Effect of epitendinous suture caliber on the tensile strength of repaired canine flexor tendons

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OBJECTIVE
To determine the effect of epitendinous suture (ES) caliber on the tensile strength of flexor tendon repairs in cadaveric specimens from dogs.

SAMPLE
60 cadaveric superficial digital flexor tendons (SDFTs) from 30 skeletally mature dogs.

PROCEDURES
Specimens were randomly assigned to 5 suture caliber groups (n = 12 SDFTs/group). After sharp transection, SDFTs were repaired by placement of a simple continuous circumferential ES created with size-0, 2-0, 3-0, 4-0, or 5-0 polypropylene suture. Constructs were preloaded to 2 N and load tested to failure. Loads at yield, peak, and failure and mode of failure were compared among groups by statistical methods.

RESULTS
Yield, peak, and failure loads for SDFT repair constructs were positively correlated with ES caliber and did not differ between the size-0 and 2-0 groups on pairwise comparisons. Yield load was significantly greater for size-0, 2-0, and 3-0 groups than for the 4-0 and 5-0 groups. Peak and failure loads were significantly greater for the size-0 and 2-0 groups than for the remaining groups. Most size-0 (12/12), 2-0 (12/12), and 3-0 (10/12) group constructs failed because of ES pull-through; several constructs in the 4-0 group (5/12) and most in the 5-0 group (11/12) failed because of ES breakage.

CONCLUSIONS AND CLINICAL RELEVANCE
Results suggested size-0 and 2-0 sutures should be considered when placing an ES for flexor tendon repairs in dogs. However, in vivo studies are needed determine the effects of increasing ES caliber on clinical outcomes for dogs undergoing these procedures. (*Am J Vet Res* 2021;82:510–515)

Tendon repair is performed primarily by surgical intervention to achieve direct contact healing between tendon ends.[1,2] The principles of tendon repair to achieve satisfactory postoperative function include creation of a strong enough construct to transmit force associated with active limb use while preventing formation of a gap > 3 mm at the repair site.[2–4]

Tendon repairs that incorporate an ES and a core suture are commonly applied during surgery at the distal aspects of the extremities in people.[5] These repairs provide increased strength, compared with core suture placement alone, and decrease the occurrence of gap formation between tendon ends.[6,7] Regarding core suture placement, factors shown to be important surgical considerations include the number of suture knots,[8,9] number of suture strands,[8,10] suture materials used,[11] caliber of the suture,[12] and depth of suture purchase,[13] all of which influence the strength of the suture construct. In veterinary medicine, the 3-loop pulley and LL techniques are commonly used in clinical settings, with the 3-loop pulley shown to have greater strength on the basis of loads sustained prior to construct failure.[14,15]

Placement of a circumferential ES in a simple continuous,[16] Silfverskiöld cross-stitch,[17] or interlocking horizontal mattress[18] pattern is widely performed to augment core suture placement in patients requiring tendon repair. In an ex vivo study of SDFTs from canine cadavers, Putterman et al[19] found that circumferential ES placement with a continuous technique in addition to a core 3-loop pulley or LL suture significantly increased the ultimate tensile strength at the repair site by 133% and 151%, respectively, compared with core suture use alone.[19] Cocca et al[20] demonstrated that ES placement in addition to core suture placement was more relevant than the type of ES pattern used to improve the biomechanical properties of SDFT repair in another ex vivo study with cadaveric specimens from dogs. Other recent investigations with cadaveric samples from dogs have shown that increasing the distance of ES bites from the transec-

ABBREVIATIONS

ES	Epitendinous suture
LL	Locking-loop
SDFT	Superficial digital flexor tendon

tion site[21] and increasing the depth of ES bites toward the center of the tendon substance (allowing a greater degree of suture-tissue interaction)[22] each result in improved construct strength. Tested experimental constructs of tendon repair predominantly fail by means of suture pull-through or failure of the suture itself. The occurrence of suture pull-through is largely dependent on the inherent strength of tissues and their structural integrity, which is commonly diminished in dogs affected by chronic degenerative changes.[12,23,24] Suture breakage, however, is largely influenced by the caliber, type, and material properties of the suture used for tendon repair.[12,17,23] Studies of specimens from human[12] and canine cadavers[25] revealed that increasing the caliber of core sutures used for tendon repair is significantly associated with increasing overall strength of the repair.[12] Owing to the variations in the size and body weight of canine patients, selection of the most appropriate suture caliber and technique for tendon repair can be technically challenging. To the authors knowledge, the effect of ES caliber on construct strength following flexor tendon repair has not been evaluated for dogs, and this warrants careful biomechanical assessment prior to use for canine patients in a clinical setting.

The purpose of the study reported here was to evaluate the effect of suture caliber (US Pharmacopeia size) used for placement of an ES in a simple continuous circumferential pattern, without the addition of a core suture, on the tensile strength of flexor tendon repairs in cadaveric specimens from dogs. Our hypothesis was that tendon repair constructs created with larger-caliber suture would have greater tensile strength than those created with smaller-caliber suture.

Materials and Methods

Specimen collection

Cadaveric forelimbs were harvested from 30 medium-sized to large-breed dogs > 1 year of age with body weights of 28 to 34 kg. Specimens were obtained immediately after euthanasia by IV administration of a commercial euthanasia solution[a] for reasons unrelated to the present study. An institutional animal care and use committee approval was not required by our academic institution owing to the secondary use of cadaveric tissues for the study. Cadavers were excluded from the study if examination revealed evidence of gross orthopedic abnormalities. History and demographic information were unavailable for many dogs, and variables such as age, sex, and weight of individual animals were not recorded.

Specimen preparation

The SDFTs of both forelimbs from each dog were individually isolated into musculotendinous units from the origin on the medial humeral condyle to the entheses on the distal phalanges as previously described.[21,22] Following dissection and tendon labeling at room temperature (21°C), specimens were individually stored in saline (0.9% NaCl) solution–soaked gauze sponges at –20°C in 1-gallon-capacity impervious bags[b] until the day of testing. Tissues were dissected, and specimens were frozen ≤ 6 hours after euthanasia. Prior to the day of specimen testing, tendons were thawed at room temperature (21°C) for 10 hours by means of a previously validated technique in a thermostatically controlled environment.[26] On the day of testing, tendons were transected at a measured distance of 2.5 cm distal to the musculotendinous junction with a No. 10 scalpel blade. All tenotomies were performed by 1 board-certified veterinary surgeon (DJD). An impervious polypropylene cutting board was used as a base to achieve a perpendicular cut across the tendon. Cuts were made at a 90° angle to the direction of longitudinal collagen fibers. After the tenotomy, the proximal tendon stump was photographed[c] alongside a calibrated surgical ruler[d] with 1-mm markings for later measurement of tendon cross-sectional area with an imaging software program.[e]

The specimens were randomly assigned to 5 suture caliber groups (12/group; size-0, 2-0, 3-0, 4-0, or 5-0) by use of an electronic random number generator.[f] Specimens from the right and left forelimbs from the same dog were controlled from being assigned to the same group. The ESs for constructs in each group were created with polypropylene suture[g] from 1 manufacturer. A straight needle was used to aid in accurate suture passage. Transected tendon ends were repaired by placement of a simple continuous circumferential ES with bites placed 2 to 3 mm apart at a depth of 2 mm and a distance of 8 mm from the tenotomy site in a proximodistal direction for each segment of a given tendon. Optical magnification[h] was used, with the tendon placed next to a calibrated surgical ruler[d] to allow for consistency among the repairs. At the time of surgical repair, two 22-gauge needles[i] were used for holding tendon ends in the correct orientation and close apposition to mimic tendon repairs as they would be performed at our tertiary referral hospital. When the ES pattern was complete, the surgeon ensured that the tendon ends were apposed, then placed a single square knot followed by 3 additional throws and cut the suture end to a length of 3 mm. Absence of tissue bunching at the repair site was achieved while ensuring that a subjectively equal amount of tension was placed on the sutures prior to knot tying. To allow assessment of only the ES, no core sutures were placed. One investigator (DJD) experienced with tendon repairs performed all repairs and was aided throughout by a trained surgical assistant (Y-JC).

Biomechanical testing

A materials testing machine[j] was used for biomechanical testing with a custom testing jig modified by one of the authors (DJD). The testing jig was used to secure the humeral bone segment of the construct with a 4.0-mm-diameter metal rod placed in a mediolateral direction through the supratrochlear foramen of the humeral bone segment and attached to a 500-N load cell that was mounted on the testing machine. The paw of each forelimb was securely mounted with a manual-compression bone clamp.[k] A high-definition digital video camera[l] was placed 20 cm from the palmar aspect of the construct, and a ruler marked with 1-mm increments was placed adjacent to the construct, near the repair site and in the camera field of view for each recorded test.

The musculotendinous unit of each construct was aligned axially for load testing. To establish consistency of initial test conditions for all constructs, musculotendinous units were preloaded to 2 N, and all elongation measurements were zeroed prior to proceeding with each test. When a load of 2 N was reached, automated software allowed for simultaneous recording of the video data, and this recording was synchronized with the load data. Constructs were then distracted at a rate of 20 mm/min until catastrophic failure occurred while the test was recorded with the digital camera.

Following biomechanical testing, load-displacement curves were generated with software[m] for evaluation of yield, peak, and failure loads. Yield load indicated the limit of elastic behavior and the beginning of plastic deformation and was defined as the load at which there was a change in the linear region of the load-displacement curve. Peak load was defined as the greatest measured load on the construct during testing prior to catastrophic failure. Finally, failure load was defined as the load at which the suture broke or pulled through the tendon tissue or tissue rupture occurred. To aid in accurate identification of the described load data points, a software program[n] was customized for use in the study. The mechanism of construct failure was documented and confirmed by review of the video recordings by 1 study investigator (Y-JC). All load data were independently reviewed by another study investigator (DJD).

Statistical analysis

A power analysis was performed and established that a sample size of 11 constructs/group would provide ≥ 80% power to detect a mean ± SD difference of 25 ± 10 N among groups with a 95% confidence level. A Shapiro-Wilk test confirmed parametric distribution for continuous variables in the data set, and these results were reported as mean ± SD. A linear mixed-effects model was used to compare yield, peak, and failure loads among groups while controlling for effects of the left and right forelimbs and cadaver. Least squares means were used for pairwise comparisons with Bonferroni adjustments for multiple comparisons. The Fisher exact test was used to compare proportional distributions of failure mode among groups. All statistical analyses were performed with a commercially available software program.[o] Values of $P < 0.05$ were considered significant.

Results

All SDFTs were successfully transected, sutured, and biomechanically tested; none were excluded from statistical analysis. There was no significant ($P = 0.856$) difference in the distribution of left versus right forelimbs across the suture caliber groups. The mean ± SD tendon cross-sectional area for all specimens was 0.23 ± 0.04 cm^2, and area measurements did not differ among groups ($P = 0.390$).

Biomechanical test data

Mean ± SD yield, peak, and failure loads for suture caliber groups are provided **(Table 1)**. The yield load differed significantly ($P < 0.001$) among groups. Pairwise comparisons indicated no difference in the yield load between the size-0 and 2-0 ($P = 0.645$) or 3-0 ($P = 0.054$) groups or between the 2-0 and 3-0 ($P = 0.655$) groups. However, the load at yield was significantly ($P < 0.001$ for all comparisons) greater for the size-0, 2-0, and 3-0 groups, compared with the 4-0 and 5-0 groups.

Peak load and load at failure each differed significantly ($P < 0.001$) among groups. No difference was detected in peak load between the size-0 and 2-0 groups ($P = 1.00$). However, the size-0 and 2-0 groups each had significantly higher peak loads, compared with the 3-0 ($P = 0.002$ and $P = 0.003$, respectively), 4-0 ($P < 0.001$), and 5-0 ($P < 0.001$) groups (Table 1). Similar findings were observed for the load at failure, with no difference between the size-0 and 2-0 groups ($P = 1.00$). However, the load at failure was significantly greater for the size-0 and 2-0 groups, compared with the 3-0 ($P < 0.002$), 4-0 ($P < 0.001$), and 5-0 ($P < 0.001$) groups.

Modes of construct failure

All repaired constructs failed as a result of ES pull-through or ES breakage. Mechanisms of failure were significantly ($P < 0.001$) different among groups, with suture pull-through being the most common mode of failure in the size-0 (12/12 constructs), 2-0 (12/12), and 3-0 (10/12) groups. In contrast, 5 of 12 constructs

Table 1—Comparison of mean ± SD yield, peak, and failure loads during biomechanical (tensile force) testing of cadaveric canine SDFTs that were transected and repaired by placement of a circumferential ES in a simple continuous pattern with polypropylene suture of various calibers (n = 12 constructs/group).

	Load (N)		
Group	Yield	Peak	Failure
Size 0	129.39 ± 29.83[a]	148.65 ± 25.58[a]	145.42 ± 27.73[a]
2–0	110.71 ± 39.47[a]	147.31 ± 28.68[a]	144.30 ± 29.04[a]
3–0	92.24 ± 34.75[a]	100.77 ± 41.96[b]	96.32 ± 42.59[b]
4–0	67.32 ± 12.94[b]	75.00 ± 14.81[b]	73.69 ± 14.52[b]
5–0	54.15 ± 18.61[c]	61.06 ± 13.69[c]	59.97 ± 13.12[c]

The yield, peak, and failure loads each differ significantly ($P < 0.001$) among groups (linear mixed-effects analysis).

[a–c]Within a column, values with different superscript letters differ significantly ($P < 0.05$) between suture size groups (pairwise comparisons).

in the 4-0 group and 11 of 12 constructs in the 5-0 group failed by means of suture breakage. No groups had evidence of tissue failure in locations other than the repair site.

Discussion

The present study investigated the strength of SDFT repairs performed by placement of a circumferential ES created in a simple continuous pattern with size-0, 2-0, 3-0, 4-0, and 5-0 polypropylene suture in cadaveric specimens from dogs (n = 12 constructs/group). Our results showed that the use of size-0 or 2-0 suture significantly increased the strength of these tendon repair constructs, with greater mean peak and failure loads than constructs created with all smaller-caliber sutures evaluated and greater yield loads than those created with 4-0 and 5-0 suture. In addition, repairs created with size-0 and 2-0 suture failed exclusively by suture pull-through, indicating that these ESs were stronger than the surrounding tendon tissues and tendon-suture interactions.

Previous experimental studies[12,13,23,27–31] have shown that the strength of tendon repair constructs and resistance to deformation are influenced by a number of interrelated factors. One study[27] showed that modified Kessler core suture repair created with an 8-strand suture technique required significantly greater force to cause anastomotic separation, compared with a 4-strand technique, for repair of flexor tendons in dogs used for human medical research. Investigators of other studies[13,28] have recommended a 7- to 10-mm length of purchase for both LL core sutures and interlocking horizontal mattress ESs combined with core sutures (in a cross-locked cruciate pattern) to provide greater strength and resistance to gap formation at the tendon repair site.[13,28] Regarding the caliber of the suture material used for flexor tendon repair, a previous study[29] of human cadaveric tissue showed that the use of 3-0 suture resulted in a nearly 3-fold increase in fatigue strength (the ability of the repair to resist construct failure by cyclic loading), compared with 4-0 suture; in that study, no difference in fatigue strength was found between modified Kessler core suture repairs created with a 2-strand or 4-strand technique or between those performed with a locking or nonlocking repair pattern. Other investigators used 3 different suture patterns for core suture repair (with no ES component) of human cadaveric flexor digitorum profundus tendons with 2-0, 3-0, 4-0, or 5-0 braided polyester suture and found significant, almost linear increases in construct strength as suture caliber increased.[12] Our study results for ES repair of canine SDFTs were similar to those findings and in general agreement with results of a study[23] that evaluated multiple factors for repair of simulated lacerations in cadaveric zone II tendons from human hands and found that increasing the suture caliber for the ES component from 6-0 to 5-0 improved the loads tolerated prior to failure. In the present study, the use of larger-caliber sutures (size-0 or 2-0) for creation of the ES increased construct strength, with respective mean yield, peak, and failure loads ranging from approximately 1.6 to 2.4, 2.0 to 2.4, and 2.0 to 2.4 times those for 4-0 or 5-0 sutures.

Differences in the results from biomechanical studies of flexor tendon repair constructs may relate to differences in suture caliber and composition, bite depth into the tendon substance, and bite distance of the ES from the transection site. In the previously described study[23] of cadaveric human tendons and an in vivo study[30] of core and ES modifications for repair of transected digital flexor tendons in the manus of dogs, the investigators demonstrated that construct strength was affected by factors such as suture strand length and suture purchase from the transection site, as well as suture caliber. We recognize that the caliber of the ES is not the only component that should be considered when choosing and designing an optimal tendon repair technique for implementation in dogs.

Construct failure in the present study resulted from ES breakage or ES pull-through. Suture breakage occurs along the length of the ES or at the level of the knot when the ES represents the weaker portion of the repair. When the holding capacity of the tissue being tested is weaker than the ES, tissue pull-through will likely be the predominant mode of failure.[17] Tissue failure occurs when the suture repair is stronger than the musculotendinous unit, origin, or enthesis in the construct being loaded. Failure as a result of suture pull-through was found for all constructs in the size-0 (12/12) and 2-0 (12/12) groups and most constructs (10/12) in the 3-0 group of the present study. Conversely, fewer than half of the constructs in the 4-0 group (5/12) and only 1 of 12 in the 5-0 group failed as a result of suture pull-through, with the remainder having suture breakage. This was similar to the findings in the aforementioned study[12] of human digital flexor tendon repairs created with a modified core Kessler pattern, in which investigators found that a higher proportion of failures was caused by suture pullout when 2-0 or 3-0 suture was used (100% and 60%, respectively), compared with 4-0 or 5-0 suture (3% and 0%, respectively). In another study,[29] 100% (60/60) of human digital flexor tendon repair constructs created with 4-0 suture failed because of suture breakage, whereas 6 of 10 created with 3-0 suture failed because of suture pull-through, when a nonlocking modified Kessler suture pattern was used. The differences in failure mode among the suture caliber groups in our study may have indicated that the weakest point in the repaired suture construct was the interaction between the suture and tendon tissue when size-0 or 2-0 polypropylene suture was used. This suggested that increasing the caliber of suture used to create an ES could potentially decrease the occurrence of suture breakage in clinical cases, in addition to improving the biomechanical properties of repairs.[23]

Some limitations applied to the study reported here. We did not evaluate the occurrence of repair site gap formation and the loads at which this developed. Gap formation of > 3 mm between tendon ends significantly increases the risk of another rupture at the repair site, especially during the early stages of tendon healing.[3,32] Therefore, minimization of gap formation at the repair site is important for successful repair and the likely progression of healing in clinical patients. In preliminary experiments prior to this study, no gapping was observed in the smaller-caliber suture groups (4-0 and 5-0), likely because ES failure occurred by breakage prior to the development of an identifiable gap between tendon ends. In addition, clinical scenarios cannot be accurately replicated with the ex vivo test methods that we used. Processes that are clinically important, such as the inflammatory changes associated with surgery, damage to the tenuous tendon blood supply, degenerative changes to collagen fibers, and protracted healing process, could not be assessed. We also aimed to standardize SDFT size by use of specimens from a fairly homogenous sample of adult dogs, and therefore, the results may be applicable only to dogs of similar sizes and with similarly sized tendons. The effect of ES caliber on the tensile strength of SDFT repairs for smaller dogs or repairs of sheathed or flatter canine tendons remains an area for future evaluation. We did not evaluate the effect of glide function in the present study, and larger-caliber suture can indirectly increase suture bulk at the repair site, increasing glide resistance.[33] We also did not cyclically evaluate repaired constructs, and cyclic testing has been shown to more closely represent conditions that are present in vivo, compared with an acute distraction-to-failure model.[34] In the study reported here, we purposefully followed the methodologies of previous studies[12,13,23,29–31] to allow meaningful conclusions to be drawn in attempts to recreate acute distractive forces placed on tendon repairs in the immediate postoperative period. Finally, our study only tested the effects of various calibers of polypropylene suture used to create ES, without concurrent evaluation of a core suture, which would routinely be placed in a clinical setting.[19,20] Our study provided information for veterinary surgeons, allowing a greater understanding of how suture caliber affects the tensile strength of SDFT repairs performed with an ES. However, it is important to recognize that successful tendon repair relies on a number of important interrelated factors.

In conclusion, our results suggested size-0 and 2-0 sutures should be considered when placing an ES for flexor tendon repairs in dogs. Patient size should be considered, and clinical judgement must be used in determining the caliber of suture most suitable for ES placement during repair of ruptured flexor tendons in dogs. Further studies are necessary to evaluate other factors that can influence the biomechanical properties of tendon repair constructs from dogs and assess the effects of ES caliber in vivo with a controlled randomized clinical trial.

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Footnotes

a. Euthasol, Virbac AH Inc, Fort Worth, Tex.
b. Ziplock, SC Johnson & Son Inc, Racine, Wis.
c. iPhone 8 camera, Apple Inc, Cupertino, Calif.
d. Medline, Northfield, Ill.
e. ImageJ, NIH, Bethesda, Md.
f. Research Randomizer, version 4.0, Urbaniak GC, Plous S. Available at: randomizer.org. Accessed May 4, 2021.
g. Surgipro, Covidien Ltd, Dublin, Ireland.
h. 4.5X surgical loupe, Surgitel, General Scientific Corp, Ann Arbor, Mich.
i. Monoject hypodermic needle, Covidien Ltd, Dublin, Ireland.
j. Instron model 5967, Instron Inc, Norwood, Mass.
k. SKU 1652-1, Sawbones, Vashon Island, Wash.
l. Lumix DMC-FZ200, Panasonic Corp, Newark, NJ.
m. Bluehill 3, Instron Inc, Norwood, Mass.
n. Matlab R2018b, Mathworks, Natick, Mass.
o. SAS, version 9.4, SAS Institute Inc, Cary, NC.

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