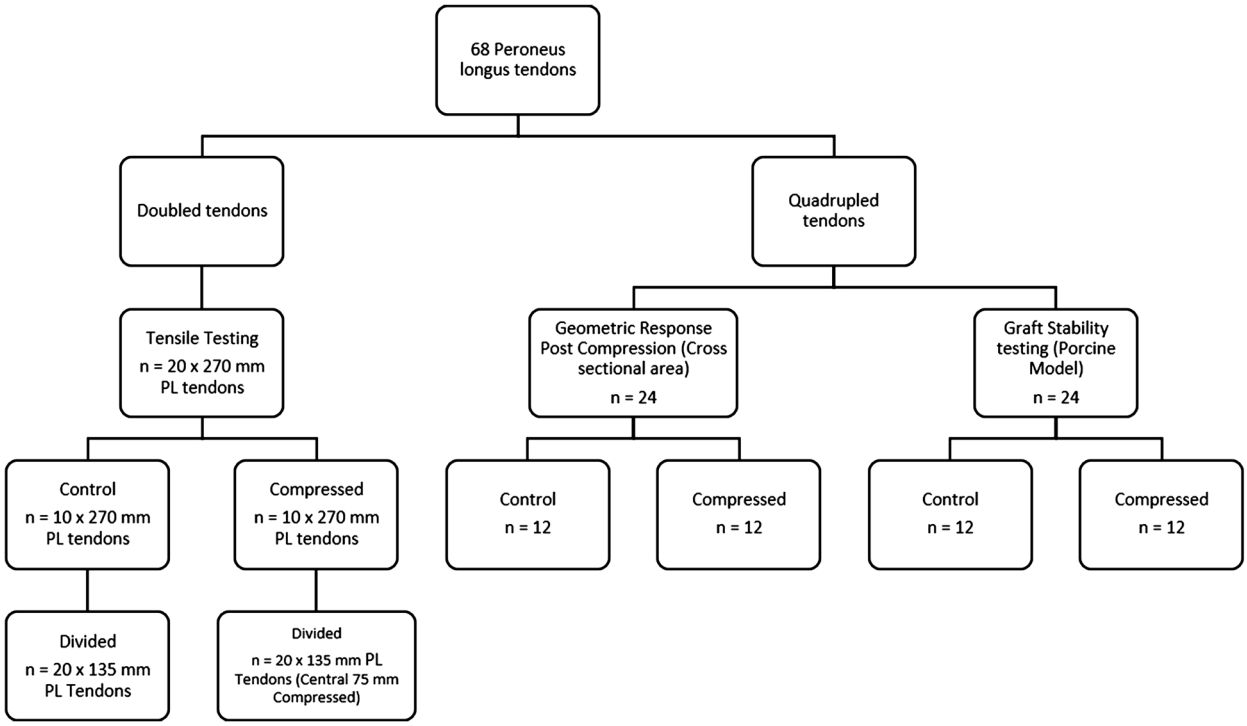

Fig. 1 A total of 68 peroneus longus allograft tendons were allocated across each arm of the study

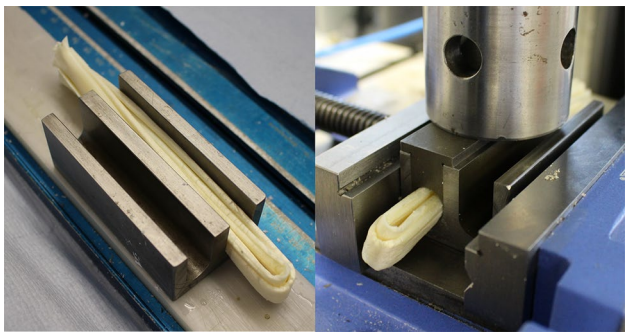

Fig. 2 A pair of peroneus longus allograft tendons doubled over to create a four-strand soft tissue graft. The graft was placed into a custom-designed compression block mounted within a materials testing machine

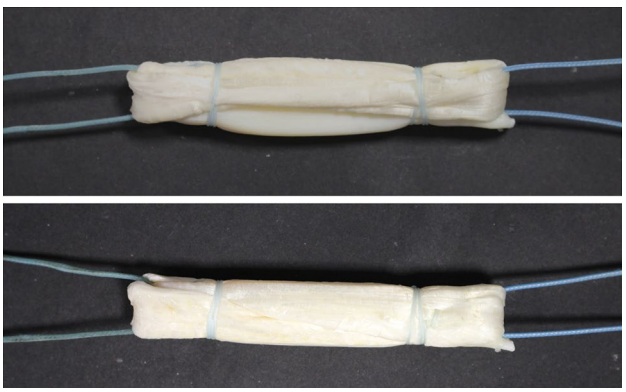

Fig. 3 **a** An uncompressed quadrupled peroneus longus allograft tendon. **b** The same graft took on a more compact and uniform shape following compression downsizing

this were not easily produced in the clinical setting without specialist equipment. Each PL tendon was then divided into half and post-compression CSA measurements were taken at the same positions. Thirty PL tendons ($n = 24$ in Fig. 1, plus a further six from the pilot testing) were quadrupled as previously described [14], to create a clinical ACL graft structure, and underwent the same protocol (Fig. 3).

ACL graft geometric response: in vitro simulation

Twenty-four quadrupled PL tendons were divided into 12 pairs consisting of one control and one graft compressed along its entire length as described above and shown in Fig. 2. These were submerged in Ringer's solution (Merck, Darmstadt, Germany) at 37 °C under 20 N of tension [15]. The mean CSA was measured at 2, 4, 6, 8 and 12 h as previously described.

Tensile testing

Forty single-stranded grafts (the two groups of 20 divided grafts in Fig. 1) were divided into matched pairs within control and compressed groups, and the mean CSA of each graft recorded before testing. An Instron 8874 servohydraulic materials testing machine (Instron, High Wycombe, UK) was used for the biomechanical testing. Each specimen was mounted in a pair of cryo-jaws positioned to match the central 40 mm of the tendon to approximate the intra-articular length of an ACL graft [16]. Liquid CO_2 was used to freeze the jaws before and after tightening to prevent slippage of the tendon in the clamps [17]. Each clamp had rounded edges to reduce the stress riser at the tendon interface. Two types of test were conducted: a non-destructive cyclic tensile loading test followed by an ultimate tensile stress (UTS) test.

After applying a pre-conditioning loading of 20 cycles of 0–70 N, the load on the tendon was increased to 145 N and then 1000 cycles between 70 and 220 N were applied at a frequency of 1 Hz. This protocol represented the loads experienced by the ACL during normal walking [18]. The mean, range, minimum and maximum loads and length for each cycle were recorded.

The specimens were then tensile tested to failure at a crosshead speed of 1000 mm/min, recording position and load. These data and the CSA were used to calculate the stiffness, UTS and Young's modulus for each tendon.

ACL graft fixation stability: in vitro simulation

Twenty-four large porcine femurs were stripped of all soft tissue and the medial condyle resected at the level of the inter-condylar notch. The femoral shaft was mounted in a stainless steel pot of 60 mm diameter using poly-methylmethacrylate (Simplex Rapid, Kemdent, UK). The CSA of 12 control and 12 compressed quadrupled PL tendons was measured and the appropriate socket was created to a depth of 30 mm using a FlipCutter (Arthrex Inc., Naples, FL, USA). Cortical fixation was achieved with a 15-mm RetroButton (Arthrex). Each specimen was placed in Ringer's solution at 37 °C for 8 h with a 20 N tensile load applied to the graft. The servohydraulic Instron materials testing machine was used to conduct the same cyclic loading protocol outlined above. The specimen was mounted in a specifically designed test rig with several degrees of freedom so that the ACL graft socket was aligned on the loading axis, with an orientation of approximately to 30° in the coronal plane [19] and 60° to the long axis of the porcine femur in the sagittal plane (Fig. 4). After pre-conditioning, the tensile load was held at 50 N while a black 2-0 nylon suture was passed in a 'figure of 8' fashion through all four graft strands 5 mm from the bony aperture. Similarly, a black plastic marker mounted on the bone produced a sharply defined edge 2 mm from the bony aperture. Any increase of the distance between the tendon- and bone-mounted markers was taken to represent slippage of the graft out of the bone tunnel. Eighteen-megapixel digital images were recorded at a rate of 25/s (Canon EOS 100D) and processed using Image J (NIH) and cyclic displacement was recorded over 1000 cycles.

Fig. 4 In vitro ACL reconstruction simulation: a quadrupled peroneus longus allograft mounted within a porcine femur with the bone tunnel aligned to the loading axis of a servohydraulic Instron 8874 materials testing machine

Research ethics committee approval

This study was approved by the Imperial College Healthcare Tissue Bank, HTA licence 12275, REC Wales approval 12/WA/0196, project R13058.

Statistical analysis

To assess the repeatability of the moulding technique to measure CSA, intraclass correlation coefficients (ICC) were determined. Paired samples *t* tests were used to analyse pre- and post-compression CSA for each of the two graft constructs. The independent-samples *t* test was used to analyse data from the quantification of the graft compression technique, cyclic loading and ultimate load to failure testing.

The CSA and cyclic motion data from the two in vitro simulations were analysed using a one-way analysis of variance (ANOVA) with a post hoc Tukey's test. The level of significance was set at 0.05. Statistical analysis was performed using statistical software (Statistical Package for the Social Sciences (SPSS) 21.0; SPSS Chicago IL, USA).

In addition, pilot test data from six specimens were used to perform relevant prospective power calculations (using software at https://www.stat.ubc.ca), to confirm that the numbers of tests would exceed the thresholds of 80% power at 95% confidence. This showed that eight pairs of values would give a power of 96% with 95% confidence for one-tailed difference between two dependent means.

Results

Power calculation

With groups of 12 pairs of values, the retrospective power analysis found that an actual power of 0.996 was achieved with 95% confidence, based on graft compression data.

Graft compression

The CSA of the metal bar was 87.4 mm^2, and the moulded sections had a mean of 87.4 ± 1.6 mm^2. Test–retest analysis of the alginate moulding technique revealed high repeatability, with a mean difference of 3 mm^2 (that is, approximately ± 3%) on different test days, and an ICC of > 0.9. The mean initial CSA of the doubled PL tendons as a four-strand construct was 87 ± 9 mm^2 [mean ± standard deviation (SD)], equivalent to a graft diameter of 10.5 mm. A significant reduction in CSA was observed between the control and compressed groups ($P < 0.001$) of 16 ± 8% following a mean compressive pressure of 471 ± 196 kPa, giving a reduced diameter of 9.6 mm. In the quadrupled format, the CSA was 90 ± 9 mm^2, equivalent to 10.7 mm in diameter. The same protocol resulted in a significant reduction in CSA ($P < 0.001$) by 13% ± 4% following a mean compressive pressure of 447 ± 132 kPa, giving a reduced diameter of 10.0 mm.

ACL graft geometric response: in vitro simulation

The CSAs of the grafts before they were compressed were 91 ± 10 and 88 ± 8 mm^2, which equate to graft diameters of 10.8 and 10.6 mm, respectively. With immersion in Ringer's solution, significant increases of the CSA of compressed quadrupled grafts were observed at every time interval with a mean change in CSA at 12 h of 19 ± 9% (17 mm^2; $P < 0.004$), while the control group changed by 2 ± 5% [2 mm^2; non-significant (n.s.)], leading to swollen graft diameters of 11.8 and 10.7 mm, respectively (Fig. 5).

Tensile testing

No significant differences were found between the control and compressed groups in terms of cyclic strain, UFL, stiffness, UTS or Young's modulus (Table 1).

ACL graft fixation stability

No significant differences in cyclic displacement were observed between the control and compressed groups at any stage over 1000 cycles. Cyclic displacements of 0.3 ± 0.1 mm were observed at 1000 cycles for both the control and compressed groups.

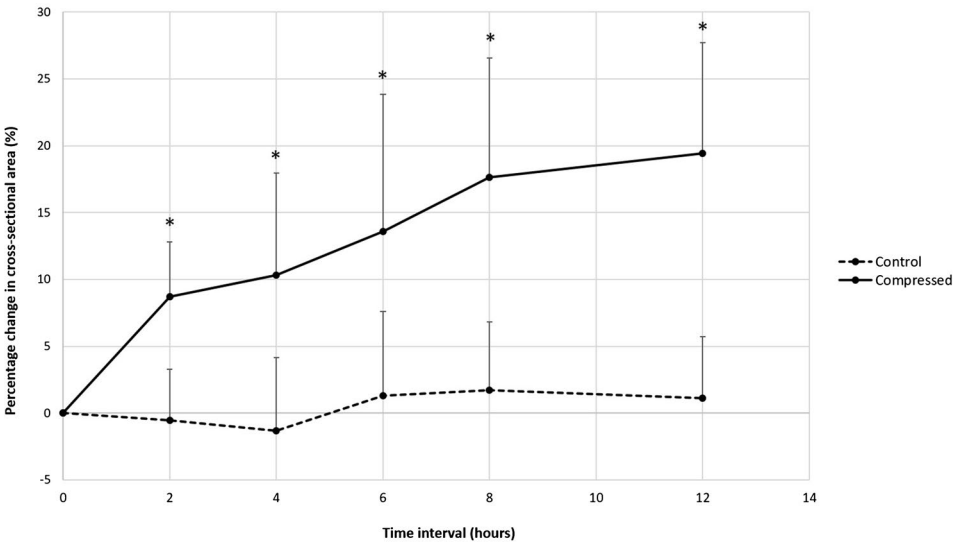

Fig. 5 ACL graft geometric response: in vitro simulation—percentage change in quadrupled allograft cross-sectional area over a 12-h period submerged in Ringer's solution at 37 °C under 20 N of tension (mean ± SD; $n = 24$; $^*P < 0.05$). Solid line: significant swelling of previously compressed grafts; interrupted line: non-significant change of CSA of uncompressed controls

Table 1 Biomechanical data (mean ± SD)

	ΔLength (mm)—1000 cycles	Cyclic strain (%)	Ultimate failure load (N)	Stiffness (N/mm)	Ultimate tensile stress (MPa)	Young's modulus (MPa)
Control	1.6 (± 1.1)	4 (± 3)	1629 (± 430)	329 (± 98)	82 (± 24)	671 (± 185)
Compressed	1.8 (± 1.0)	5 (± 2)	1664 (± 332)	343 (± 71)	93 (± 21)	764 (± 164)

Discussion

The most important finding in this study was that a four-strand ACL allograft construct could be compressed so that it would fit into a bone tunnel with 1 mm smaller diameter, thus reducing bone removal by approximately 20%. As had been hypothesised, there were no significant changes to the inherent biomechanical properties of the ACL allograft material following graft compression. However, contrary to our hypothesis, although compressed grafts were found to expand when soaked in Ringer's solution, this expansion in situ did not lead to greater resistance to slipping from the bone tunnel under cyclic loading.

Compression downsizing offers the opportunity for a 21% (size 11–10 mm) to 31% (size 8–7 mm) smaller bone tunnel CSA with no compromise in the biomechanical properties of the reconstruction, enabling a smaller graft tunnel to be used, which may be advantageous if revision surgery is required. With the advent of 'anatomic' ACL reconstruction, studies have highlighted the importance of graft placement within the native ACL attachment sites both in vitro [20] and in vivo [21, 22]; a smaller tunnel size would make it easier to keep the aperture within the ACL attachment area. This is of particular importance in the context of the small female knee or in a child. ACL injury has become increasingly common in children and adolescents as the number of young athletes involved in high-demand sport increases [23, 24], such cases reportedly account for 3% of all ACL injuries [25]. Recent anatomical studies have highlighted that the ACL attaches posterior to the anterior edge of the lateral inter-condylar ridge [26], and a 9-mm graft may encroach beyond the ACL attachment even when correctly positioned. Some surgeons utilise a four-tunnel (double-bundle) technique and in this context a reduction in tunnel size would make it easier to maintain a bony bridge and avoid coalescence between pairs of tunnels [27]. Similarly, in the context of the multi-ligament knee injury, smaller tunnels decrease the chances of tunnel convergence during graft fixation.

Despite the re-expansion of the compressed grafts, which was expected to increase the pull-out strength from the bone tunnel, no significant difference was observed in graft fixation stability at the bony aperture when compared to the control group. However, extravasation of synovial fluid has been proposed as a possible cause of osteolysis at the graft–osseous interface, leading to tunnel enlargement [5, 6]. Despite not adding any mechanical stability to the graft, the press-fit created by compression downsizing and re-expansion may impede or avoid extravasation of synovial fluid into the graft tunnels; further clinical investigation is required.

Hwang et al. [28] performed a prospective study of 35 patients in whom the femoral ACL graft tunnels were under drilled by 0.5 mm and the graft forced through. They reported no significant difference in total tunnel volume between these and the control specimens when evaluated using computed tomography at 1 year, no difference in International Knee Documentation Score or KT-2000 laxity. However, allografts are often oedematous and the perioperative results presented in this study show that an allograft can be compressed and passed into a socket of 1 mm smaller diameter, representing 20–30% preservation of bone stock. Although no definitive clinical benefit has yet been identified, the clinical authors of the present study have now adopted ACL graft compression to preserve native bone stock whilst maintaining ease of graft passage in both the primary (autograft) and revision (autograft/allograft) setting. This is done easily by pulling the freshly prepared graft into progressively smaller sizing tubes.

The major limitation of this study is that it used chemically treated PL allografts due to the number of samples required, their consistency, cost and availability. The morphology of the PL tendon is partially cylindrical and partially ribbon-like, these were allocated equally throughout to minimise any bias. This study was not intended to support allografts rather than autografts, and the authors are unaware of evidence to suggest that the graft compression would have a different effect on autografts, other than the practical observation that fresh autografts are less oedematous and so only reduce 0.5 mm diameter. Other limitations are: the data relate only to the situation immediately post-surgery with a limited number of cycles, excluding biological factors such as ligamentisation of the ACL graft; porcine femurs were used for more consistent cancellous bone quality when compared to human cadaveric specimens which are older than the patient demographic for ACL reconstruction. The structural and material properties of the PL allografts found in this study were in line with published data on ligaments and grafts: the ultimate failure load (UFL) of the ACL was found to be 2160 N in the young adult [29] whilst Noyes

et al. [30] reported a UFL of 1725 N and a UTS of 37.8 MPa. Similarly, UFL values of 1784–2900 and 2421 N have been reported for the commonly used patellar tendon and doubled semitendinosus/gracilis hamstring grafts, respectively [30, 31]. Pearsall et al. [32] tested fresh-frozen PL autografts, reporting a mean UFL of 2483 N and a mean stiffness of 244 N/mm in doubled PL tendons.

Conclusion

The clinical implication of this study is that graft compression, which does not increase cost or operative time, appears to preserve bone stock whilst aiding surgical technique. No detrimental biomechanical effects on chemically treated allograft PL tendons were observed after a compression protocol that used easily applicable pressures. Following graft compression, these tendons significantly increased in size during in vitro joint simulation. Although no significant difference was observed in graft fixation stability between groups, graft compression should aid graft passage, anatomic positioning within the attachment and preservation of bone stock during ACL reconstruction.

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Compliance with ethical standards

Conflict of interest None declared.

Ethical approval This study was approved by the Imperial College Healthcare Tissue Bank, HTA licence 12275, REC Wales approval 12/WA/0196, project R13058.

Informed consent Not required for this study, which was covered by a Research Ethics Committee permit.

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