

Ex Vivo Biomechanical Assessment of a Novel Multi-Strand Repair of Canine Tendon Lacerations

Chiara P. Curcillo¹ Daniel J. Duffy¹  Yi-Jen Chang¹ George E. Moore²

¹ Department of Clinical Sciences, College of Veterinary Medicine, North Carolina State University, Raleigh, North Carolina, United States

² Veterinary Administration, College of Veterinary Medicine, Purdue University, West Lafayette, Indiana, United States

Address for correspondence Daniel J. Duffy, BVM&S(Hons.), MS, FHEA, MRCVS, Diplomate ACVS-SA & ECVS, Department of Clinical Sciences, College of Veterinary Medicine, North Carolina State University, 1052 William Moore Drive, Raleigh, NC 27607, United States (e-mail: djduffy@ncsu.edu).

Vet Comp Orthop Traumatol

Abstract

Objective This study aimed to evaluate the effect of increasing the number of suture strands traversing the transection site, level of suture purchase and depth of suture penetrance on the biomechanical properties of repaired gastrocnemius tendons.

Study Design Thirty-eight adult cadaveric gastrocnemius tendons were randomized, transected and repaired with either two-, four- or six-strand locking multi-level repair. Tensile loads required to create a 1 and 3 mm gap, yield, peak and failure loads and failure mode were analysed. Significance was set at $p < 0.05$.

Results Mean \pm standard deviation yield, peak and failure force for six-strand repairs was 90.6 ± 22.1 N, 111.4 ± 15.2 N and 110.3 ± 15.1 N respectively. This was significantly greater compared with both four-strand (55.0 ± 8.9 N, 72.9 ± 7.8 N and 72.1 ± 8.2 N) and two-strand repairs (24.7 ± 8.3 N, 36.5 ± 6.0 N and 36.1 ± 6.3 N) respectively ($p < 0.001$). Occurrence of 3 mm gap formation was significantly less using six-strand repairs ($p < 0.001$). Mode of failure did not differ between groups with all repairs (36/36; 100%) failing by suture pull-through.

Conclusion Pattern modification by increasing the number of suture strands crossing the repair site, increasing points of suture purchase from the transection site and depth of suture penetrance is positively correlated with repair site strength while significantly reducing the occurrence of gap formation in a canine cadaveric model. Additional studies *in vivo* are recommended to evaluate their effect on tendinous healing, blood supply and glide resistance prior to clinical implementation.

Keywords

- biomechanical properties
- tendon repair
- suture
- canine tendon injury
- animal models of human disease

Introduction

Tendons are a highly organized connective tissues that transmit muscle derived force to bone¹ with lacerations or degenerative injuries seldom spontaneously resolving without surgical intervention.² Of tendinous injuries, which account for $<1\%$ of all musculoskeletal diagnosis,³ the common calcaneal tendon (CCT), also known as the Achilles tendon in humans, represents the most common site of tendinopathy in the dog^{2,4} and can be acute or chronic in nature.^{5–10} The CCT, which functions primarily to maintain

extension of the talocrural joint,^{11–13} is comprised of three distinct units; the paired gastrocnemius tendons (GT), the superficial digital flexor tendon and the common or accessory tendon composed of the biceps femoris, gracilis and semitendinosus muscular contributions.¹¹ Surgical intervention followed by postoperative immobilization and rehabilitation is indicated in cases of CCT laceration.² The goal of surgical intervention is to prevent or minimize gap formation and maintain tendinous blood supply at the repair site.¹⁴ A successful repair allows for direct contact healing and minimizes the development of a fibrous scar.^{15–17} With

received

March 19, 2020

accepted after revision

January 15, 2021

© 2021. Thieme. All rights reserved.
Georg Thieme Verlag KG,
Rüdigerstraße 14,
70469 Stuttgart, Germany

DOI <https://doi.org/10.1055/s-0041-1725014>.
ISSN 0932-0814.

gaps >3 mm at the repair site, leading to the deposition of scar tissue, which is mechanically inferior and leads to increased risk of re-rupture following surgical intervention.^{14–18}

Suture tenorrhaphy is the most commonly utilized surgical repair technique in both veterinary and human medicine.^{19–23} Both locking loop (LL) and three-loop pulley (3LP) patterns are frequently implemented for canine tenorrhaphy.^{18,24–27} Within the human literature, it has been demonstrated that increasing the number of strands traversing the repair site increases the overall strength of tendon repair.^{19,28–38} In the veterinary literature, the 3LP has been demonstrated to be biomechanically superior, compared with the LL pattern,³⁹ likely as it relies on multiple divergent passes and utilizes six strands that cross the repair site.^{25,39} The LL has previously been demonstrated to be inferior to 3LP regarding loads tolerable prior to failure and ultimate tensile strength.^{24,33} However, some of the technical differences between patterns, such as ease of suture placement, repeatability, uniform apposition along the length of the repair site, reduction in repair site bunching and less distortion of the repair site upon removing slack from the construct and suture knotting among LL repairs.^{16,26,40,41} Modifications to the LL pattern, such as increasing the number of suture strands, increasing purchase from the repair site and depth of suture placement, may result in a comparable or superior pattern to the 3LP and warrants further investigation. Biomechanical analysis using an equine model has shown that double and triple LL sutures had similar strength to a 3LP repair.⁴²

To date, knowledge regarding surgical factors and successful outcomes during human tendinous repair have been established in comparative animal studies.^{14,16,19,32,43,44} In addition to the number of core suture strands crossing the repair,⁴⁴ the calibre of the core suture,^{45,46} depth of core suture penetrance,⁴⁷ placement of an epitendinous suture,⁴⁰ depth of epitendinous placement⁴⁸ and increasing the distance of epitendinous suture bite placement from the transection site⁴⁸ have all been identified as important surgical factors regarding the repair. Sequential increases in each of these evaluated variables resulted in increased strength, stiffness and resistance of the repair to the development of gap formation.^{34,38–41} To date within veterinary literature, the direct effects of tenorrhaphy pattern modifications increasing the number of suture strands traversing the repair site, suture purchase and depth of suture penetrance in to the tendinous tissue on the mechanical properties of time-zero canine tendon repairs are lacking and warrant further focused evaluation.

The objective of this study was to evaluate the effect of increasing the number of suture strands traversing the transection site, level of suture purchase and depth of suture penetrance on the biomechanical properties of canine GT repairs. We hypothesized that this novel pattern modification of a locking repair technique will improve the biomechanical properties of repaired canine gastrocnemius constructs and loads tolerable at the repair site while decreasing the occurrence of gap formation between tendon ends.

Materials and Methods

Specimen Collection

The protocol of tendinous acquisition, specimen cryopreservation, isolation of individual gastrocnemius musculotendinous units, suture placement, pattern refinement and biomechanical evaluation of the tested constructs was determined based on a prior pilot study. Cadavers were serially obtained, weighing 26 to 32 kg and received within 1 hour from a local animal shelter, following intravenous euthanasia (Beuthanasia-D Solution, Merck, Whitehouse Station, New Jersey, United States) for reasons unrelated to this study. Due to secondary use of cadaveric tissues, our institution did not require an animal care and use committee protocol for the purposes of this study.

Specimens

Paired hindlimbs were harvested from 18 medium-to-large breed dogs that were skeletally mature and whose sex was not recorded. An orthopaedic examination was performed by a board-certified small animal surgeon (D.J.D.) on all cadavers to confirm the absence of musculoskeletal disease or gross tendon pathology. Any cadaveric limbs with visible abnormalities of bone, tendon or musculotendinous unit were excluded from study inclusion.

All components of the CCT were individually isolated. The gastrocnemius, originating from the supracondylar eminence on the caudodistal femur and inserting on the tuber calcanei, was carefully dissected in entirety along its length (►Fig. 1A). Using a number 10 Bard-Parker blade, the superficial digital flexor tendon was transected 2 cm proximal to its musculoskeletal junction and removed distal to the calcaneal tuberosity (►Fig. 1B). The distal femur was then transected using a band saw (Delta Power Equipment Corp., Anderson, South Carolina, United States) 5 cm proximal to the femorotibial joint articulation. The talocrural joint was then disarticulated by a sharp incision with a number 10 Bard-Parker blade by controlled incision into the joint capsule and transection of the supporting collateral ligaments. To facilitate rigid bone clamp fixation during experimental testing, tissue distal to the tarsus was left intact. At a level 1 cm proximal to the femoral trochlear groove, a 4.5 mm bone tunnel was drilled in a mediolateral direction into the distal femoral diaphysis. Isolated paired hindlimbs from the same dog were individually wrapped in saline (0.9% NaCl) soaked gauze and stored within a thermostatically controlled environment at -20°C. Specimens were then completely thawed at 21°C for 10 to 12 hours prior to definitive testing.

Following a single controlled thaw cycle, each specimen received a standardized tenotomy in a transverse plane across the paired GTs using a number 10 Bard-Parker scalpel blade. Tendinous transection was performed on a hard, durable surface to provide counter pressure and allow a uniform cut to be made. Tenotomies were performed at a measured distance of 2 cm proximal to the tuber calcanei using a calibrated mm ruler. Following transection, the distal tendon stump of the GT was photographed at a set distance of 8 cm (iPhone 8 camera, Apple Inc., Cupertino, California,

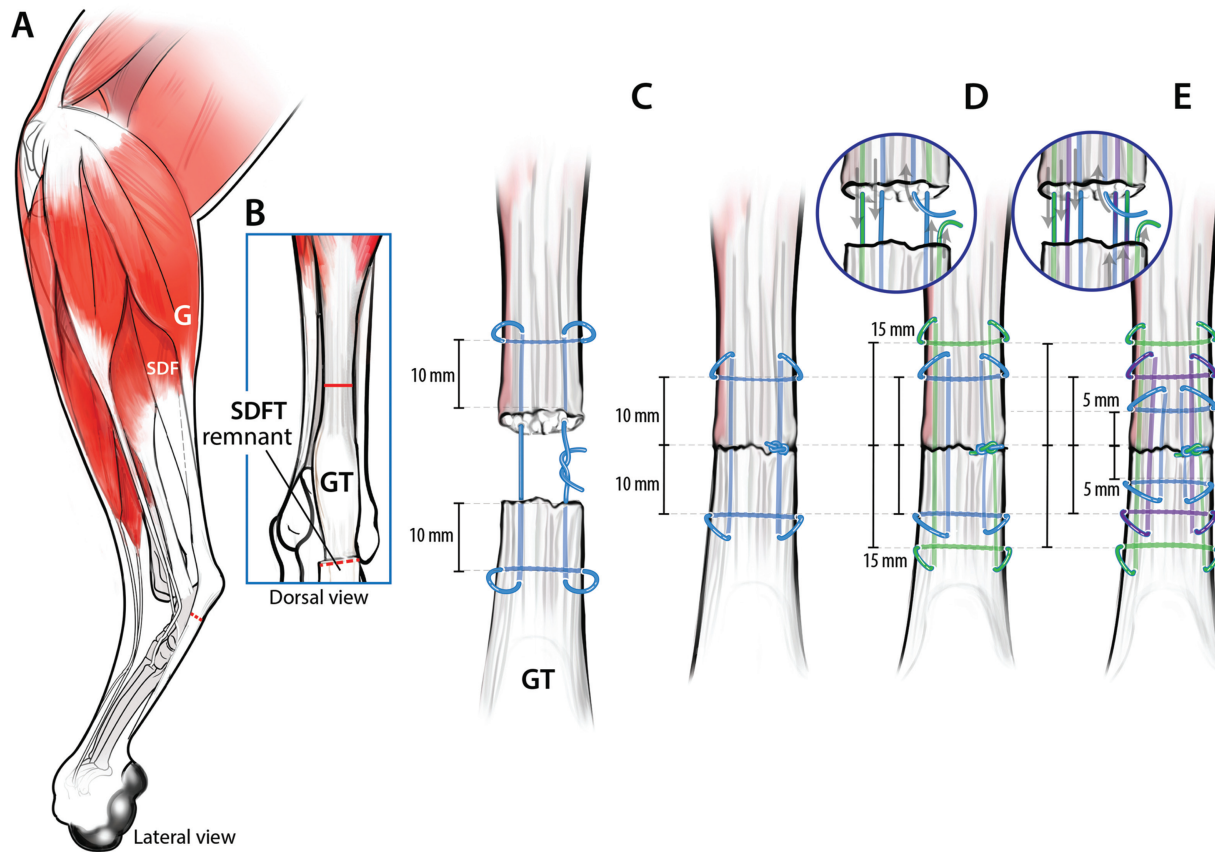


Fig. 1 Schematic representation of the canine gastrocnemius tendons (GT) (1A) in a cadaveric model. The superficial digital flexor tendon has been removed (1B). For group 1, a two-strand repair is pictured, with transverse bites placed 10 mm from the level of transection (1C). For group 2, a four-strand repair was completed by placing the first and second transverse bites 10 mm and 15 mm from the tendon end respectively (1E). For group 3, a six-strand repair placed transverse bites placed 5, 10 and 15 mm from the site of tendon transection respectively (1F). Insert show a close-up of the strands crossing at the level of the tendinous transection.

United States) from the construct. An imaging software program (ImageJ, National Institute of Health, Bethesda, Maryland, United States) was used to calculate the mean tendon cross-sectional area (CSA). The CSA was measured three times by a single investigator (C.C.) and the mean and standard deviation calculated to minimize possible error.

Experimental Groups

Tendons were then randomly assigned to one of three equally sized experimental groups using a random number generator (Research Randomizer, Lancaster, Pennsylvania, United States). All core surgical repairs used 2-0 USP polypropylene suture (Surgipro, Covidien Ltd., Dublin, Ireland). Group 1 was repaired with a core LL pattern, two-strand repair, as previously described by Tomlinson and Moore,⁴¹ starting by introducing the suture at the proximal transected tendon end. Transverse bites were taken 10 mm from the tendon ends (→Fig. 1C). Group 2 specimens were repaired with a novel four-strand repair utilizing a locking design with the transverse suture bite purchase 10 mm from the repair site with the second sequential transverse bite placed 15 mm from tenotomy site (→Fig. 1D). Finally, group 3 tendons were repaired with a novel six-strand repair with the first set, second and third transverse suture bites placed 5, 10 and 15 mm from the site of tendinous transection in each tendon

stump respectively (→Fig. 1E). For all groups, apposition of the tendon ends was achieved by manually tightening the suture, with equal tension that was subjectively assessed placed on each strand, prior to knot tying, to avoid development of bunching at the repair site. Upon pattern completion, the knot was tied using a square knot followed by three throws. Suture was cut 3 mm from the knot, buried within the repair site tendon repairs were completed with a new sterile suture pack, opened immediately prior to use. A single investigator (C.C.) performed all repairs under the direct supervision of a board-certified surgeon (D.J.D.) experienced with tendinous repairs in both clinical and research settings. Testing was completed in three separate sessions and the results collated. Saline (0.9% NaCl) solution was used to prevent against desiccation and keep specimens moist throughout the duration of tissue processing, repair and experimental testing.

Mechanical Testing

A materials testing machine (Instron Model 5944, Instron Inc., Norwood, Massachusetts, United States) connected to a modified bone fixation clamp (Bone clamp, SKU 1652-1, Sawbones, Vashon Island, Washington, United States) was used for experimental testing at room temperature (21°C) (→Fig. 2). The proximal part of the construct was secured by placing a 4 mm

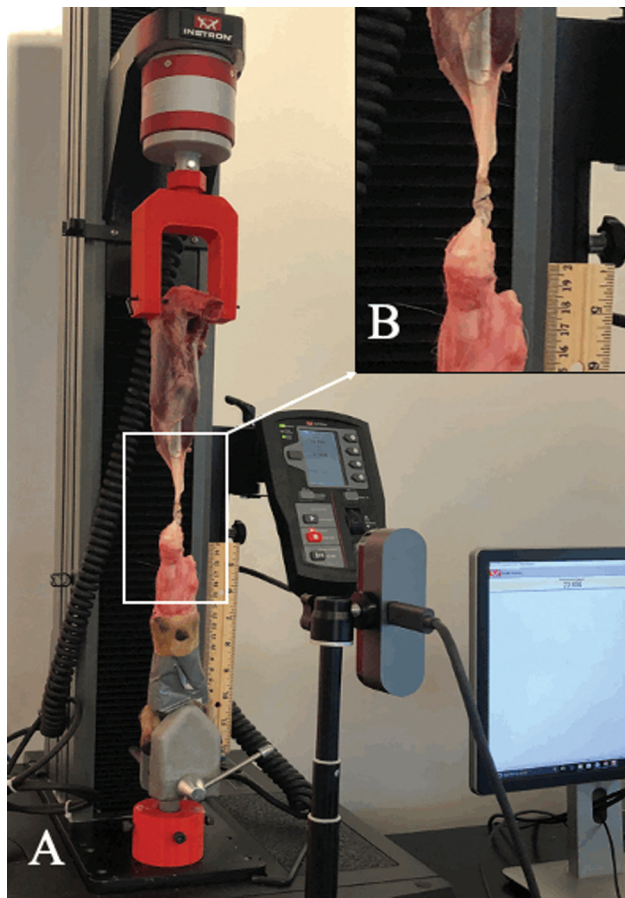


Fig. 2 Mechanical tensile testing apparatus with a gastrocnemius tendons (GT) specimen loaded within the custom testing jig (A). Magnified photographic image of a transected GT repaired with a two-strand locking pattern with a buried knot using 2-0 polypropylene (B).

diameter stainless steel bolt transversely through the previously drilled bone tunnel in the distal femur. To mimic the load application of a GT repair splinted in extension *in vivo*, repaired constructs were positioned with the calcaneus in a vertical orientation. A high-speed camera (Panasonic Lumix FZ200, Panasonic Corporation, Newark, New Jersey, United States) orientated perpendicularly with the tenorrhaphy site was used to film each separate test in high definition. A calibrated ruler was axially aligned with each specimen within the viewing window of the camera to allow for computed assessment of gap formation. The camera was placed at a measured distance 20 cm from the construct and each test captured at 50 frames/second.

Following initial calibration and pre-loading to 2N to establish a consistent resting length, a distraction rate of 20 mm/minute was applied to the repaired construct. Continuous distraction was continued until the point of catastrophic failure. Testing software (Bluehill 3, Instron Inc., Norwood, Massachusetts, United States) collected load (Newtons, N) and displacement (mm) data at a frequency of 100 Hz. Using a graphical plot to display the load-displacement curve within a custom software program (Matlab R2018b, Mathworks, Natick, Massachusetts, United States), the force required to achieve yield, peak and failure loads was identified. The point along the plot at which there was a non-

linear deformation of the construct was defined as the yield force. The maximum force applied during each test was defined as the peak force. The load at which failure of the construct or suture failed, or when there was a sharp decrease in the load displacement curve was defined as the failure force. Failure occurred by either suture breakage, suture pull through or direct failure of the tendinous tissue itself. Gap formation was assessed upon retrospective review of high-speed video footage, using the smallest distance between tendon ends to measure 1 and 3 mm gaps respectively. An imaging software program with a calibrated digital caliper (ImageJ, National Institute of Health, Bethesda, Maryland, United States) was used to measure development of gaps in sequentially assessed video frames to determine the exact time points and respective loads at which 1 and 3 mm gaps developed at the repair site. If constructs failed before formation of an identifiable gap, no gapping was reported. Mechanism of construct failure was assessed and confirmed upon review of the video data by a single investigator (C.C.). Collated data were recorded and reviewed using a commercially available spreadsheet program (Microsoft Excel, Microsoft Corp, Redmond, Washington, United States). A single investigator (D.J.D.) reviewed all collated data.

Statistical Analysis

A priori power analysis was performed following a pilot study. Based on the data and failure loads of a previous study,²⁴ a sample size calculation determined that 12 tendons per group would provide at least 90% power to detect a mean difference between groups of $30 \text{ N} \pm 5 \text{ N}$ at a 5% α error rate in independent measures. Pilot data were not included within the final statistical analysis. Data were assessed for parametric distribution with the Shapiro–Wilk test for normality. Differences in sample means were assessed by analysis of covariance. Pairwise comparisons of least square means were conducted with Scheffe or Dunn adjustment for multiple comparisons. Proportional distributions in occurrence of 3 mm gapping were compared between pattern groups with Fisher's exact test. All analyses were performed using statistical software (Statistical software, SAS v.9.4, SAS Institute Inc, Cary, North Carolina, United States). p -Values < 0.05 were considered statistically significant.

Results

All tendons were included within the final statistical analysis with no specimens rejected during collection or biomechanical testing. There was an equal distribution of right and left limbs ($p = 0.717$) among experimental groups. There was no difference in CSA between experimental groups (group 1: $0.10 \text{ cm}^2 \pm 0.02 \text{ cm}^2$; group 2: $0.09 \text{ cm}^2 \pm \text{standard deviation}$ [SD] 0.01 cm^2 ; group 3: $0.09 \text{ cm}^2 \pm \text{SD}$ 0.02 cm^2) ($p = 0.245$).

Load Data

A six-strand repair ($90.6 \pm 22.1 \text{ N}$) increased yield loads by $1.6\times$ and $3.7\times$ compared with four-strand ($55.0 \pm 8.9 \text{ N}$) and two-strand repairs ($24.7 \pm 8.3 \text{ N}$) respectively ($p < 0.001$) (**►Fig. 3**). Force application required to reach peak loads

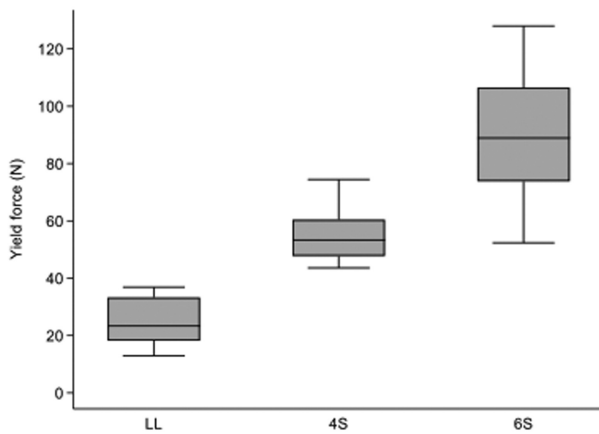


Fig. 3 Box and whisker plot depicting yield force of tenorrhaphies repaired with a two-strand locking repair (Group 1), a four-strand locking repair (Group 2) and six-strand locking repair (Group 3) using 2-0 USP polypropylene. Yield force differed significantly between groups ($p < 0.001$). Overall, results showed that the number of strands crossing the repair site increased, yield load also increased. Boxes represent interquartile range, the horizontal line in each box represents the median and whiskers extend to the highest and lowest values. LL, locking loop.

was 1.5 \times and 3.1 \times greater using a six-strand repair (111.4 ± 15.2 N) compared with four-strand (72.9 ± 7.8 N) and two-strand repairs (36.5 ± 6.0 N), respectively ($p < 0.001$). Failure loads followed a similar trend among experimental groups (\rightarrow Fig. 4). A six-strand repair (110.3 ± 15.1 N) resulted in a 1.5 \times and 3.1 \times increase in load to failure when compared with a four-strand repair (72.1 ± 8.2 N) and two-strand repair (36.1 ± 6.3 N) respectively ($p < 0.001$) (\rightarrow Table 1).

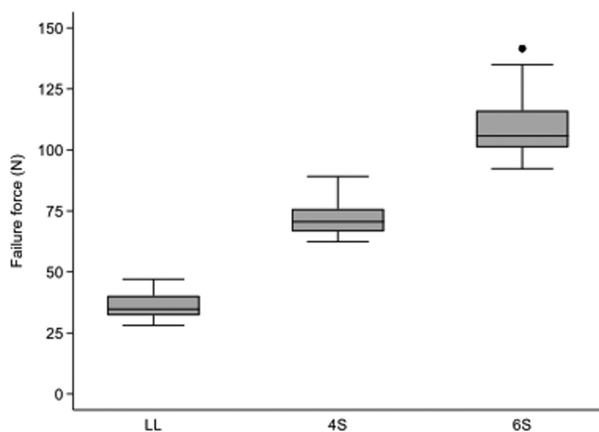


Fig. 4 Box and whisker plot showing failure loads of tenorrhaphies repaired with a two-strand (Group 1), four-strand (Group 2) and six-strand locking repair (Group 3) using 2-0 USP polypropylene. Failure force differed significantly between groups ($p < 0.001$). Results showed that as the number of strands crossing the repair site increased, failure load also increased proportionally. Boxes represent interquartile range, the horizontal line in each box represents the median and whiskers extend to the highest and lowest values. LL, locking loop.

Table 1 Mean \pm SD yield, peak and failure force in Newtons (N) for canine gastrocnemius tendons repaired with a two-strand (Group 1), four-strand (Group 2) and a six-strand locking repair (Group 3) using 2-0 USP polypropylene

Repair	Yield force (N)	Peak force (N)	Failure force (N)
2-Strand	24.7 ± 8.3^a	36.5 ± 6.0^a	36.1 ± 6.3^a
4-Strand	55.0 ± 8.9^b	72.9 ± 7.8^b	72.1 ± 8.2^b
6-Strand	90.6 ± 22.1^c	111.4 ± 15.2^c	110.3 ± 15.1^c

Abbreviation: SD, standard deviation.

Note: Different superscript letters denote significant differences between groups. There was a significant difference in yield ($p < 0.002$), peak ($p < 0.003$) and failure loads ($p < 0.003$) between experimental groups. Yield, peak and failure loads differed between all groups respectively ($p < 0.001$).

Development of Gap Formation between Tendon Ends

Results differed among groups regarding force application required to create a 1 mm gap between tendon ends ($p < 0.001$) with all specimens (36/36) demonstrating development of a 1 mm gap at the repair site. A six-strand repair ($101. \pm 20.1$ N) required significantly greater load to cause a 1 mm gap compared with four-strand (65.0 ± 10.2 N) and two-strand repairs (29.9 ± 5.6 N) respectively. When comparing the occurrence of 3 mm gap at the repair site, six-strand repairs had reduced occurrence of 3 mm gapping ($p < 0.001$) while requiring significantly greater force to cause 3 mm gap formation ($p < 0.001$). Of the six-strand repairs, 25% (3/12) of repaired constructs developed a 3 mm gap under tension (122.5 ± 26.6 N). For four-strand repair constructs, 66.7% (8/12) of constructs developed a 3 mm gap under tension (67.7 ± 13.0 N). Of two-strand repair constructs, 100% (12/12) of repaired constructs developed evidence of 3 mm gapping under tension (32.6 ± 6.9 N).

Mode of Construct Failure

Mechanisms of construct failure did not differ between experimental groups ($p = 0.869$). Failure mechanism among all construct repairs (36/36, 100%) occurred by pull-through of the suture through the tendon substance.

Discussion

This study investigated the biomechanical properties and occurrence of gap formation when using a novel tenorrhaphy design by increasing the number of strands crossing the repair, level of suture bites from the transection site and depth of suture penetrance in a canine tendon laceration model. In agreement with our hypothesis, results of this study show that increasing the number of suture strands crossing the repair and increasing the level of suture purchase from the transection site are positively correlated with repair site strength while reducing the occurrence of gap formation in a cadaveric gastrocnemius model. While multiple sources in both the veterinary and human literature report variations of multi-strand repairs,^{19,25-39} the authors of the current study sought to modify existing patterns to

develop a novel six-strand locking repair technique that would offer technical benefits similar to the LL pattern such as ease of suture placement and reduction in repair site bunching.^{40,41} Within the human literature, a review by Rawson and colleagues³⁸ reported several suture repair techniques. Two of these patterns, a modified locking Kessler and a four-strand double modified Kessler, were similar to two- and four-strand repair utilized in this study, with the exception of a buried knot, though suture depth and distance from the transection site were not reported. Locking patterns, such as those presented here, offer the surgeon the benefit of patterns acting as a secure anchor as the suture grasps, tightens and consequently holds tendon fibers.³⁸ Given the known benefit of increasing the number of strands traversing the repair site, this study developed and tested a pattern which increased the number of strands crossing the repair site of a commonly used LL pattern by taking subsequent bites further away from the tendinous transection site and increasing the depth of suture penetrance into the core of the tendinous substance. Moores and colleagues²⁴ described a four-strand repair that consisted of 2 separate LL patterns. The present study sought to simplify placement and reduce repair site bulk by removing the requirement for multiple knots. In an equine tendon model, Easley and colleagues⁴² used a similar locking repair to the present study. However, this repair required smaller locking suture purchase points with bites placed at the same distance from the tenotomy site, resulting in a repair that the aforementioned study authors described as difficult to re-create with unpredictable modes of failure due to smaller portions of tendon fibrils being grasped by the suture. The pattern utilized in the present study was subjectively simple to place while grasping a larger portion of tendon fibrils. However, these hypotheses should be interpreted with caution until a direct study comparing these two patterns is performed.

Following review and evaluation of human tenorrhaphy patterns following distal extremity tendon repair of the human hand, it has been demonstrated that overall repair site strength can be improved by increasing the number of strands crossing the repair, which resulted in the development and modification of several six-strand patterns.^{19,28,31–33} In a study by Al-Qattan and Al-Turaiki,³³ using an experimental ovine model, they found the failure force of a two-, four- and six-strand repair technique was positively correlated to increasing the number of strands crossing the repair site (48.0 N, 73.1 N and 93.3 N respectively). Similarly, our study found a significant improvement in repair site strength by increasing the number of strands crossing the repair. Increasing the number of suture strands and points of suture purchase in a six-strand repair was superior to a four- and two-strand repair with failure forces of 110 N, 72 N and 36 N, respectively. Al-Qattan and Al-Turaiki further investigated clinical outcomes *in vivo* in human clinical trials following use of three 'figure of eight' sutures for profundus digitorum tendon repairs, examining 50 fingers in 45 patients.³³ Outcome of this novel six-strand repair technique resulted in a 2% rupture rate (1/50), with the remaining 98% of repairs having a good to excellent clinical outcome. While our study could not assess clinical success given its *ex*

vivo nature, the authors hope that similar improvements will be seen following clinical evaluation if a six-stranded locking pattern is utilized for canine core tendon repairs. These hypotheses should be interpreted with caution until focused *in vivo* assessment in canine patients is performed.

Within the veterinary literature, the 3LP has been shown to be biomechanically superior, compared with LL patterns.³⁹ These findings likely relate to strands crossing the repair using a 3LP pattern compared with a two-strand repair using a LL suture pattern.²⁵ While not directly compared in the present study, the results presented here suggest that a suture pattern that increases the number of strands and points of suture purchase and suture depth into the tendon substance can result in similar biomechanical properties compared with previously reported 3LP force data.^{18,24,25,27,50} In an equine tendon laceration model,⁴² Easley and colleagues found that both double and triple LL repairs achieved similar loads to failure as a 3LP and the triple LL was stronger than both the double LL and LL suture alone. Everett and colleagues⁵⁰ reported similar findings in an equine tendon laceration model when comparing a 3LP to a six-strand suture, with the latter shown to improve strength and resistance to suture pull-through. Moores and colleagues²⁴ compared 3LP to two LL patterns and found loads to failure to be similar between experimental groups (3LP; 60.1 ± 3.9 N, 2 x LL; 61.0 ± 3.9 N). This is similar to maximal values recorded in our study using a four-strand repair (72.9 N). Wilson and colleagues²⁵ also found load to failure of a 3LP repair on a GT to be less than that reported for our six-strand pattern modifications at 80.9 N. Maximum loads tolerated by a six-strand modification in the present study were found to be 111.4 N, suggesting that this pattern may be biomechanically superior to a 3LP alone when compared indirectly to prior studies. While a 3LP group was purposefully not included in the current study, in an effort to allow the effect of a two-, four- and six-strand locking repair to be directly compared, the authors acknowledge a direct comparison study using similarly sized suture material would be necessary to further elucidate these findings.

Decreasing the occurrence of gap formation plays an important role in reducing the risk of repair failures postoperatively.^{14–18} Our study showed that increasing the number of strands crossing the transection site increases the force required to cause occurrence of 1 and 3 mm gaps, respectively, thus supporting increasing the number of strands that cross the repair site resulting in a biomechanically superior repair, as reported in prior studies.^{19,28,31–34,44} In a biomechanical study by Dunlap and colleagues,²² a non-locking pre-manufactured loop suture technique was compared with a 3LP core suture. In that study they found the force to achieve a 1 mm gap to be 65 N in the 3LP group using 2–0 polypropylene suture. In the present study, a six-strand locking repair using 2–0 polypropylene suture required 101.3 N to cause a 1 mm gap, significantly greater than that reported for 3LP core repairs alone. In that same study,²² investigators demonstrated that 127.2 N was required to cause development of a 3 mm gap in the 3LP repair group alone. Similarly, our study found that mean loads required to create a 3 mm gap with a six-strand locking repair was

122.5 N. In the present study, however, it should be noted that only 25% (3/12) of the six-strand locking repairs developed a 3 mm gap under maximal tension prior to catastrophic failure. Based on our results, we hypothesize that a six-strand locking repair may translate *in vivo* to improve tendinous healing while decreasing the risk of repair failures and decreasing the occurrence 3 mm gap formation. These findings must be interpreted with caution as further studies are necessary using an *in vivo* model to assess if a six-strand locking repair results in improved healing with decreased rates of postoperative failure compared with 3LP repairs alone.

In prior studies by Easley and colleagues,⁴² and Moores and colleagues,²⁴ investigators found that repeated triple LL patterns were associated with longer suturing times compared with a 3LP pattern. Although subjectively assessed, we found six-strand locking repairs relatively easy to place while achieving uniform apposition along the whole length of the repair site. In agreement with a study by Moores and colleagues,²⁴ in which the majority of double LL repairs resulted in failure by suture pull through, 100% of the present study's repaired constructs failed by suture pull through. This may be explained by greater force distribution and load sharing between the suture and tendinous substance. We postulate that additional suture purchase afforded by multi-strand, multi-level repairs, result in the tendon-suture interface being weaker than the suture material itself leading to differences in failure mode seen between studies. When tensioned, a locking pattern results in tendon fibres and collagen bundles being locked or grasped between suture loops.³⁸ Larger tendons result in the ability of suture to grasp and interact with a greater number of collagen fibrils, resulting in a stronger repair, and ultimately, the suture material itself is more likely to be the weakest point of the repair.^{39,42,51} The smaller size of the canine GT and resultant smaller number of available fibrils available for suture purchase, result in a weaker tendon-suture interface, and can likely explain why two-stranded locking repairs failed by suture pull-through, opposed to similar repairs resulting in failure by suture breakage in larger equine tendons.^{42,51}

In the present study, we demonstrated that the strength of a novel locking pattern can be improved proportionally by pattern modification to increase the number of suture strands traversing the surgical repair, increasing the level of locking suture bites and depth of penetrance of the core suture. Currently in canine patients, immobilization is recommended postoperatively to decrease the risk of repair failures.^{52,53} However, methods such as external coaptation have been associated with a high degree of postoperative morbidity.⁵⁴ In contrast, in human tenorrhaphy, current recommendations following surgical repair are immediate active controlled rehabilitation.²⁸ We hypothesize that the degree of postoperative failures and reliance on postoperative immobilization may be reduced with the use of a multi-strand, multi-level locking repair when used in addition other methods of support and augment, such as placement of an epitendinous suture.^{40,55} However, this was not evaluated in this *in vitro* study and warrants further investigation.

A limitation of the present study was our inability to assess the effect of six-strand locking repairs on glide function and the implications of knot burial within the repair site. In human zone II tendon repair following distal extremity laceration, glide resistance is an important consideration improving tendinous excursion and may decrease the risk of tendon re-rupture.⁵⁶ In dogs following tendinous repair, restoration of tensile strength and minimization of gap formation between tendon ends is often prioritized over glide function, as it is thought to be of less clinical significance compared with human repairs.^{2,4} Numerous repair methods utilized in veterinary medicine use externally placed knotting techniques,^{18,22,24,25,27,39,40} which have been shown to adversely affect glide resistance and increase the overall force required for active motion.^{38,56} Glide resistance has been identified as an important factor in decreasing the risk of tendinous re-rupture during this period, especially in regard to passive rehabilitation.⁵⁶ Identification of novel tenorrhaphy techniques that increase repair site strength, reduce the occurrence of gap formation while potentially allowing for earlier controlled mobilization, are necessary and of critical importance in further validating results gathered from such studies. This may lead to more rigorous stipulations in techniques for future *ex vivo* studies. Due to our studies inherent *ex vivo* design, important factors such as the effect of suture interaction and knot placement on tendinous blood supply and intrinsic healing were not assessed. Additionally, this study did not employ a cyclical loading protocol, which is likely more representative of clinical conditions appreciable in the live animal.⁵⁷ Instead, this study purposefully employed a single load to failure testing protocol to represent acute force application to the musculotendinous unit in the immediate postoperative period and allow direct comparison between prior studies using similar methodologies.^{24,25,43}

In conclusion, use of a novel six-strand locking repair resulted in a significantly stronger repair in a canine tendinous laceration model. Increasing the number of suture strands crossing the repair while increasing the points of suture purchase and depth of suture penetrance from the transection site was shown to be positively correlated with repair site strength while significantly reducing the occurrence of gap formation between tendon ends. Additional studies are recommended *in vivo*, to determine effect on clinical function, tendinous healing and glide resistance prior to clinical use.

Authors' Contributions

D.J.D. conceptualized and designed the study. All authors acquired, analyzed and interpreted the data. They drafted, revised and approved the submitted manuscript and are publically accountable for relevant content.

Conflict of Interest

None declared.

Acknowledgments

The authors would like to thank Medtronic, Inc., Mansfield, Massachusetts, USA, for providing the surgical suture.

References

- O'Keefe RJ, Jacobs JJ, Chu CR, Einhorn TA. Orthopaedic Basic Science: Foundations of Clinical Practice. 4th edition. Rosemont: American Academy of Orthopaedic Surgeons; 2018
- Muscle and tendon disorders. In: Tobias K, Johnston S, eds. Veterinary Surgery: Small Animal. St. Louis: Elsevier; 2018: 1319–1322
- Johnson J, Austin C, Breur G. Incidence of canine appendicular musculoskeletal disorders in 16 veterinary teaching hospitals from 1980 through 1989. *Vet Comp Orthop Traumatol* 1994;07(02):56–69
- Slatter DH. Textbook of Small Animal Surgery. 3rd edition. Vol. 2; Philadelphia: Saunders; 2003
- Corr SA, Draffan D, Kulendra E, Carmichael S, Brodbelt D. Retrospective study of Achilles mechanism disruption in 45 dogs. *Vet Rec* 2010;167(11):407–411
- Katayama M. Augmented repair of an Achilles tendon rupture using the flexor digitorum lateralis tendon in a toy poodle. *Vet Surg* 2016;45(08):1083–1086
- Morton MA, Whitelock RG, Innes JF. Mechanical testing of a synthetic canine gastrocnemius tendon implant. *Vet Surg* 2015;44(05):596–602
- Diserens KA, Venzin C. Chronic Achilles tendon rupture augmented by transposition of the fibularis brevis and fibularis longus muscles. *Schweiz Arch Tierheilkd* 2015;157(09):519–524
- Morton MA, Thomson DG, Rayward RM, Jiménez-Peláez M, Whitelock RG. Repair of chronic rupture of the insertion of the gastrocnemius tendon in the dog using a polyethylene terephthalate implant. Early clinical experience and outcome. *Vet Comp Orthop Traumatol* 2015;28(04):282–287
- Worth AJ, Danielsson F, Bray JP, Burbidge HM, Bruce WJ. Ability to work and owner satisfaction following surgical repair of common calcanean tendon injuries in working dogs in New Zealand. *N Z Vet J* 2004;52(03):109–116
- Evans H, de Lahunta A. Miller's Anatomy of the Dog. 4th edition. St. Louis: Elsevier Saunders; 2013
- King M, Jerram R. Achilles tendon rupture in dogs. *Compend Contin Educ Pract Vet* 2003;25:613–620
- Meutstege FJ. The classification of canine Achilles' tendon lesions. *Vet Comp Orthop Traumatol* 1993;06(01):53–55
- Gelberman RH, Boyer MI, Brodt MD, Winters SC, Silva MJ. The effect of gap formation at the repair site on the strength and excursion of intrasynovial flexor tendons. An experimental study on the early stages of tendon-healing in dogs. *J Bone Joint Surg Am* 1999;81(07):975–982
- Lister GD, Kleinert HE, Kutz JE, Atasoy E. Primary flexor tendon repair followed by immediate controlled mobilization. *J Hand Surg Am* 1977;2(06):441–451
- Kessler I, Nissim F. Primary repair without immobilization of flexor tendon division within the digital sheath. An experimental and clinical study. *Acta Orthop Scand* 1969;40(05):587–601
- Möller M, Movin T, Granhed H, Lind K, Faxén E, Karlsson J. Acute rupture of tendon Achilles. A prospective randomised study of comparison between surgical and non-surgical treatment. *J Bone Joint Surg Br* 2001;83(06):843–848
- Gall TT, Santoni BG, Egger EL, Puttlitz CM, Rooney MB. In vitro biomechanical comparison of polypropylene mesh, modified three-loop pulley suture pattern, and a combination for repair of distal canine Achilles' tendon injuries. *Vet Surg* 2009;38(07): 845–851
- Strickland JW. Development of flexor tendon surgery: twenty-five years of progress. *J Hand Surg Am* 2000;25(02):214–235
- Dona E, Turner AWL, Gianoutsos MP, Walsh WR. Biomechanical properties of four circumferential flexor tendon suture techniques. *J Hand Surg Am* 2003;28(05):824–831
- Guzzini M, Lanzetti RM, Proietti L, et al. Interlocking horizontal mattress suture versus Kakiuchi technique in repair of Achilles tendon rupture: a biomechanical study. *J Orthop Traumatol* 2017; 18(03):251–257
- Dunlap AE, Kim SE, McNicholas WT Jr. Biomechanical evaluation of a non-locking pre-manufactured loop suture technique compared to a three-loop pulley suture in a canine calcaneus tendon avulsion model. *Vet Comp Orthop Traumatol* 2016;29(02):131–135
- Cervi M, Brebner N, Liptak J. Short- and long-term outcomes of primary Achilles tendon repair in cats: 21 cases. *Vet Comp Orthop Traumatol* 2010;23(05):348–353
- Moore AP, Owen MR, Tarlton JF. The three-loop pulley suture versus two locking-loop sutures for the repair of canine Achilles tendons. *Vet Surg* 2004;33(02):131–137
- Wilson L, Banks T, Luckman P, Smith B. Biomechanical evaluation of double Krackow sutures versus the three-loop pulley suture in a canine gastrocnemius tendon avulsion model. *Aust Vet J* 2014; 92(11):427–432
- Moore AP, Comerford EJ, Tarlton JF, Owen MR. Biomechanical and clinical evaluation of a modified 3-loop pulley suture pattern for reattachment of canine tendons to bone. *Vet Surg* 2004;33(04): 391–397
- Savage R, Risitano G. Flexor tendon repair using a "six strand" method of repair and early active mobilization. *J Hand Surg Am* 1989;14B:396–399
- Wong YR, Tay SC. A biomechanical study of a novel asymmetric 6-strand flexor tendon repair using porcine tendons. *Hand (N Y)* 2018;13(01):50–55
- Wichelhaus A, Beyersdoerfer ST, Vollmar B, Mittlmeier T, Gierer P. Four-strand core suture improves flexor tendon repair compared to two-strand technique in a rabbit model. *BioMed Res Int* 2016; 2016:4063137
- Silfverskiöld KL, Andersson CH. Two new methods of tendon repair: an in vitro evaluation of tensile strength and gap formation. *J Hand Surg Am* 1993;18(01):58–65
- Shaieb MD, Singer DI. Tensile strengths of various suture techniques. *J Hand Surg [Br]* 1997;22(06):764–767
- Al-Qattan MM, Al-Turaiki TM. Flexor tendon repair in zone 2 using a six-strand 'figure of eight' suture. *J Hand Surg Eur Vol* 2009;34(03):322–328
- Hatanaka H, Manske PR. Effect of suture size on locking and grasping flexor tendon repair techniques. *Clin Orthop Relat Res* 2000;(375):267–274
- Labana N, Messer T, Lautenschlager E, Nagda S, Nagle D. A biomechanical analysis of the modified Tsuge suture technique for repair of flexor tendon lacerations. *J Hand Surg [Br]* 2001;26(04):297–300
- Barrie KA, Tomak SL, Cholewicki J, Wolfe SW. The role of multiple strands and locking sutures on gap formation of flexor tendon repairs during cyclical loading. *J Hand Surg Am* 2000;25(04): 714–720
- Viinikainen A, Göransson H, Huovinen K, Kellomäki M, Rokkanen P. A comparative analysis of the biomechanical behaviour of five flexor tendon core sutures. *J Hand Surg [Br]* 2004;29(06):536–543
- Rawson S, Cartmell S, Wong J. Suture techniques for tendon repair; a comparative review. *Muscles Ligaments Tendons J* 2013;3(03):220–228
- Berg RJ, Egger EL. In vitro comparison of the three loop pulley and locking loop suture patterns for repair of canine weightbearing tendons and collateral ligaments. *Vet Surg* 1986;15(01):107–110
- Putterman AB, Duffy DJ, Kersh ME, Rahman H, Moore GE. Effect of a continuous epitendinous suture as adjunct to three-loop pulley and locking-loop patterns for flexor tendon repair in a canine model. *Vet Surg* 2019;48(07):1229–1236
- Tomlinson J, Moore R. Locking loop tendon suture use in repair of five calcanean tendons. *Vet Surg* 1982;11(03):105–109

- 42 Easley KJ, Stashak TS, Smith FW, Van Slyke G. Mechanical properties of four suture patterns for transected equine tendon repair. *Vet Surg* 1990;19(02):102–106
- 43 Gelberman RH, Manske PR, Akeson WH, Woo SL-Y, Lundborg G, Amiel D. Flexor tendon repair. *J Orthop Res* 1986;4(01):119–128
- 44 Winters SC, Gelberman RH, Woo SLY, Chan SS, Grewal R, Seiler JG III. The effects of multiple-strand suture methods on the strength and excursion of repaired intrasynovial flexor tendons: a biomechanical study in dogs. *J Hand Surg Am* 1998;23(01):97–104
- 45 Taras JS, Raphael JS, Marczyk SC, Bauerle WB. Evaluation of suture caliber in flexor tendon repair. *J Hand Surg Am* 2001;26(06):1100–1104
- 46 Duffy DJ, Curcillo CJ, Chang YJ, Moore GE. Effect of suture caliber on the tensile strength of tenorrhaphies in cadaveric canine tendons. *Am J Vet Res* 2020;81(09):714–719
- 47 Tang JB, Zhang Y, Cao Y, Xie RG. Core suture purchase affects strength of tendon repairs. *J Hand Surg Am* 2005;30(06):1262–1266
- 48 Duffy DJ, Cocca CJ, Kersh ME, Kim W, Moore GE. Effect of bite distance of an epitendinous suture from the repair site on the tensile strength of canine tendon constructs. *Am J Vet Res* 2019;80(11):1034–1042
- 49 Duffy DJ, Chang YJ, Gaffney LS, Fisher MB, Moore GE. Effect of bite depth of an epitendinous suture on the biomechanical strength of repaired canine flexor tendons. *Am J Vet Res* 2019;80(11):1043–1049
- 50 Everett E, Barrett JG, Morelli J, DeVita R. Biomechanical testing of a novel suture pattern for repair of equine tendon lacerations. *Vet Surg* 2012;41(02):278–285
- 51 Jann HW, Stein LE, Good JK. Strength characteristics and failure modes of locking-loop and three-loop pulley suture patterns in equine tendons. *Vet Surg* 1990;19(01):28–33
- 52 Nielsen C, Pluhar GE. Outcome following surgical repair of Achilles tendon rupture and comparison between postoperative tibiotarsal immobilization methods in dogs: 28 cases (1997–2004). *Vet Comp Orthop Traumatol* 2006;19(04):246–249
- 53 Guerin S, Burbidge H, Firth E, Fox S. Achilles tenorrhaphy in five dogs: a modified surgical technique and evaluation of a cranial half cast. *Vet Comp Orthop Traumatol* 1998;11:205–210
- 54 Meeson RL, Davidson C, Arthurs GI. Soft-tissue injuries associated with cast application for distal limb orthopaedic conditions. A retrospective study of sixty dogs and cats. *Vet Comp Orthop Traumatol* 2011;24(02):126–131
- 55 Cocca CJ, Duffy DJ, Kersh ME, Kim W, Groenewold A, Moore GE. Biomechanical comparison of three epitendinous suture patterns as adjuncts to a core locking loop suture for repair of canine flexor tendon injuries. *Vet Surg* 2019;48(07):1245–1252
- 56 Zhao C, Amadio PC, Zobitz ME, An KN. Gliding characteristics of tendon repair in canine flexor digitorum profundus tendons. *J Orthop Res* 2001;19(04):580–586
- 57 Pruitt DL, Manske PR, Fink B. Cyclic stress analysis of flexor tendon repair. *J Hand Surg Am* 1991;16(04):701–707