Challenging the Perceptions of Human Tendon Allografts

Influence of Donor Age, Sex, Height, and Tendon on Biomechanical Properties

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Background: The use of allograft tendons has increased for primary and revision anterior cruciate ligament reconstruction, but allograft supply is currently limited to a narrow range of tendons and donors up to the age of 65 years. Expanding the range of donors and tendons could help offset an increasing clinical demand.

Purpose: To investigate the effects of donor age, sex, height, and specific tendon on the mechanical properties of a range of human lower leg tendons.

Study Design: Descriptive laboratory study.

Methods: Nine tendons were retrieved from 39 fresh-frozen human cadaveric lower legs (35 donors [13 female, 22 male]; age, 49-99 years; height, 57-85 inches [145-216 cm]) including: Achilles tendon, tibialis posterior and anterior, fibularis longus and brevis, flexor and extensor hallucis longus, plantaris, and flexor digitorum longus. Tendons underwent tensile loading to failure measuring cross-sectional area (CSA), maximum load, strain at failure, ultimate tensile strength, and elastic modulus. Results from 332 tendons were analyzed using mixed-effects linear regression, accounting for donor age, sex, height, and weight.

Results: Mechanical properties were significantly different among tendons and were substantially greater than the effects of donor characteristics. Significant effects of donor sex, age, and height were limited to specific tendons: Achilles tendon, tibialis posterior, and tibialis anterior. All other tendons were unaffected. The Achilles tendon was most influenced by donor variables: greater CSA in men (β = 15.45 mm^2; Šidák adjusted $P < .0001$), decreased maximum load with each year of increased age (β = −17.20 N per year; adjusted $P = .0253$), and increased CSA (β = 1.92 mm^2 per inch; adjusted $P < .0001$) and maximum load (β = 86.40 N per inch; adjusted $P < .0001$) with each inch of increased height.

Conclusion: Mechanical properties vary significantly across different human tendons. The effects of donor age, sex, and height are relatively small, are limited to specific tendons, and affect different tendons uniquely. The findings indicate that age negatively affected only the Achilles tendon (maximum load) and challenge the exclusion of donors aged >65 years across all tendon grafts.

Clinical Relevance: The findings support including a broader range of tendons for use as allografts for anterior cruciate ligament reconstruction and reviewing the current exclusion criterion of donors aged >65 years.

Keywords: musculoskeletal; graft; biomechanics of tendon; tissue mechanics; ACL; reconstruction

Anterior cruciate ligament (ACL) tear is one of the most common and debilitating knee injuries in sports. Reconstructive surgery using a graft is widely accepted as the best treatment strategy for younger or active patients with higher functional demands or patients with chronic knee instability.[15,34] Rates of primary and revision ACL reconstruction have respectively increased by 43% and 127% between the years 2000 and 2015.[41]

While tendon autografts (surgically retrieved from the patient) are generally considered the standard for ACL reconstruction, the use of tendon allografts (sourced from human tissue donors) has increased for primary and revision surgery,[19] with usage as high as 42.4% and 78.8%, respectively.[26] Tendon autografts have the advantage of providing greater fixation strength, superior short-term

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mechanical properties, and good long-term results,[8] but they are limited by significant donor-site morbidity and availability constraints when adequate tendons are sourced in young patients[33] and for use in multiligament and revision surgery.[3] Allografts provide a solution to overcome these issues, although they are associated with slower incorporation and the potential for disease transmission.[3] Although tendon allografts have been shown to have a significantly higher rerupture rate in younger highly active patients (age, <25 years), they provide comparable clinical outcomes with autografts in patients aged ≥35 years,[38] particularly when nonirradiated or nonchemically treated grafts are used.[19] The most common tendons used as allografts for ACL reconstruction include bone−patellar tendon–bone (BPTB), hamstring (semitendinosus with or without gracilis tendon), tibialis anterior, tibialis posterior, and Achilles tendon.[10,11] Additional tendons with use as autografts for a range of reconstructive applications, such as fibularis longus,[18] flexor hallucis longus,[28] plantaris,[1,22,31] and flexor digitorum longus,[16] may have uses as allografts for specific cases of ACL injury. The final decision on graft source and type needs to be personalized and cater to the functional demands and age of the patient (ie, withstand certain loads and be appropriately sized), but it is also heavily influenced by surgeon preference.[2]

Up to 96.5% of surgeons surveyed believe that donor age negatively affects the success of tendon allografts (22.5%, small effect; 47.3%, moderate; 26.7%, strong), and many report that they will not accept allografts from donors aged >40 years.[2] While no standards for donor age have been set by the US Food and Drug Administration or the American Association of Tissue Banks, leading tissue banks accept donors up to the age of 65 years.[27] Some of the most important considerations for the selection of any tendon graft are the initial tensile mechanical properties. The effect of donor age on tendon strength has mixed reports, with some studies describing an inverse relationship[14,21,24,35,36] while others cite no effect of aging.[4-6,13,14,39,40] Importantly, most studies report on only a single anatomic tendon, and collectively these studies cover a narrow range of different tendons. Thus, our current knowledge of the effects of age and other donor variables on potential allograft tendon mechanical properties is often extrapolated from trends observed in restricted cohorts, generalized across different human tendons, and collated from studies that use different testing parameters that may not be directly comparable.

Knowledge of the variables that influence the mechanical properties of tendons may assist surgeons and patients to make informed decisions on graft selection and may expand graft supplies to offset an increasing clinical demand via the inclusion of a broader range of eligible donors and different tendons. The present study aimed to determine the effects of donor age, sex, height, and tendon on the mechanical properties of a range of lower leg tendons commonly used as grafts, either for ACL reconstruction or other applications. The outcomes of the study will determine if different tendons are uniquely influenced by donor variables.

METHODS

Tissue Acquisition

A total of 39 fresh-frozen adult human cadaveric lower legs from 35 donors (13 female, 22 male) were sourced from 2 cohorts for inclusion in this study; 31 lower legs were sourced from an older cohort: 27 donors (10 female, 17 male) with a mean age of 81 years (range, 68-99 years) and a mean height of 66.75 inches (range, 64-72 inches [163-183 cm]). Four donors had paired limbs (2 female, 2 male), as opposed to a single limb from the remaining 23 donors. Any tendons damaged during retrieval were excluded from subsequent testing. Eight additional lower leg pairs were sourced from a younger cohort: 8 donors (3 female, 5 male) with a mean age of 56 years (range, 49-65 years) and mean height of 68.40 inches (range, 57-85 inches [145-216 cm]). Tendons were retrieved from one of the paired limbs. Any tendons damaged during retrieval from the younger cohort were replaced by the same tendon in the contralateral limb. Ethics approval was granted by the Northern Sydney Local Health District Human Research Ethics Committee.

Specimen Retrieval

Nine distinct tendons were retrieved from each donor limb: Achilles tendon, tibialis posterior and anterior, fibularis (peroneus) longus and brevis, flexor and extensor hallucis

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longus, plantaris, and flexor digitorum longus. These specific tendons were selected by their availability in tibial plateau to toe-tip specimens. All tendons were retrieved by two orthopaedic surgeons (N.H., P.H.) in the same sequence to facilitate accurate identification, labeling, and safe removal. Each tendon was identified and incised at its distal insertion and then followed and released proximally at its musculotendinous junction (full dissection protocol in the Appendix Methods, available in the online version of this article). After removal, tendons were wrapped in saline-soaked gauze, sealed in double zipper-locked plastic bags, and stored at −20°C.

Specimen Preparation

Tendons were thawed at room temperature for one hour. Muscle, fat, and loose connective tissue were removed using a scalpel and by wiping dry gauze distal to proximal to avoid fiber separation. Visual inspection verified integrity. The free tendon was marked using picrosirius red dye at the midpoint and 25 mm proximal and distal to the midpoint, identifying the desired 50-mm gauge length. Cross-sectional area (CSA) was measured at the narrowest region using a custom micrometer device as previously published.[9] Briefly, the tendon was placed under a glass slide attached to a ratchet micrometer. The slide was lowered until the torque limit was reached, and the specimen height was recorded from the micrometer. The specimen was photographed through the glass slide. The width was measured using ImageJ (Version 1.52k; National Institutes of Health). CSA was calculated by assuming a rectangular cross section and multiplying the height and width.

Biomechanical Testing

Specimens were gripped in custom cryogenic clamps, cooled using dry ice, and attached to a servohydraulic testing machine (8874; Instron Corp). Specimens were first secured in the top clamp (using the dye to align the desired gauge length), before being lowered and fastened in the bottom clamp, ensuring that the specimen was slack when the grips were tightened. Specimens were loaded under uniaxial tension to failure at 2.5 mm/s (5% strain/s). Force was measured at 100 Hz using a 25-kN load cell (Dynacell 2527-201; Instron); crosshead displacement was measured at 100 Hz (8874; Instron); and mode of failure was determined visually (midsubstance, edge of clamps, within clamps, or grip slip) using the picrosirius dye lines. Only specimens that failed within the gauge length (midsubstance to edge of clamps) were included in the analysis of mechanical outcomes.

Maximum load at failure was defined as the maximum measured force. The working gauge length was defined as the distance between clamps when a force threshold of 10 N was reached. Displacement data were converted to strain by dividing the change in displacement by the working gauge length. Force data were converted to stress by dividing the force by the CSA. A custom MATLAB program (Version 9.4; MathWorks) generated stress-strain curves for each specimen to determine ultimate tensile strength (UTS; stress at maximum measured force) and strain at failure (strain at maximum measured force). Elastic modulus was objectively calculated by fitting a fifth-order polynomial to the stress-strain data between 0% strain and strain at failure and determining the maximum gradient. This was plotted back on the stress-strain data to visually confirm that it was within what we would classify as the "linear" region.

Statistical Analysis

Results were analyzed using multivariate mixed-effects linear regression models. Two approaches were performed assuming that the effects of donor variables were either generalizable across different tendons by excluding interaction terms or tendon specific via the inclusion of interaction terms. The overall "generalized" model was used to evaluate the main effects of age, sex, and tendon on each outcome (CSA, maximum load, UTS, strain at failure, and elastic modulus), correcting for donor height and weight. Multiple comparisons between tendons were adjusted using the Šidák correction. Height was a significant predictor for most outcomes and therefore evaluated in the tendon-specific models alongside age and sex. The tendon-specific models were used to determine the effects of age, sex, and height on each tendon via the inclusion of interaction terms for sex and tendon, age and tendon, and height and tendon. Weight remained a covariate in the models. Multiple comparisons of age, sex, and height were adjusted using the Šidák correction. Regression models included random intercept terms at the limb and donor levels to account for the nonindependence of tendons retrieved from the same limb and from paired limbs of the same donor, respectively. Effects were considered significant where P- or Šidák adjusted P-value $< .05$. All statistical analyses were performed using Stata/IC (Version 13.1; StataCorp).

RESULTS

The plantaris tendon was absent in 6 donors (4 female, 2 male). Thirteen tendons damaged during retrieval, preparation, or biomechanical testing were excluded from all analyses. CSA data from 332 specimens were included in the analysis. Sixteen tendons that slipped or failed within the clamps during tensile testing were excluded from the analysis of mechanical outcomes. Of the 316 tendons, the relative distribution of failure locations by tendon type is summarized in Appendix Table A1 (available online). Seven tendons failed within the gauge region, but elongation also occurred outside the gauge region at the first tooth within the clamps; thus, these tendons were excluded from strain-based analyses (strain at failure and elastic modulus). Absolute values and final sample sizes for each tendon and outcome are summarized in Appendix Table A2.

Effect of the Tendon

The specific tendon had the largest influence on tensile properties (Figure 1; Appendix Table A3, available online).

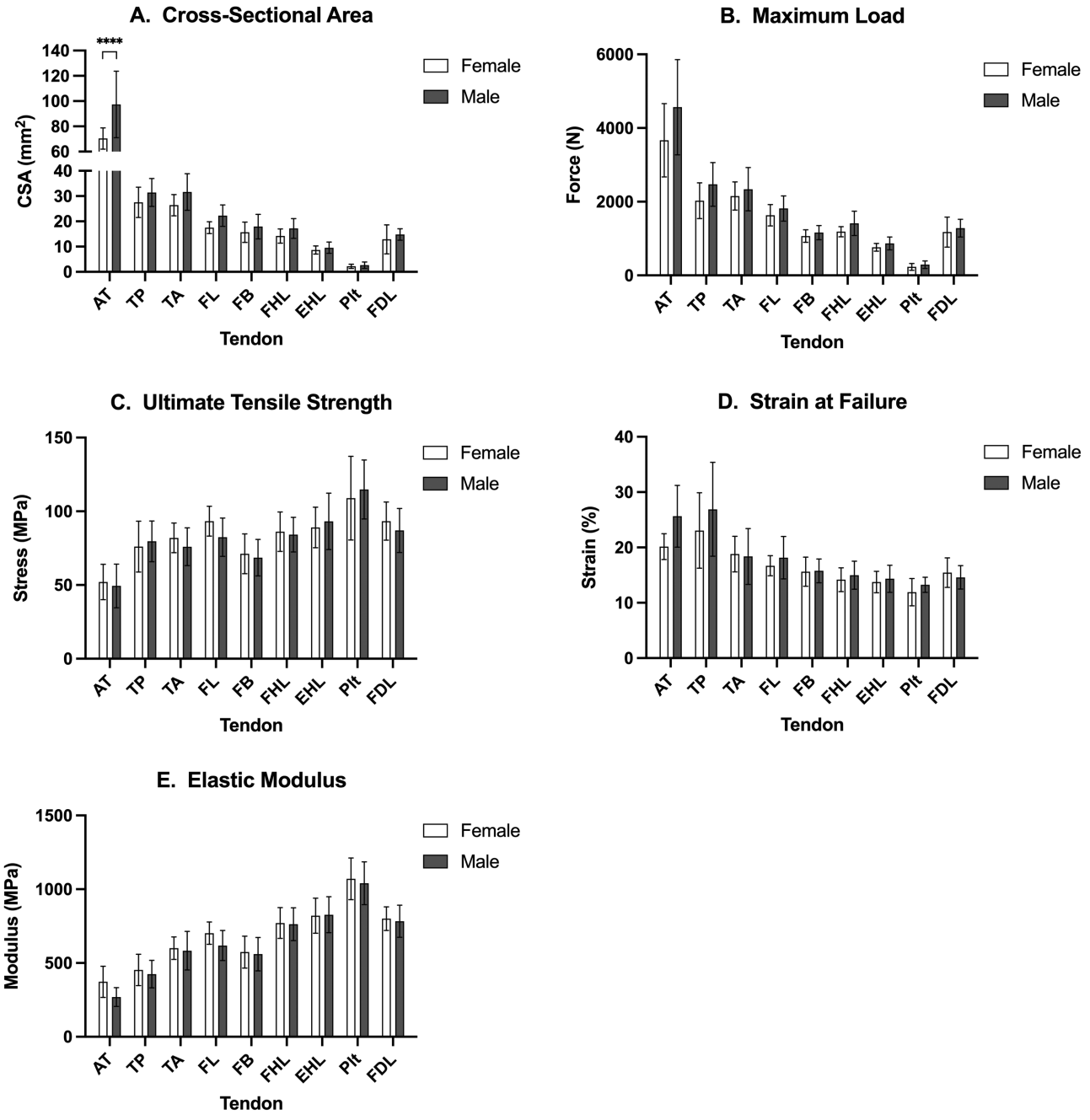

Figure 1. Effect of sex on individual lower leg tendon tensile properties. Data are presented as mean ± SD. Statistically significant sex differences within specific tendons as determined from the mixed-effects regression models: ****$P < .0001$ (Šidák adjusted). AT, Achilles tendon; CSA, cross-sectional area; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; TA, tibialis anterior; TP, tibialis posterior.

On average, the Achilles tendon had the largest CSA (87.17 mm^2) and maximum load (4206.57 N) and the second highest strain at failure (23.41%) but had the lowest UTS (50.47 MPa) and elastic modulus (311.14 MPa). Conversely, the plantaris tendon had the smallest CSA (2.53 mm^2), maximum load (271.70 N), and strain at failure (12.84%) but the greatest UTS (112.99 MPa) and elastic modulus (1050.11 MPa). When controlling for age, sex, height, and weight, most comparisons among specific tendons were significantly different (Šidák adjusted $P < .05$), revealing largely unique tendon properties across all outcomes measured (Appendix Table A3). The few exceptions were no significant differences between flexor hallucis longus and flexor digitorum longus tendons for any outcome. Tibialis posterior and anterior tendons also showed no significant differences in CSA, maximum load,

TABLE 1
Tendon-Specific Effects of Donor Sex[a]

	CSA, mm^2			Maximum Load, N			UTS, MPa			Strain at Failure, %			Elastic Modulus, MPa		
Variable	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value
Female	Ref			Ref			Ref			Ref			Ref		
Male															
AT	15.45	7.48, 23.42	**<.0001**	307.93	–189.55, 805.41	.5589	–4.94	–20.56, 10.68	.9868	3.36	–0.55, 7.27	.1462	–91.67	–207.46, 24.12	.2296
TP	3.79	–4.08, 11.67	.8378	257.37	–264.66, 779.41	.8185	–4.70	–21.11, 11.71	.9934	1.75	–2.29, 5.8	.9063	–50.38	–170.12, 69.35	.9198
TA	1.63	–6.33, 9.59	.9995	–32.03	–537.95, 473.89	≥.999	–5.69	–21.57, 10.19	.9697	–1.18	–5.03, 2.67	.9895	7.32	–106.71, 121.34	≥.999
FL	2.32	–5.51, 10.14	.9917	–23.10	–510.9, 464.69	≥.999	–13.04	–28.34, 2.25	.1536	0.52	–3.18, 4.21	≥.999	–85.18	–194.59, 24.24	.2491
FB	0.88	–7.3, 9.06	≥.999	21.65	–525.9, 569.2	≥.999	–1.86	–19.12, 15.39	≥.999	–0.82	–5, 3.35	.9996	28.27	–95.33, 151.86	.9988
FHL	1.58	–6.26, 9.42	.9996	94.81	–415.68, 605.3	.9998	–2.53	–18.57, 13.5	.9999	0.51	–3.37, 4.39	≥.999	–3.34	–118.11, 111.44	≥.999
EHL	0.05	–7.91, 8.01	≥.999	58.19	–453.27, 569.64	≥.999	4.18	–11.89, 20.25	.9968	–0.06	–4.02, 3.9	≥.999	37.73	–79.42, 154.88	.9851
Plt	–0.11	–9.33, 9.11	≥.999	75.91	–494.15, 645.96	≥.999	8.76	–9.22, 26.74	.8283	1.28	–3.07, 5.62	.9921	–14.17	–142.9, 114.56	≥.999
FDL	0.93	–6.9, 8.75	≥.999	30.16	–456.63, 516.95	≥.999	–8.28	–23.54, 6.98	.7245	–1.35	–5.04, 2.34	.9652	4.43	–104.75, 113.6	≥.999

[a]Adjusted results from the multilevel mixed-effects linear regression analysis using the Šidák correction. *P* values are Šidák adjusted with significance identified in bold ($P < .05$). β, effect size; AT, Achilles tendon; CSA, cross-sectional area; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; Ref, reference; TA, tibialis anterior; TP, tibialis posterior; UTS, ultimate tensile strength.

and UTS, while differences were observed in strain at failure (β = −6.74%; adjusted $P < .0001$) and elastic modulus (β = 154.63 MPa; adjusted $P < .0001$).

Effect of Sex

When the effect of sex was generalized across all tendons (Appendix Table A3, available online), the only outcome that differed significantly was CSA, being significantly larger in men versus women (β = 2.95 mm^2; $P = .0125$). When tendon-specific effects were analyzed (Figure 1, Table 1), significant sex differences were replicated only in the Achilles tendon, with CSA significantly greater in men versus women (β = 15.45 mm^2; adjusted $P < .0001$). The effect of sex on CSA within the Achilles tendon was ~5-fold greater than the generalized effect, while there were no sex differences in any of the other tendons. This suggests that Achilles tendon measurements skewed the observed generalized effect and, at the same time, the generalized effect masked the true magnitude of the unique effect of sex on the Achilles tendon.

Effect of Age

When the effect of age was generalized across all tendons (Appendix Table A3, available online), advancing age had a significant but small negative effect on UTS (β = −0.19 MPa per year of increased age; $P = .0072$) and elastic modulus (β = −1.07 MPa per year; $P = .0312$). No changes in CSA, maximum load, or strain at failure were observed. However, when the tendon-specific effects of age were analyzed (Figure 2, Table 2), significant changes were limited to a few tendons and outcomes, where advancing age significantly decreased maximum load in the Achilles tendon (β = −17.20 N per year; adjusted $P = .0253$), increased maximum load in the tibialis anterior (β = 16.68 N per year; adjusted $P = .0365$), and increased strain at failure in the tibialis posterior (β = 0.25% per year; adjusted $P < .0001$).

Effect of Height

When the effect of height was generalized across all tendons (Appendix Table A3, available online), increasing height had a significant effect on 4 of 5 outcomes: CSA, maximum load, strain at failure, and elastic modulus. Outcomes that increased with increasing donor height included CSA (β = 0.36 mm^2 per inch of height; $P = .0010$), maximum load (β = 23.84 N per inch; $P = .0035$), and strain at failure (β = 0.13% per inch; $P = .0115$), whereas elastic modulus decreased (β = –4.93 MPa per inch; $P = .0009$). No changes in UTS were observed. When the tendon-specific effects of donor height were analyzed (Figure 2, Table 3), outcomes and tendons were not consistently affected. With increasing height, the Achilles tendon significantly increased in CSA (β = 1.92 mm^2 per inch; adjusted $P < .0001$) and maximum load (β = 86.40 N per inch; adjusted $P < .0001$), while strain at failure significantly increased in the tibialis posterior tendon (β = 0.50% per inch; adjusted $P = .0070$). As with the effect of sex, the significant tendon-specific effect sizes were several factors larger than the generalized effects, and while the generalized analysis indicated a significant change in elastic modulus with donor height, there was no significant change within individual tendons.

DISCUSSION

The most important finding of this study was that the effects of donor age, sex, and height are relatively small, are limited to specific tendons, and affect these tendons differently, as summarized in Table 4. The specific tendon, rather than donor age, sex, or height, may represent the more important consideration for graft selection. Furthermore, the effects of donor variables should not be generalized across all tendons, as demonstrated by the differences in significant results when using a generalized versus tendon-specific analytic approach. This is particularly

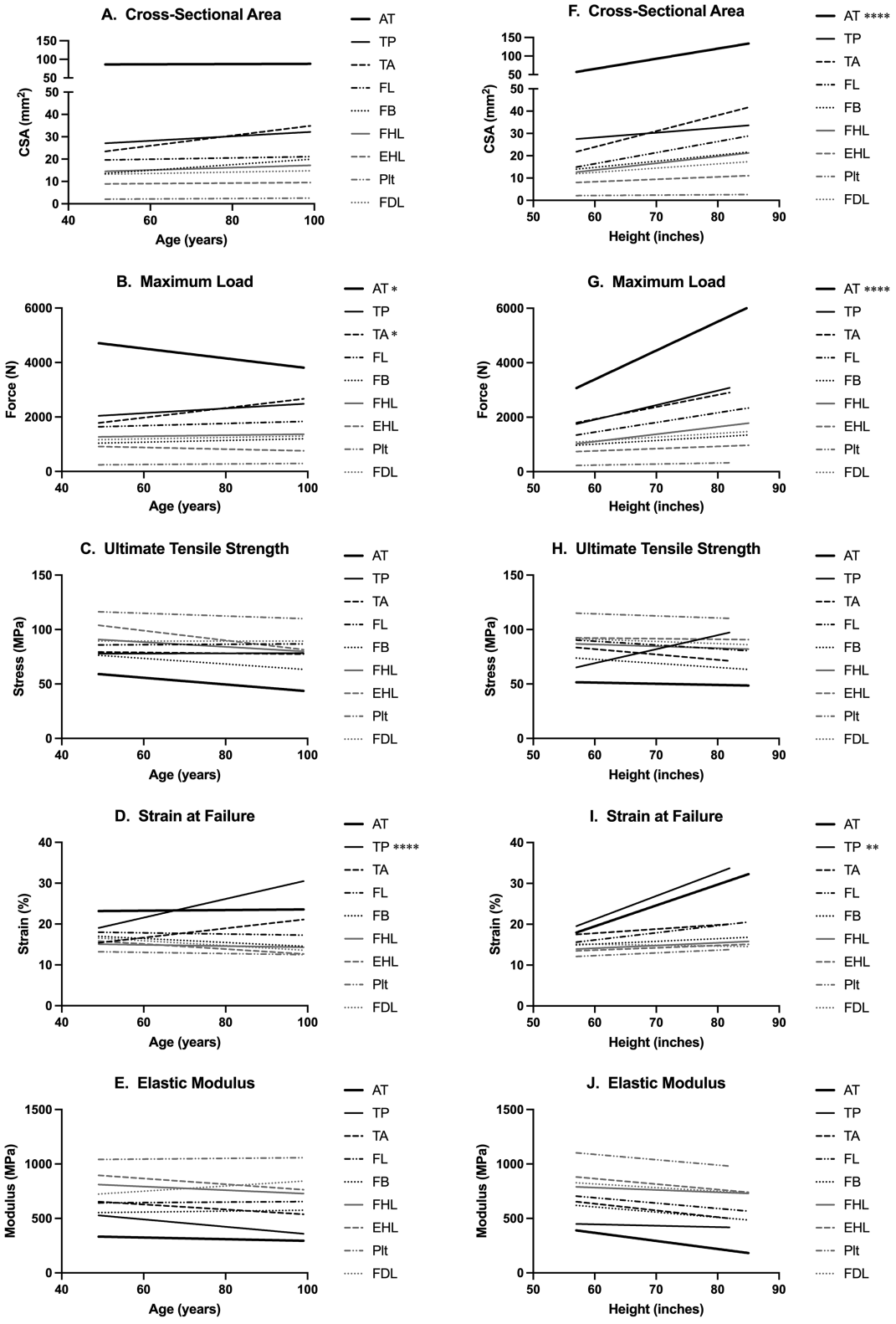

Figure 2. Effects of donor age (A-E) or height (F-J) on individual lower leg tendon tensile properties. Data are presented as simple linear regressions. Statistically significant effects of age and height within individual anatomic tendons as determined from the mixed-effects regression models (all Šidák adjusted): *$P < .05$. **$P < .01$. ****$P < .0001$. AT, Achilles tendon; CSA, cross-sectional area; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; TA, tibialis anterior; TP, tibialis posterior.

TABLE 2
Tendon-Specific Effects of Donor Age[a]

	CSA, mm^2			Maximum Load, N			UTS, MPa			Strain at Failure, %			Elastic Modulus, MPa		
Variable	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value
Age, y															
AT	0.11	−0.15, 0.36	.9133	−17.20	−33.14, −1.26	**.0253**	−0.35	−0.85, 0.15	.4025	0.01	−0.12, 0.14	≥.999	−1.01	−4.85, 2.84	.9966
TP	0.11	−0.14, 0.37	.9063	8.96	−7.82, 25.75	.7418	0.02	−0.51, 0.55	≥.999	0.25	0.12, 0.38	**<.0001**	−3.65	−7.49, 0.19	.0750
TA	0.22	−0.03, 0.48	.1326	16.68	0.6, 32.77	**.0365**	−0.08	−0.59, 0.42	.9999	0.11	−0.01, 0.23	.0971	−2.57	−6.18, 1.03	.3596
FL	0.03	−0.22, 0.28	≥.999	1.98	−13.92, 17.87	≥.999	−0.05	−0.55, 0.45	≥.999	−0.02	−0.14, 0.1	≥.999	−0.23	−3.79, 3.33	≥.999
FB	0.12	−0.14, 0.37	.8734	1.79	−14.92, 18.5	≥.999	−0.30	−0.82, 0.23	.6841	−0.06	−0.19, 0.07	.8703	0.40	−3.36, 4.16	≥.999
FHL	0.05	−0.2, 0.31	.9994	0.81	−15.09, 16.72	≥.999	−0.25	−0.75, 0.25	.7990	−0.02	−0.14, 0.11	≥.999	−1.92	−5.48, 1.65	.7351
EHL	0.00	−0.25, 0.25	≥.999	−4.63	−20.49, 11.24	.9926	−0.46	−0.96, 0.04	.0914	−0.07	−0.19, 0.06	.7390	−2.70	−6.32, 0.91	.2985
Plt	−0.01	−0.27, 0.26	≥.999	−1.50	−18.09, 15.09	≥.999	−0.16	−0.68, 0.36	.9874	−0.02	−0.14, 0.11	≥.999	0.00	−3.72, 3.72	≥.999
FDL	0.03	−0.23, 0.28	≥.999	1.56	−14.16, 17.28	≥.999	−0.06	−0.55, 0.44	≥.999	−0.07	−0.19, 0.05	.6903	2.16	−1.35, 5.68	.5675

[a]Adjusted results from the multilevel mixed-effects linear regression analysis using the Šidák correction. *P* values are Šidák adjusted with significance identified in bold ($P < .05$). β, effect size; AT, Achilles tendon; CSA, cross-sectional area; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; TA, tibialis anterior; TP, tibialis posterior; UTS, ultimate tensile strength.

TABLE 3
Tendon-Specific Effects of Donor Height[a]

	CSA, mm^2			Maximum Load, N			UTS, MPa			Strain at Failure, %			Elastic Modulus, MPa		
Variable	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value	β	95% CI	*P* Value
Height, in															
AT	1.92	1.2, 2.64	**<.0001**	86.40	40.91, 131.89	**<.0001**	−0.05	−1.47, 1.37	≥.999	0.35	0, 0.69	.0532	−4.49	−14.8, 5.82	.9029
TP	−0.03	−0.74, 0.69	≥.999	34.91	−19.09, 88.9	.4984	1.37	−0.33, 3.08	.2064	0.50	0.09, 0.91	**.0070**	−0.75	−12.96, 11.45	≥.999
TA	0.56	−0.15, 1.28	.2317	42.27	−10.22, 94.76	.2106	−0.36	−2.01, 1.29	.9992	0.17	−0.23, 0.57	.9160	−8.51	−20.44, 3.43	.3617
FL	0.31	−0.4, 1.02	.8998	31.83	−13.28, 76.94	.3759	0.13	−1.28, 1.54	≥.999	0.14	−0.2, 0.48	.9296	−2.24	−12.31, 7.83	.9991
FB	0.16	−0.58, 0.89	.9993	7.89	−40.99, 56.77	.9999	−0.39	−1.92, 1.15	.9974	0.10	−0.27, 0.47	.9940	−7.85	−18.81, 3.11	.3558
FHL	0.16	−0.55, 0.88	.9988	17.56	−28.54, 63.67	.9554	−0.20	−1.64, 1.25	≥.999	0.03	−0.32, 0.37	≥.999	−3.42	−13.73, 6.89	.9818
EHL	0.04	−0.68, 0.75	≥.999	0.49	−44.45, 45.42	≥.999	−0.42	−1.82, 0.99	.9912	0.05	−0.3, 0.39	≥.999	−8.29	−18.41, 1.83	.1928
Plt	−0.01	−0.9, 0.88	≥.999	−5.88	−61.19, 49.44	≥.999	−0.82	−2.56, 0.93	.8568	−0.01	−0.43, 0.41	≥.999	−5.54	−18.01, 6.93	.8917
FDL	0.07	−0.64, 0.79	≥.999	7.14	−37.38, 51.67	.9999	0.06	−1.33, 1.45	≥.999	0.03	−0.31, 0.36	≥.999	−4.97	−14.91, 4.97	.8061

[a]Adjusted results from the multilevel mixed-effects linear regression analysis using the Šidák correction. *P* values are Šidák adjusted with significance identified in bold ($P < .05$). β, effect size; AT, Achilles tendon; CSA, cross-sectional area; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; TA, tibialis anterior; TP, tibialis posterior; UTS, ultimate tensile strength.

true when effects are determined from data sets that include or solely focus on individual tendons that have notable changes with donor characteristics, such as the Achilles tendon from the subset of tendons used in this study. Finally, although sex and height are not used as donor selection requirements, donor age >65 years is a strict exclusion criterion.[27] While the generalized effect of aging was observed to decrease the UTS and elastic modulus, the effect sizes were small, and for perspective, donor height was more influential. More importantly, analysis within specific tendons revealed a negative effect of aging only on the maximum load of the Achilles tendon. Therefore, the findings have important implications for the current maximum age of tissue donors accepted, suggesting that age should be less emphasised as an overall exclusion criterion but rather a factor to consider for specific tendons (eg, Achilles tendon). Increasing the inclusion age for tissue donors and the use of tendons that show small changes with individual donor characteristics represent areas to increase tendon allograft supply to meet a growing clinical need.

Biomechanical properties represent one of the most important considerations for graft selection. The specific tendon has the largest effect on tensile properties, with each exhibiting largely unique properties across all outcomes measured. Because donor-site morbidity and functional deficits created do not need to be considered when using allografts, it is not surprising that the three tendons exhibiting the largest CSA and maximum failure loads (Achilles tendon, tibialis posterior, and tibialis anterior) are those commonly used as allografts for ACL reconstruction.[10] Interestingly, these preferred allograft candidates with the highest failure loads possess a lower UTS and elastic modulus and demonstrate the greatest variability with sex, aging, and increasing height. Conversely the plantaris tendon, utilized as an autograft for upper limb[1] and ankle[31] reconstructions, has the smallest CSA and lowest maximum load at failure but possesses the greatest UTS and elastic modulus. Tendons with a smaller CSA (eg, hamstring semitendinosus and gracilis tendons at ~14 and 8 mm^2, respectively)[30] are widely used for ACL reconstructions via looped and bundled configurations to produce an

TABLE 4
Summary of Tendon-Specific Effects of Donor Sex, Age and Height on Human Tendon Mechanical Properties. The Statistically Significant Effects of Donor Variables on Mechanical Properties are Identifiable by an Arrow Indicating the Direction of the Effect of: Sex, Advancing Age and Increasing Height.[a]

Tendon	Cross-Sectional Area			Maximum Load			Ultimate Tensile Strength			Strain at Failure			Elastic Modulus		
	Sex	↑Age	↑Height	Sex	↑Age	↑Height	Sex	↑Age	↑Height	Sex	↑Age	↑Height	Sex	↑Age	↑Height
AT	↑ Males		↑		↓	↑									
TP											↑	↑			
TA					↑										
FL															
FB															
FHL															
EHL															
Plt															
FDL															

[a]AT, Achilles tendon; EHL, extensor hallucis longus; FB, fibularis brevis; FDL, flexor digitorum longus; FHL, flexor hallucis longus; FL, fibularis longus; Plt, plantaris; TA, tibialis anterior; TP, tibialis posterior.

appropriately sized load-bearing graft construct. Noyes et al[30] defined the maximum load of single-strand semitendinosus and gracilis tendons as 1216 and 838 N respectively. Thus, all tendons except the plantaris tendon in the current study possess comparable or higher failure loads as single strands (Appendix Table A2, available online). This warrants further investigation to assess the potential of expanding the range of allograft tendons used for ACL reconstruction to include the fibularis longus and brevis, flexor and extensor hallucis longus, and flexor digitorum longus tendons. Despite the high material strength of the plantaris tendon, its small CSA and maximum load make it unlikely to be useful for intra-articular ACL reconstruction in isolation. However, Josipović et al[22] examined concurrent reconstruction of the ACL and anterolateral ligament to restore additional rotational stability using semitendinosus and plantaris tendon autografts, respectively. Because the anterolateral ligament is injured in 90% of patients with ACL ruptures and plantaris autografts are reported for other reconstructive applications,[1,31] scope remains for future investigations into the expansion of allografts available to include the plantaris tendon.

The generalized effect of sex was observed to influence CSA when correcting for age, tendon, height, and weight. Yet, when data with interaction terms for donor variables and specific tendons were analyzed, significant sex differences were observed only in the Achilles tendon (see Figure 1). This suggests that the sex differences observed for the CSA in the initial models were an oversimplification and largely driven by the sex differences in the Achilles tendon. These results caution against ignoring the interactions present between donor variables and distinct tendons, particularly when the data set includes tendons with large donor-specific differences, such as the Achilles. To confirm the effect size that the study was powered to detect, we performed a post hoc sensitivity power analysis using G*Power (Version 3.1; HHU),[12] computing an effect size of 0.03 with the following parameters: $\alpha = .05$, power = 0.80, total sample size = 332, number of tested predictors = 3 (age, sex, height), and total number of predictors = 8 (age, sex, height, weight, tendon, age × tendon, sex × tendon, height × tendon). Interestingly, sex differences have been indicated for human ACL,[7] but consideration of patient and donor sex for the selection of graft materials is not commonly reported. Equivalent in vitro sex studies on human tendons are relatively limited, and to our knowledge, this study is one of the first to evaluate the tensile failure mechanics of a broad range of tendons in a single study. Thermann et al[37] similarly found that Achilles tendons from male patients had larger CSAs, but they also cited a higher maximum failure load and UTS in male patients, which was not observed in our study. Hashemi et al[17] stated that patellar tendon failure properties were independent of sex, and Castile et al[6] reported no overall effect of sex on nonfailure mechanical properties (toe modulus, linear modulus, transition stress, transition strain) on a range of tendons, ligaments, and capsules. However, this latter report did not include any tendons investigated in the present study. Divergence in study conclusions may be associated with differences in donor cohorts and the tissues investigated, as well as the tendon dissection, preparation, and mechanical testing methods employed. Nevertheless, our findings are similar to human in vivo[20,23,25,42] and animal in vitro[32] studies citing sex differences in Achilles tendon properties, notably a larger CSA in males,[20,23,25,32,42] while demonstrating an absence of sex differences in a range of other tendons.

Interestingly, when the regression models did not include donor height and weight as covariates, generalized sex differences were found for all outcomes except for UTS (ie, differences observed for CSA, maximum load, strain at failure, elastic modulus; all $P < .05$), with sex differences again primarily affecting the Achilles tendon (differences observed for CSA, maximum load, and strain at failure; all $P < .05$). Anthropometric measurements were a part of the regression models to explore whether any sex differences identified were related to more physical versus intrinsic biological differences. When body mass index

(BMI) was initially included as a covariate, it was not predictive of any outcome (all $P > .05$). However, when donor height and weight were separate covariates, height became a significant predictor for all outcomes where sex was previously significant (CSA, maximum load, strain at failure, elastic modulus) (Appendix Table A3, available online), and only CSA was significantly affected by height and sex. This suggests that an aspect of the sex differences that we initially observed in the absence of height and weight covariates was driven by the differences in physical size between men and women. Height has been evaluated as a variable of interest only in an in vivo study of nonfailure Achilles tendon properties.[29] While BMI may represent an appropriate variable to control for participant size in human in vivo studies, weight fluctuations during end-of-life events may confound meaningful cadaveric weight and BMI values. Therefore, there may be merit in controlling for height and weight separately within in vitro studies of human cadaveric tissues.

The generalized effects of aging were small but had significant negative effects on tendon UTS and elastic modulus. For perspective, the modeled effect size of age on UTS (Appendix Table A3, available online) was equivalent to a 9.5-MPa decrease in strength over the 50-year age range (49-99 years). In contrast, differences in UTS among anatomic tendons were up to 62.51 MPa, and donor height was more influential than age for the majority of outcomes. Importantly, when tendon-specific changes were evaluated, the effect of age varied, and results were insignificant for most tendons and outcomes (Figure 2). The few significant changes observed were in the three largest tendons (Achilles tendon, tibialis posterior, and tibialis anterior), and the only negative effect of aging was a decrease in the maximum load of the Achilles tendon. This was equivalent to a 17.20-N decrease per year of age (Table 2). In comparison, a 1-inch increase in donor height was equivalent to an 86.40-N increase in maximum load (Table 3). Published results are mixed, with numerous studies reporting no effect of age,[4-6,13,14,39,40] and others finding an inverse relationship,[14,21,24,35,36] although most investigated only the patellar tendon. Of the exceptions, Balsly et al[4] found no significant correlations between age and strength or between age and elastic modulus in irradiated BPTB, tibialis anterior, semitendinosus tendon, or fascia lata. Similarly, Greaves et al[14] indicated that age had no significant effect on failure load, failure strain, UTS, or linear stiffness in nonirradiated single-strand tibialis anterior and posterior tendons, although UTS did decrease with age in nonirradiated double-stranded tendons. In contrast, Lewis and Shaw[24] noted a moderate decrease in Achilles tendon UTS with aging but an insignificant effect on elastic modulus. Swank et al[36] found very weak correlations between aging and all mechanical properties tested in tibialis posterior tendons ($R^2 = 0.012$-0.063). They stated that UTS slightly increased up to the ages of 40 to 49 years then a slightly decreased with further aging. However, the authors concluded that aging explained at most 6% of the variation observed and was unlikely to be clinically relevant. Despite conflicting results from generally restricted cohorts, most surgeons believe that aging decreases tendon strength and negatively affects graft performance,[2] and tissue banks will not accept tendons from donors >65 years.[27] Our results highlight the small and tendon-specific effect of aging on biomechanical properties, especially relative to other donor variables.

A strength of this study was the range of distinct anatomic tendons tested from an extensive human cadaveric cohort, allowing within-study comparisons under the same testing conditions, including test operator. Given the logistical and financial challenges of procuring human cadaveric tissue, human tendon studies generally report on a single or narrow range of specific tendons or small cohorts. This is one of the largest biomechanical studies on human tendon failure properties, consisting of 332 tendons from donors of both sexes spanning an age of 49 to 99 years. This facilitated the investigation of tendon-specific effects of donor variables across a range of tendons, identifying that different tendons are influenced uniquely by donor age, sex, and height. Furthermore, as far as we are aware, no previous studies have revealed an effect of donor height on human tendon tensile failure properties.

Nevertheless, limitations existed. Tendons were tested as unmodified single strands. Autograft and allograft tendons are generally modified to meet the surgical and biomechanical requirements of an ACL graft. Grafts made from larger CSA tendons, such as BPTB and Achilles tendon, are cut down to an appropriate width to fit within the intra-articular space while still possessing a sufficient failure load. Medium-sized tendons, such as tibialis posterior or anterior, are looped into double-stranded constructs, and smaller hamstring tendons are bundled into multi-stranded grafts. As noted by Greaves et al,[14] graft-specific configurations may alter the outcomes of mechanical testing and associated conclusions regarding the effects of age. Another possible limitation is the age range of donors tested, with our youngest donors still considered to be the upper age limit of tissue banks (≤65 years). However, the implication of not including younger donors is unclear. Swank et al[36] examined tibialis anterior tendons from donors aged 15 to 79 years and found that strength started to slowly decline from the age of 50 years, whereas Shelton et al[35] noted a decrease in failure load and failure stress in BPTB from donors aged 15 to 30 versus 51 to 65 years.

Limitations also existed in some of our methodologies. Strain was calculated from the change in crosshead displacement, assuming a homogeneous strain field between the grips. While we excluded specimens that were observed to strain outside the gauge length from strain-based analyses (strain at failure and elastic modulus), there was still the limitation that using this method may under- or overestimate the strain in comparison with the generally more accepted method of using an optically tracked strain. Finally, elastic modulus is often reported as the gradient of the so-called linear region of the stress-strain curve, without specifying how this region is identified. In this study, we used an automated and objective method to calculate the elastic modulus, and we visually confirmed that it was within what we would classify the linear region. To calculate this maximum gradient, we considered using a moving average over a range of data points or fitting

a curve to the data and calculating the gradient. When these 2 possible methods were compared for this data set, they largely produced similar values with similar variances; however, the moving average method occasionally determined the modulus outside of what we would have classified as the linear region and produced slightly higher variability.

CONCLUSION

The findings of this study support the inclusion of a broader range of different tendons for use as allografts for ACL reconstruction and warrant further investigation to evaluate these new allograft candidates tested as graft constructs. Donor sex, age, and height had relatively small effects on mechanical properties, but, most important, they were limited to specific tendons (Achilles tendon, tibialis posterior, tibialis anterior) and affected these tendons in different ways, demonstrating that the effects of donor variables should not be generalized. Finally, the effect of age negatively affected only the maximum load of Achilles tendon. Together, these results challenge the clinical relevance of using age as an exclusion criterion across all tendons and in isolation to all other donor variables.

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REFERENCES

1. Alva A, Kumar V, Sreekumar R, Ng CY, Talwalkar SC. Autologous tendon grafts used in upper limb surgery. *Open J Orthop*. 2013;3(2):74-78. doi:10.4236/ojo.2013.32014
2. American Academy of Orthopaedic Surgeons. *Allografts for ACL Reconstruction Survey Report*. American Academy of Orthopaedic Surgeons; 2013.
3. Baer GS, Harner CD. Clinical outcomes of allograft versus autograft in anterior cruciate ligament reconstruction. *Clin Sports Med*. 2007;26(4):661-681. doi:10.1016/j.csm.2007.06.010
4. Balsly CR, Cotter AT, Williams LA, Gaskins BD, Moore MA, Wolfinbarger L. Effect of low dose and moderate dose gamma irradiation on the mechanical properties of bone and soft tissue allografts. *Cell Tissue Bank*. 2008;9(4):289-298. doi:10.1007/s10561-008-9069-0
5. Blevins FT, Hecker AT, Bigler GT, Boland AL, Hayes WC. The effects of donor age and strain rate on the biomechanical properties of bone–patellar tendon–bone allografts. *Am J Sports Med*. 1994;22(3):328-333. doi:10.1177/036354659402200306
6. Castile RM, Cannon PC, Smith MV, Brophy RH, Lake SP. Donor age and sex have limited effects on the mechanical and microstructural properties of human connective tissues. *J Orthop Res*. 2022;40(8):1844-1852. doi:10.1002/jor.25200
7. Chandrashekar N, Mansouri H, Slauterbeck J, Hashemi J. Sex-based differences in the tensile properties of the human anterior cruciate ligament. *J Biomech*. 2006;39:2943-2950. doi:10.1016/j.jbiomech.2005.10.031
8. Chang SKY, Egami DK, Shaieb MD, Kan DM, Richardson AB. Anterior cruciate ligament reconstruction: allograft versus autograft. *Arthroscopy*. 2003;19(5):453-462. doi:10.1053/jars.2003.50103
9. Choi RK, Smith MM, Martin JH, et al. Chondroitin sulphate glycosaminoglycans contribute to widespread inferior biomechanics in tendon after focal injury. *J Biomech*. 2016;49(13):2694-2701. doi:10.1016/j.jbiomech.2016.06.006
10. Duchman KR, Lynch TS, Spindler KP. Graft selection in anterior cruciate ligament surgery: who gets what and why? *Clin Sports Med*. 2017;36(1):25-33. doi:10.1016/j.csm.2016.08.013
11. Edgar CM, Zimmer S, Kakar S, Jones H, Schepsis AA. Prospective comparison of auto and allograft hamstring tendon constructs for ACL reconstruction. *Clin Orthop Relat Res*. 2008;466(9):2238-2246. doi:10.1007/s11999-008-0305-5
12. Faul F, Erdfelder E, Buchner A, Lang AG. Statistical power analyses using G*Power 3.1: tests for correlation and regression analyses. *Behav Res Methods*. 2009;41(4):1149-1160. doi:10.3758/BRM.41.4.1149
13. Flahiff CM, Brooks AT, Hollis JM, Vander Schilden JL, Nicholas RW. Biomechanical analysis of patellar tendon allografts as a function of donor age. *Am J Sports Med*. 1995;23(3):354-358. doi:10.1177/036354659502300319
14. Greaves LL, Hecker AT, Brown CH. The effect of donor age and low-dose gamma irradiation on the initial biomechanical properties of human tibialis tendon allografts. *Am J Sports Med*. 2008;36(7):1358-1366. doi:10.1177/0363546508314394
15. Grevnerts HT, Fältström A, Sonesson S, Gauffin H, Carlfjord S, Kvist J. Activity demands and instability are the most important factors for recommending to treat ACL injuries with reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2018;26(8):2401-2409. doi:10.1007/s00167-018-4846-1
16. Guyton GP, Jeng C, Krieger LE, Mann RA. Flexor digitorum longus transfer and medial displacement calcaneal osteotomy for posterior tibial tendon dysfunction: a middle-term clinical follow-up. *Foot Ankle Int*. 2001;22(8):627-632. doi:10.1177/107110070102200802
17. Hashemi J, Chandrashekar N, Slauterbeck J. The mechanical properties of the human patellar tendon are correlated to its mass density and are independent of sex. *Clin Biomech (Bristol, Avon)*. 2005;20:645-652. doi:10.1016/j.clinbiomech.2005.02.008
18. He J, Tang Q, Ernst S, et al. Peroneus longus tendon autograft has functional outcomes comparable to hamstring tendon autograft for anterior cruciate ligament reconstruction: a systematic review and meta-analysis. *Knee Surg Sports Traumatol Arthrosc*. 2021;29(9):2869-2879. doi:10.1007/s00167-020-06279-9
19. Hulet C, Sonnery-Cottet B, Stevenson C, et al. The use of allograft tendons in primary ACL reconstruction. *Knee Surg Sports Traumatol Arthrosc*. 2019;27(6):1754-1770. doi:10.1007/s00167-019-05440-3
20. Intziegianni K, Cassel M, Hain G, Mayer F. Gender differences of Achilles tendon cross-sectional area during loading. *Sports Med Int Open*. 2017;1(4):e135-e140. doi:10.1055/s-0043-113814
21. Johnson GA, Tramaglini DM, Levine RE, Ohno K, Choi NY, Woo SLY. Tensile and viscoelastic properties of human patellar tendon. *J Orthop Res*. 1994;12(6):796-803. doi:10.1002/jor.1100120607
22. Josipović M, Vlaić J, Serdar J, et al. Plantaris tendon: a novel graft for anterolateral ligament reconstruction and additional reinforcement for anterior cruciate ligament autografts in combined reconstructive procedures. *Knee Surg Sports Traumatol Arthrosc*. 2020;28(8):2604-2608. doi:10.1007/s00167-020-05885-x
23. Lepley AS, Joseph MF, Daigle NR, et al. Sex differences in mechanical properties of the Achilles tendon: longitudinal response to repetitive loading exercise. *J Strength Cond Res*. 2018;32(11):3070-3079. doi:10.1519/JSC.0000000000002386

24. Lewis G, Shaw KM. Tensile properties of human tendo Achillis: effect of donor age and strain rate. *J Foot Ankle Surg*. 1997;36(6):435-445. doi:10.1016/S1067-2516(97)80096-8
25. Magnusson SP, Hansen M, Langberg H, et al. The adaptability of tendon to loading differs in men and women. *Int J Exp Pathol*. 2007;88(4):237-240. doi:10.1111/j.1365-2613.2007.00551.x
26. Maletis GB, Inacio MCS, Funahashi TT. Analysis of 16,192 anterior cruciate ligament reconstructions from a community-based registry. *Am J Sports Med*. 2013;41(9):2090-2098. doi:10.1177/0363546513493589
27. Mauro CS, Rihn JA, Harner CD. Safety issues for musculoskeletal allografts. In: Johnson DL, Mair SD, eds. *Clinical Sports Medicine*. Mosby; 2006:111-116. doi:10.1016/B978-032302588-1.50015-4
28. Meter J, Bascharon R, Amesur A, et al. Revision Achilles tendon repair using posterior tibial tendon allograft and flexor hallucis longus transfer. *Arthrosc Tech*. 2022;11(3):e359-e363. doi:10.1016/j.eats.2021.11.001
29. Morrison SM, Dick TJM, Wakeling JM. Structural and mechanical properties of the human Achilles tendon: sex and strength effects. *J Biomech*. 2015;48(12):3530-3533. doi:10.1016/j.jbiomech.2015.06.009
30. Noyes FR, Butler DL, Grood ES, Zernicke RF, Hefzy MS. Biomechanical analysis of human ligament grafts used in knee-ligament repairs and reconstructions. *J Bone Joint Surg Am*. 1984;66(3):344-352.
31. Pagenstert G, Valderrabano V, Hintermann B. Lateral ankle ligament reconstruction with free plantaris tendon graft. *Tech Foot Ankle Surg*. 2005;4:104-112. doi:10.1097/01.btf.0000152574.09654.80
32. Pardes AM, Freedman BR, Fryhofer GW, Salka NS, Bhatt PR, Soslowsky LJ. Males have inferior Achilles tendon material properties compared to females in a rodent model. *Ann Biomed Eng*. 2016;44(10):2901-2910. doi:10.1007/s10439-016-1635-1
33. Pennock AT, Ho B, Parvanta K, et al. Does allograft augmentation of small-diameter hamstring autograft ACL grafts reduce the incidence of graft retear? *Am J Sports Med*. 2017;45(2):334-338. doi:10.1177/0363546516677545
34. Raines BT, Naclerio E, Sherman SL. Management of anterior cruciate ligament injury: what's in and what's out? *Indian J Orthop*. 2017;51(5):563-575. doi:10.4103/ortho.IJOrtho_245_17
35. Shelton TJ, Delman C, McNary S, Taylor JR, Marder RA. Aging decreases the ultimate tensile strength of bone–patellar tendon–bone allografts. *Arthroscopy*. 2021;37(7):2173-2180. doi:10.1016/j.arthro.2021.02.042
36. Swank KR, Behn AW, Dragoo JL. The effect of donor age on structural and mechanical properties of allograft tendons. *Am J Sports Med*. 2015;43(2):453-459. doi:10.1177/0363546514557246
37. Thermann H, Frerichs O, Biewener A, Krettek C, Schandelmaier P. Biomechanical studies of human Achilles tendon rupture. Article in German. *Unfallchirurg*. 1995;98(11):570-575.
38. Widner M, Dunleavy M, Lynch S. Outcomes following ACL reconstruction based on graft type: are all grafts equivalent? *Curr Rev Musculoskelet Med*. 2019;12(4):460-465. doi:10.1007/s12178-019-09588-w
39. Wilson TW, Zafuta MP, Zobitz M. A biomechanical analysis of matched bone-patellar tendon-bone and double-looped semitendinosus and gracilis tendon grafts. *Am J Sports Med*. 1999;27(2):202-207. doi:10.1177/03635465990270021501
40. Yanke A, Bell R, Lee A, Shewman EF, Wang V, Bach BR. Regional mechanical properties of human patellar tendon allografts. *Knee Surg Sports Traumatol Arthrosc*. 2015;23(4):961-967. doi:10.1007/s00167-013-2768-5
41. Zbrojkiewicz D, Vertullo C, Grayson JE. Increasing rates of anterior cruciate ligament reconstruction in young Australians, 2000-2015. *Med J Aust*. 2018;208(8):354-358. doi:10.5694/mja17.00974
42. Zhang X, Deng L, Xiao S, Li L, Fu W. Sex differences in the morphological and mechanical properties of the Achilles tendon. *Int J Environ Res Public Health*. 2021;18(17):8974. doi:10.3390/ijerph18 178974