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Effect of diameter reduction by compression on peroneus longus allograft integrity: A biomechanical analysis

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ABSTRACT

Background: There is broad consensus regarding the minimum required graft diameter in anterior cruciate ligament (ACL) reconstruction surgery; a diameter of at least 8 mm is recommended when using hamstring tendon grafts. However, it remains unclear what happens when a larger-diameter graft is compressed to a smaller diameter, and how this alteration may affect the biomechanical properties of the graft. The objectives is determine whether the biomechanical strength of compressed allografts is preserved after reducing their diameter, and whether compression techniques could be considered a viable option for smaller patients such as women and children. Our hypothesis is that a compressed graft may retain similar strength to the original, despite having a reduced diameter.

Methods: This was a cadaveric biomechanical case-control study using 30 peroneus longus (PL) tendon allografts. The grafts were divided into three groups: Group 0 (8 mm uncompressed grafts), Group 1 (9 mm uncompressed grafts), and Group 2 (10 mm with a graft tube, compression to 9 mm). Each graft was tested using a Zwick/Roell Z005 tensile testing machine, undergoing cyclic loading between 50 and 250 N at 1 Hz for 1000 cycles, followed by continuous tensile loading to failure. The maximum failure force (Fmax) was recorded for each graft. Statistical analyses were performed using STATA v.18.5.

Results: Group 1 (9 mm uncompressed grafts) showed a mean maximum failure force of 1902 N (SD: 432.12; 95 % CI). Group 2 (compressed grafts) showed a mean Fmax of 1608 N (SD: 233.12; 95 % CI), with a statistically significant difference between the two groups ($p = 0.037$). Group 0 (native 8 mm grafts) showed a mean Fmax of 1341 N (SD: 936.67; 95 % CI). No statistically significant difference was found between Group 0 and Group 2 ($p = 0.196$).

Conclusions: The results indicate that the compressed group had lower resistance compared to the original 9 mm grafts. However, when compared to the smaller-diameter grafts, no significant difference in strength was observed. Nonetheless, the compressed grafts showed a tendency to withstand higher loads than the native 8 mm grafts.

1. Background

One of the most common knee ligament injuries is the rupture of the anterior cruciate ligament (ACL), and ACL reconstruction (ACLR) is a frequently performed procedure. It has been reported that there are between 100,000 and 250,000 patients per year with ACL injury in the US[1,2] and its reconstruction has been performed in 60,000 to 175,000 patients.[2] There are various techniques, types of graft, and systems of fixation for ACLR. There are multiple options for ACLR including allografts. There have been reports suggesting that peroneos longus (PL) graft may be a superior alternative graft for the ACL, because of the tensile strength or the regenerative capacity of the graft after the implantation.[3,4]

However, inherent variability in graft diameter has been noted as a disadvantage,[5] since smaller graft diameters have been correlated with an increased risk of re-rupture.[6] Magnussen et al.[7] evaluated hamstring

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autograft diameter as a predictor of graft failure and the need for revision. In a study of 256 patients undergoing ACL reconstruction with hamstring autografts, grafts larger than 8.5 mm had a revision rate of 1.7 %.[8] In contrast, grafts measuring 7–7.5 mm and 7 mm or less had revision rates of 11.3 % and 33 %, respectively. The goal is to obtain a graft size that is appropriate for each patient according to their anatomy and functional demands.[9]

There have been various methods described to augment the final graft diameter achieved from a standard hamstring harvest.[10,11] These propose that these methods often use more hamstring strands to create the desired graft thickness. To optimize graft diameter, a compression tube can be used to enhance tubularization, reduce graft diameter to facilitate passage, and maintain maximal strength. To date, only one study have evaluated whether the biomechanical characteristics of the graft are affected by the use of compression tubes.[12]

Our purpose is compare the biomechanical characteristics of the peroneus longus allograft after diameter reduction using compression tubing. The objective of this cadaveric biomechanical study is to investigate whether reducing the diameter of a peroneus longus allograft with a compression tube preserves greater tensile strength compared to an allograft of smaller original diameter.

2. Methods

Ethical approval was obtained from the Health and Disability Ethics Committee of our center. We are not requesting funding, cadaveric tendons (which will be provided by Sports Medicine LifeNet Health-LifeNet Health), laboratory facilities, access to the biomechanical testing machine (Zwick/Roell Z005), the biomechanics engineer, or the software required for data analysis.

Materials:

- Laboratory.
- Graft Compression Tubes Arthrex® (Fig. 1).
- Testing machine (Zwick/Roell Z005) (Fig. 2).
- 30 cadaveric peroneus tendons (non-irradiated).
- 30 #2 FiberWire® (Arthrex, Naples, Florida, USA).

All donors have to meet organ bank criteria for tissue donation and all allografts have to go through the same sterilization process, non-irradiated. 30 fresh frozen peroneus tendons will be recovered from 15 human donors, divided in 3 experimental groups by graft size:

Group 0: 10 freshly frozen peroneus tendons of 8 mm (double string) (Control Group).



Fig. 1. Graft compression tubes Arthrex®.



Fig. 2. Zwick/Roell Z005 tensile testing machine.

Group 1: 10 freshly frozen peroneus tendons of 9 mm (double string).
Group 2: 10 freshly frozen peroneus tendons of 10 mm (double string) with a graft tube, compression to 9 mm is achieved*.

*In case it is not possible to use 10 mm allografts to compress them to 9 mm, another available diameter could be used. The idea is to have an initial diameter that can be compressed 1 size and to be able to compare it with an allograft of the same size that has not been compressed.

A matching process for these 3 groups divisions were conducted in order to avoid any age-based bias on their biomechanical properties. Donor genders were uniformly distributed among the 3 groups with the matching process.

Specimen preparation: Each end of the tendon was sutured together using a #2 FiberWire® (Arthrex, Naples, Florida, USA) with a Krackow technique, creating a tendon loop. The sutured ends were fixed to the machine using two grooved compression clamps, attaching the ends of the suture to a stable metallic pivot under the compression plates to ensure attachment, avoiding any tendon gliding. The loop end was attached to a universal testing metallic hook (Fig. 3).

Biomechanical procedure: Each group is tested on a tensile testing machine Zwick/Roell Z005 (Ulm, Radeberg, Bickenbach, Germany), which performs cyclic loading followed by continuous tensile load in case of absence of failure in the former loading. Following pre-conditioning with 20 loading cycles ranging from 0 to 70 N, cyclic loading to the tendon varies between 50 and 250 N at 1 Hz for 1000 cycles, strength is determined by the normal strength at which the anterior cruciate ligament is put through in daily activities.[13] At the end of the cycles, grafts are elongated to failure (Fig. 3).

Test results are registered and processed by a software program. Cyclic loading is registered in a strain and load versus time graph and continuous loading is registered in a load versus time and a load versus

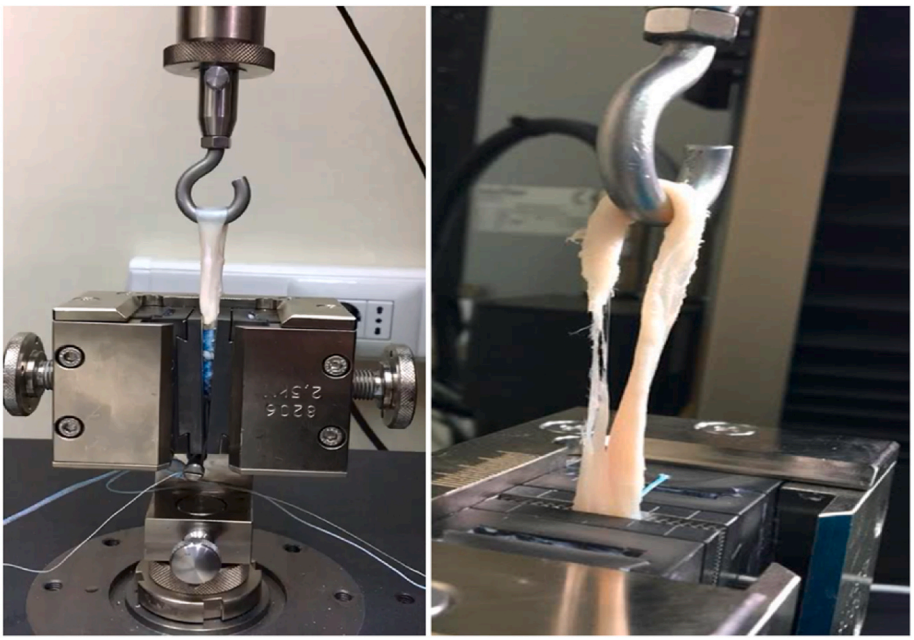


Fig. 3. The allograft undergoing cyclic loading between 50 and 250 N at 1 Hz for 1000 cycles, followed by continuous tensile loading to failure.

strain graphs. Load to first failure is determined by the first curve inflexion in the time versus load graph. Deformation is registered in millimeters.

2.1. Statistical analysis

A sample size of 10 tendons per group was calculated with a power analysis of 80 % before testing. A Shapiro-Wilks Normality test was performed to determine the distribution of the ultimate load to failure variable amongst the sample. In case of normal distribution between groups, a paired-samples *t*-test test was conducted ($\alpha = 0.05$) with the null hypothesis "the means in groups are equal", being tested against the alternate hypothesis, "the means are different between groups." In case of a non-parametric distribution a Wilcoxon signed-rank test were performed with the same null and alternative hypothesis using medians instead of means.

3. Results

This cadaveric biomechanical study evaluated the maximum tensile strength (Fmax) of peroneus longus allografts in two groups: Group 1 consisted of 9 mm diameter uncompressed grafts, and Group 2 consisted of the same 10 mm diameter grafts compressed to 9 mm using a compression tube. In Group 1, the mean Fmax was 1902 N (SD: 432), with a 95 % confidence interval of 1592.9–2211.1 N. In Group 2, the mean Fmax was 1608 N (SD: 233), with a 95 % confidence interval of 1441.2–1774.8 N. The difference between the two groups was 294 N (95 % CI: 32.2–620.2 N), which was statistically significant ($p = 0.037$), indicating that the uncompressed grafts had superior tensile strength compared to the compressed grafts (Table 1).

3.1. Two-sample t-test with equal variances

An additional comparison was made between a historical control of 8 mm diameter uncompressed grafts (mean Fmax: 1341 N; SD: 937; 95 % CI: 670.9–2011.1 N) and Group 2 (the 10 mm compressed to 9 mm grafts). Although the compressed grafts (Group 2) showed a higher mean tensile strength (1608 N vs. 1341 N), the difference was not statistically significant ($p = 0.393$), with a wide confidence interval (−908.3 to 374.3 N), reflecting considerable variability in the smaller uncompressed grafts (Table 2).

3.2. Two-sample t-test with equal variances

The results shows that the compressed group had less resistance than the original ones (9 mm diameter Max resistance 1902 N versus compressed 1608 N). When compared with the smaller graft, there wasn't a significant difference between the compressed group and the smaller group, nevertheless the compressed group shows a tendency to have more resistance than the smaller graft.

4. Discussion

The purpose of this cadaveric biomechanical study was to determine

Table 1
Statistical analysis of group 1 versus group 2.

Group|	Obs	Mean	Std. Err.	Std. Dev.	[95 % Conf. Interval]	
1 |	10	1902	136.6569	432.1471	1592.861	2211.139
2 |	10	1608	73.72321	233.1332	1441.227	1774.773
combined |	20	1755	82.75042	370.0711	1581.801	1928.199
vdiff |		294	155.2747		−32.21997	620.22
diff = mean (0) - mean (1						t = 1.8934
Ho: diff = 0						degrees of freedom = 18
Ha: diff < 0			**Ha: diff = 0**		**Ha: diff > 0**	
Pr(T < t) = 0.9628			**Pr(|T| > |t|) = 0.0745**		color = "#FF0000; ">**Pr(T > t) = 0.0372**	

Table 2
Statistical analysis of group 0 versus group 2.

Group |	Obs	Mean	Std. Err.	Std. Dev.	[95 % Conf. Interval]	
0 |	10	1341	296.2036	936.6779	670.941	2011.059
2 |	10	1608	73.72321	233.1332	1441.227	1774.773
combined |	20	1474.5	151.6739	678.3065	1157.043	1791.957
diff |		−267	305.2403		−908.2862	374.2862
diff = mean (0) - mean[1]						t = −0.8747
Ho: diff = 0						degrees of freedom = 18
Ha: diff <0			Ha: diff ! = 0			Ha: diff >0
Pr (T < t) = 0.1966			Pr (|T| > |t|) = 0.3932			color = "#FF0000; ">Pr (T > t) = 0.8034

the consequence of diameter reduction by compression using peroneo longus allograft on their tensile properties. Our study showed that by compressing a 10 mm graft to 9 mm, there is a statistically significant reduction in the maximum load to failure when the latter is compared with a 9 mm graft in its native noncompressed state. This raises the issue of whether by decreasing graft volume, compression may also reduce the biomechanical quality of the graft. The mean failure load of the uncompressed teeth at 9 mm (1902 N) was significantly greater than for the compressed group (1608 N) by an average of 294 N, this has to be considered along with the clinical implications, reduction of the ultimate tensile strength is not an equivalent to clinical failure, if the strength of the implanted graft is still higher than the anterior cruciate ligament would be subjected to during day to day activities and early rehabilitation.[14–16] Interestingly, the compressed group fared better than a historical cohort of native 8 mm grafts with respect to a higher mean load to failure (1608 N versus 1341 N) although this was not significant. This would indicate that by compressing a larger graft to a smaller diameter it may permit a superior to biomechanical strength even compared with using a smaller native graft. Such results suggest that compression could be a good method to ensure acceptable mechanical properties while resizing the graft to smaller anatomical dimensions such as those of women or children.[6,17,18]

Graft size, particularly diameter, has previously been highlighted as a predictor of revision risk in ACL reconstruction in the literature. Magnussen et al. for grafts ≤7.5 mm in size, indicating the need for methods that maintain or increase the size of grafts.[7] Our results contribute to this knowledge by indicating that let-alone compression techniques are not biomechanically equivalent to larger native-like grafts, but may have a biomechanical benefit over using small grafts by default. But there are important issues to consider when interpreting these findings. First, although compression may tailor the graft diameter it introduces variability in the microstructure of the tendon. For long-term graft behavior in vivo, internal reorganization of the collagen, water content, or changed viscoelastic characteristics could all play a role. These microstructural implications were not evaluated in this study but are an important area for future research. Additionally, the study was cadaver based. While cadaveric testing offers controlled and reproducible mechanical data, it does not take into consideration biologic integration, remodeling, or the healing potential in a live host. The allografts underwent cyclic loading and actual pull-to-failure testing in a laboratory environment, which may not accurately simulate in vivo joint forces, rehabilitation loads, or the influence of fixation techniques.

On the another hand, recent anatomic studies have demonstrated that insertion of the ACL is located posteriorly to the anterior edge of the lateral intercondylar ridge[19–21] and a 9 mm graft may overhang the native ACL footprint despite proper placement. Some surgeons prefer the double-bundle technique for anterior cruciate ligament reconstruction due to its biomechanical advantages and its more anatomical approach. This technique requires the creation of two femoral tunnels, and in this context, reducing the graft size may offer the benefit of facilitating the preservation of a bony bridge between the tunnels and preventing tunnel coalescence.[22–24] The same principle could be applied in multiligamentous knee reconstruction surgeries.

Another limitation is that the variation of the 8-mm control group is found to be broad with a large confidence interval and high standard deviation. This heterogeneity may have influenced the statistical power of detecting a significant difference between the compressed and smaller grafts group. However, the general trend still indicates that the mechanical properties are more advantageous for the compressed grafts.

Nevertheless, results are of clinical relevance. While performing surgery, surgeons frequently encounter the problem of graft securing not to mention the fact that some patients suffer from smaller size of intercondylary notch or are smaller themselves.[25,26] In those cases, it might be useful for fitting without losing this desired mechanical properties by compressing before implantation. Future work should address in vivo validation of our findings, the rate at which compressed grafts integrate as compared to uncompressed grafts, and whether observed differences in tensile strength predict clinically relevant outcomes, such as graft failure, knee stability, and return to sport. The application of compression methods for autografts and other types of allografts should also be further studied to extend its applicabilities.

5. Conclusion

This study demonstrates that compressing 10 mm peroneus longus allografts to 9 mm in diameter significantly reduces their tensile strength compared to their original uncompressed form. This suggests that mechanical integrity is compromised when downsizing grafts using compression, and caution should be exercised in clinical applications where maximum load-bearing capacity is critical. However, when compared to historical data from native 8 mm diameter grafts, the compressed 9 mm grafts did not show a statistically significant difference in tensile strength, although there was a trend toward higher resistance in the compressed group. These findings imply that, while compressing larger grafts diminishes their original strength, the resulting biomechanical properties may still be favorable compared to using smaller native grafts. In clinical practice, this may support the selective use of compression tubes to adjust graft diameter when necessary, especially if a native smaller graft is not available. Nonetheless, further in vivo research is needed to evaluate whether these biomechanical differences influence graft performance and clinical outcomes over time.

CRediT authorship contribution statement

Rafael Calvo Rodriguez: Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration. **Waldo Gonzalez Duque:** Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration. **David Figueroa Poblete:** Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration. **Minerva Itriago:** Conceptualization,

Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration. **Rafael Calvo Mena:** Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration. **Jorge Isla Villanueva:** Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing, Supervision, Project administration.

Informed consent

was obtained from all patients for the use of their data or photos.

Patient or guardian consent

This study did not involve human participants, identifiable personal data, or clinical images. Therefore, patient or guardian consent was not required.

Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional.

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Conflicts of interest

We have no conflicts of interest relevant to the content of this review.

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