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Mechanical properties and collagen cross-linking of the patellar tendon in old and young men

C. Couppé,[1,2] P. Hansen,[1] M. Kongsgaard,[1] V. Kovanen,[3] C. Suetta,[1] P. Aagaard,[4] M. Kjær,[1] and S. P. Magnusson[1,2]

[1]*Institute of Sports Medicine, Bispebjerg Hospital and Center for Healthy Aging, Faculty of Health Sciences, University of Copenhagen, Copenhagen, Denmark;* [2]*Department of Physical Therapy, Bispebjerg Hospital, Copenhagen, Denmark;* [3]*Department of Health Sciences, University of Jyväskylä, Jyväskylä, Finland; and* [4]*The University of Southern Denmark, Odense, Denmark*

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Couppé C, Hansen P, Kongsgaard M, Kovanen V, Suetta C, Aagaard P, Kjær M, Magnusson SP. Mechanical properties and collagen cross-linking of the patellar tendon in old and young men. *J Appl Physiol* 107: 880–886, 2009. First published June 25, 2009; doi:10.1152/japplphysiol.00291.2009.—Age-related loss in muscle mass and strength impairs daily life function in the elderly. However, it remains unknown whether tendon properties also deteriorate with age. Cross-linking of collagen molecules provides structural integrity to the tendon fibrils and has been shown to change with age in animals but has never been examined in humans in vivo. In this study, we examined the mechanical properties and pyridinoline and pentosidine cross-link and collagen concentrations of the patellar tendon in vivo in old (OM) and young men (YM). Seven OM (67 ± 3 years, 86 ± 10 kg) and 10 YM (27 ± 2 years, 81 ± 8 kg) with a similar physical activity level (OM 5 ± 6 h/wk, YM 5 ± 2 h/wk) were examined. MRI was used to assess whole tendon dimensions. Tendon mechanical properties were assessed with the use of simultaneous force and ultrasonographic measurements during ramped isometric contractions. Percutaneous tendon biopsies were taken and analyzed for hydroxylysyl pyridinoline (HP), lysyl pyridinoline (LP), pentosidine, and collagen concentrations. We found no significant differences in the dimensions or mechanical properties of the tendon between OM and YM. Collagen concentrations were lower in OM than in YM (0.49 ± 0.27 vs. 0.73 ± 0.14 mg/mg dry wt; $P < 0.05$). HP concentrations were higher in OM than in YM (898 ± 172 vs. 645 ± 183 mmol/mol; $P < 0.05$). LP concentrations were higher in OM than in YM (49 ± 38 vs. 16 ± 8 mmol/mol; $P < 0.01$), and pentosidine concentrations were higher in OM than in YM (73 ± 13 vs. 11 ± 2 mmol/mol; $P < 0.01$). These cross-sectional data raise the possibility that age may not appreciably influence the dimensions or mechanical properties of the human patellar tendon in vivo. Collagen concentration was reduced, whereas both enzymatic and nonenzymatic cross-linking of concentration was elevated in OM vs. in YM, which may be a mechanism to maintain the mechanical properties of tendon with aging.

tendon dimension; tendon mechanical properties; aging; collagen; hydroxylysyl pyridinoline; lysyl pyridinoline; advanced glycation end products

FORCE GENERATED BY MUSCLE is transferred to bones via tendons to produce movement. However, tendons are not entirely inextensible, but exhibit elastic and time-dependant properties that serve to influence the overall function of the muscle-tendon complex (3, 13, 17, 29, 58). Tendons have traditionally been considered relatively inert structures, but several recent reports have demonstrated that human tendons respond directly to physical activity by increased metabolic activity (15, 35, 42) and increased collagen synthesis (50, 51). Furthermore, strength training and habitual loading of tendons appear to be associated with increased tendon size (6, 21, 46), confirming that the aforementioned response to elevated loading results in a net increase of tendon tissue. These recent findings show that tendons respond and adapt to their specific loading history.

Aging is associated with a decline in muscle mass, strength, and physical function (66). However, although tendon properties influence the overall function of the muscle-tendon complex (58, 67), there is a relative lack of human data that describe possible age-associated changes in mechanical properties of tendon. Animal data show that aging yields a stronger and stiffer tendon (34, 69, 83), a weaker and more compliant tendon (27, 90, 91), or leaves the tendon unchanged (39); thus these data are inconclusive. In contrast, data on isolated human cadaver tendon suggest that the aging process largely leaves the mechanical properties unaltered (31, 40, 41). Despite the development of ultrasonography-based methods to evaluate human tendon properties in vivo (33, 37, 53), the effect of aging on the mechanical properties of human tendon in vivo remains elusive. Tendon strain in humans has been shown to decrease (47–49) or increase (43, 52, 61, 63, 70) with aging. Yet others have shown that aging leaves the mechanical properties of the patellar tendon unchanged (18). Thus the picture is incoherent, which may partly be related to methodological and design differences, the physical activity level of the sample population, and the type of tendon tested, i.e., the tendon-aponeurosis complex or the free tendon alone.

In tendon, the trivalent intermolecular pyridinoline cross-links [primarily hydroxylysyl pyridinoline (HP) and lysyl pyridinoline (LP)] stabilize the fibrillar structure of collagen and thus contribute to the mechanical properties of the tendon (7, 9, 12). These cross-links are formed from enzymatically derived covalent immature cross-links, which undergo a spontaneous conversion into more mature trivalent cross-links with collagen maturation (7). The slow turnover of mature collagen allows further cross-linking via the adventitious nonenzymatic reactions of glucose with the lysyl and arginine amino acid residues in the collagen triple helix as a true aging process (7, 59, 62). This nonenzymatic process results in the accumulation of advanced glycation end products (AGE) in tendon tissue. The most widely studied AGE is pentosidine. AGE accumulation is known to accelerate with aging and diabetes (16, 28, 62) and is believed to yield a stiffer and more load-resistant tendon (78, 79). It has been shown that AGE cross-link density in collag-

Address for reprint requests and other correspondence: S. P. Magnusson, Institute of Sports Medicine Copenhagen, Bispebjerg Hospital, Bldg. 8, 1st Floor, Bispebjerg Bakke 23, 2400 Copenhagen NV, Denmark (e-mail: P.Magnusson@mfi.ku.dk).

enous tissues (19, 38, 81, 86) and in human tendons (11, 84) is markedly higher in older than in younger individuals, and there are only sparse human data on mature cross-link density of tendon and how these are influenced by aging (11). However, recent reports of an age-associated reduction in human tendon stiffness in vivo (43, 52, 61, 63, 70) in conjunction with the notion of simultaneously elevated cross-link density and AGE accumulation are difficult to reconcile. To the best of our knowledge, there are currently no human studies examining collagen cross-linking and mechanical properties of the patellar tendon in vivo in old and young men. Therefore, the purpose of this study was to examine both the mechanical properties of human patellar tendon in vivo and collagen cross-link composition in old (OM) and young men (YM) subjects.

MATERIALS AND METHODS

Subjects. Seven OM (67 ± 3 years, 86 ± 10 kg) and 10 YM (27 ± 2 years, 81 ± 8 kg) with similar activity levels volunteered for the study (Table 1). With the use of a standardized form, participants were interviewed regarding how many hours per week they were performing organized sport or exercise on a regular basis (OM: 5 ± 6 h/wk, YM: 5 ± 2 h/wk). There were no differences in physical characteristics between OM and YM. All were healthy, did not take prescription medicine, and had no overt signs or symptoms of diabetes. In addition, the subjects had no known knee or tendon pathology. The study complied with the Declaration of Helsinki and was approved by the local ethics committee. All subjects gave their informed consent before the experiments.

Muscle and tendon dimensions. The anatomic cross-sectional area (CSA) of the quadriceps femoris muscle was measured 20 cm proximal to the tibia plateau (mid-thigh level) by magnetic resonance imaging (MRI) (General Electric, Sigma Horizon LX 1.5-Tesla, T1 weighted SE) using a lower extremity coil. The images were obtained using the following parameters: TR/TE = 500/14 ms, field of view (FOV) = 18, matrix = 512 × 512, and slice thickness = 6 mm (21, 46). Subsequently, the lean muscle mass of the quadriceps muscle (subcutaneous and intermuscular noncontractile tissues were excluded from the measurement) was manually outlined using the software program Osiris 4.19 (http://www.sim.hcuge.ch/osiris/). The mean value of three measurements of the same image was used for analysis. Patellar tendon CSA and length were determined with the use of MRI (General Electric, Sigma Horizon LX, 1.5-Tesla, T1 weighted SE) (21, 46). Patellar tendon CSA was determined by axial plane MRI using the following parameters: TR/TE = 400/14 ms, FOV = 20, matrix = 256 × 256, slice thickness = 5.0 mm, and spacing = 0 mm. The axial scans were performed perpendicular to the patellar tendon. As described in detail previously, the tendon CSA was measured *1*) just distal to the patellar insertion, *2*) just proximal to the tibia insertion, and *3*) midway between these two sites (21, 46). The patellar tendon length was determined from sagittal plane MRI using the following parameters: TR = 500, echo time (ET) = 3 × (TE: 12.4 ms), FOV = 16, matrix = 256 × 192, slice thickness = 4.0 mm, and no spacing. The patellar tendon length was obtained by measuring the distance from the dorsal insertion at the patella apex to the dorsal insertion on the tibia. Patellar tendon CSA and length were manually outlined using the software program Osiris 4.19 (http://www.sim.hcuge.ch/osiris/). The color intensity of each image was adjusted using the National Institutes of Health color scale mode of the software. Tendon CSA and length were measured using the grayscale image display. The average tendon CSA was calculated from the three levels (proximal, mid, and distal CSA) and used for analysis. The typical error percent of repeated measures of site-specific tendon CSA was 2–2.5% (21).

Table 1. *Physical characteristics of OM and YM*

	OM (n = 7)	YM (n = 10)
Age, years	67±3	27±2
Height, cm	178±9	183±4
Weight, kg	86±10	81±8
Activity level, h/wk	5±6	5±2

Values are means ± SD. There were no differences in physical characteristics between old men (OM) and young men (YM).

Mechanical properties of tendon. The details of the measurement, including the reliability of the method in our laboratory, has been reported previously (37). The within-day correlation coefficient and typical error percent results for repeated measures were 0.95 and 9.9% for tendon stiffness, 0.97 and 5.5% for tendon strain, and 0.94 and 9.4% for Young's modulus. Subjects performed a 5-min warm-up on a stationary bike to secure proper preconditioning of the tendon before testing. Thereafter, the subjects were seated in a custom-made rigid chair with both hips and knees flexed to an angle of 90°. A leg cuff, which was connected to a strain gauge (Bofors KRG-4, Bofors, Sweden) through a rigid steel rod perpendicular to the lower leg, was mounted on the leg just above the medial malleolus. An ultrasound probe (7.5 MHz, linear array B-mode; Sonoline Sienna, Siemens, Erlangen, Germany) was fitted into a custom-made rigid cast that was secured to the skin above the patellar tendon in the sagittal plane. The ultrasound probe and cast were positioned so that the patella, the patellar tendon, and the tibia were all visible within the viewing field throughout the ramped contractions (Fig. 1).

The ultrasound S-VHS video images obtained during the ramp trials were sampled at 50 Hz on a personal computer using frame-by-frame capturing software (Matrox Marvel G400-TV, Dorval, Quebec, Canada). Force was sampled on two separate personal computers at 50 Hz via a 12-bit analog-to-digital converter (dt2810A; Data Translation). The two computers were interconnected to permit synchronous sampling of all data using a custom-built trigger device (14). The subjects performed four to five slow isometric knee extensions ramps by applying gradually increasing force until maximum over a 10-s period during which patellar tendon displacement and knee extension force were measured simultaneously. Each ramp was separated by a 2-min rest period. All measurements were performed on one side, randomized to either the right or left knee. During the ramp contractions, force was sampled at 50 Hz and low-pass filtered at a 1.0-Hz cutoff frequency using a fourth-order zero-lag Butterworth filter.

Tendon force was calculated by dividing the estimated total knee extension moment by the internal moment arm, which was estimated from individually measured femur lengths (87). Tendon stress was calculated by dividing tendon force with the average of the three levels (proximal, mid, and distal) of the patellar tendon CSA determined from MRI. Tendon deformation was defined as the change in distance between the patellar apex and the tibia (37, 57). Tendon strain was calculated as the change in length related to the initial tendon length. Each single force-deformation curve was fitted to a second- or third-order polynomial fit, which yielded $R^2 > 0.98$. Tendon stiffness (Δforce/Δdeformation) and Young's modulus (Δstress/Δstrain) based on common force were calculated in the final 20% of the force-deformation and stress-strain curves, respectively (57). To compare tendon dimensions between the subjects of various body size, tendon CSA data were normalized to body weight and raised to the power of 2/3 (60).

Patellar tendon biopsies. A Bard MAGNUM biopsy instrument (C.R. Bard, Covington, GA) with a disposable core biopsy needle (14 gauge) was used. After sterilization, the skin was injected with local anesthetic (1% lidocaine), and a 3- to 5-mm-long incision was created just distal to the patella apex. The biopsy needle was inserted into the tendon surface at an ~30° angle and fired, securing a tissue sample of ~8 mg. Samples were snap-frozen in liquid nitrogen and stored at −80°C. Tendon biopsies were taken from the same side as tendon mechanical properties assessments were performed. No previous bi-

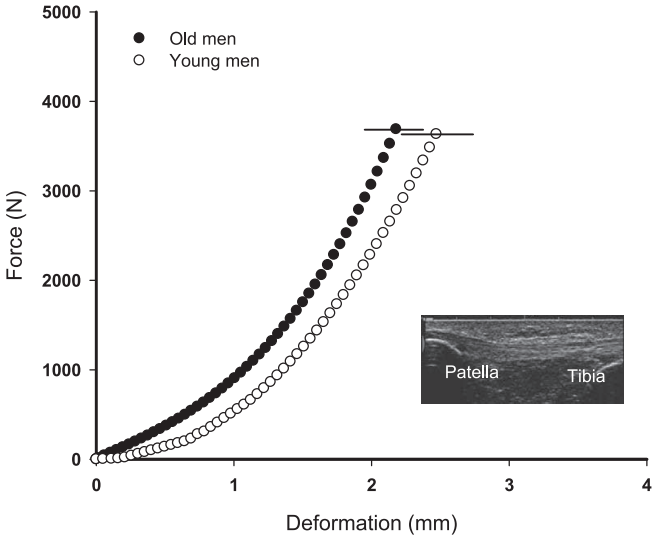

Fig. 1. The patellar tendon force-deformation relationship based on common force. Values are means ± SD of all subjects. There were no differences between old (OM) and young men (YM) with respect to tendon deformation or stiffness ($P > 0.05$).

opsy had been taken from that site in all subjects. All biopsy samples were analyzed in an investigator-blinded fashion.

Biochemical analysis. Freeze-dried tendon samples were hydrolyzed in 6 M HCl (+108 C, 24 h) and evaporated into dryness and dissolved in H_2O. Hydroxyproline, the collagen-specific amino acid, was measured spectrophotometrically (23) to quantify collagen protein (24). HP, LP, and pentosidine were analyzed via a single reversed-phase high-performance liquid chromatography (HPLC) run and detected on the basis of their natural fluorescence (10). At 0–16 min, the wavelength for HP and LP fluorescence was 400 nm for emission and 295 nm for excitation. The wavelengths were changed at 16–60 min to 328/378 nm to measure pentosidine. For the elution of the cross-links, a gradient was built up to contain 17% eluent B (75% acetonitrile with 0.13% heptafluorobutyric anhydride) at 0 min and 25% eluent B at 30 min. Eluent A was 0.13% heptafluorobutyric anhydride. Flow rate was 1 ml/min. HP was eluted at 12 min, LP at 13.5 min, and pentosidine at 23 min. The HPLC system used included Quaternary Gradient Pump unit, PU-2089 Plus, Intelligent AutosamplerAS-2057 Plus, and Intelligent Fluorescence Detector FP-2020 by Jasco. Data processing software was Jasco Chrompass. The LiChroCART 125-4 column was from Merck Hitachi. The results for HP, LP, and pentosidine are given compared with the standards injected at four different concentrations in each HPLC run. The intra-assay coefficient of variations based on duplicates within a run were 2.6%, 3.7%, and 3.9% for HP, LP, and pentosidine, respectively. The detection limit for HP and LP is 0.4 pmol and 0.05 pmol for pentosidine.

Data reduction and analysis. The two isometric ramp contractions that yielded the greatest maximum force were selected for further analysis. To make group comparisons and thereby account for differences in magnitude in isometric ramp contraction force, the trials for all subjects were subsequently analyzed to the lowest common force, as determined by the weakest subject (3953 N). Mann-Whitney *U*-tests were used to examine whether there were differences between the groups in the measured variables. Spearman rank-order correlation was used to analyze the strength of relationships between variables. $P < 0.05$ was considered significant. Results are reported as means ± SD.

RESULTS

Peak knee extensor moment was lower in OM (154 ± 41 N·m) than in YM (216 ± 62 N·m) ($P < 0.05$). Similarly, quadriceps femoris CSA was lower in OM than in YM (Table 2) ($P < 0.01$).

Table 2. *Patellar tendon dimensions and quadriceps muscle CSA for OM and YM*

	OM (n = 7)	YM (n = 10)
Tendon length, mm	43±4	43±5
Tendon CSA, mm^2	101±17	103±8
Tendon CSA, mm^2/body wt$^{2/3}$	5.34±0.48	5.47±0.25
Quadriceps muscle CSA, mm^2	6,214±705*	8,431±522

Values are means ± SD. CSA, cross-sectional area. *Significantly different from YM, $P < 0.01$.

Tendon dimensions are shown in Table 2. Tendon length did not differ between YM and OM. Absolute average tendon CSA did not differ between OM and YM. Similarly, there was no between group difference in average tendon CSA normalized for body weight.

Mechanical properties determined at maximal force are shown in Table 3. Maximal tendon force was lower in OM than in YM ($P < 0.05$). There were no differences between OM and YM with respect to tendon deformation, stiffness, strain, stress, or Young's modulus based on average tendon CSA. Mechanical properties at a common force are shown in Table 4. Again, there were no differences between OM and YM for any of the variables (Figs. 1 and 2).

Collagen concentration and cross-link density data are shown in Table 5. Collagen concentration was lower in OM than in YM ($P < 0.05$). Both HP and LP ($P < 0.05$) as well as pentosidine ($P < 0.01$) concentrations in collagen were higher in OM than in YM. Pentosidine was positively related to age in YM ($r = 0.74$, $P < 0.01$, Fig. 3) but not in OM ($r = 0.65$, $P = 0.11$). There were no significant correlations between the mechanical and biochemical variables.

DISCUSSION

To the best of our knowledge, this is the first study that has examined both the mechanical properties of the human patellar tendon in vivo and collagen cross-link densities in YM and OM with similar physical activity levels. The main findings were that the collagen concentration was lower in OM than in YM, whereas the enzymatically derived cross-links (HP and LP) were greater in OM than in YM. At the same time, the nonenzymatically derived AGE marker (pentosidine) was markedly more abundant in OM than in YM. However, despite these apparent age-related differences in the tendon collagen properties, the tendon mechanical properties in the two age groups did not diverge appreciably.

It is well known that age is associated with a loss in muscle mass and consequently a reduction in muscle function (66). It

Table 3. *Patellar tendon mechanical properties for OM and YM based on maximum force*

	OM (n = 7)	YM (n = 10)
Force, N	5,161±737*	7,415±2184
Deformation, mm	2.6±0.4	2.9±0.9
Stiffness, N/mm	3,926±1091	5,546±1871
Stress, MPa	51±8	65±24
Strain, %	6.1±0.9	6.9±2.3
Modulus, GPa	1.7±0.3	2.2±0.7

Values are means ± SD. *Significantly different from YM, $P < 0.05$.

Table 4. *Patellar tendon mechanical properties for OM and YM based on common force*

	OM (n = 7)	YM (n = 10)
Deformation, mm	2.3±0.4	2.4±0.6
Stiffness, N/mm	3,511±837	3,290±869
Stress, MPa	41±7	37±5
Strain, %	5.3±0.9	5.9±1.7
Modulus, GPa	1.5±0.4	1.4±0.4

Values are means ± SD.

Table 5. *Concentration of collagen and hydroxylysyl pyridinoline, lysyl pyridinoline, and pentosidine cross-links in the patellar tendons of OM and YM*

	OM (n = 7)	YM (n = 10)
Collagen, mg/mg dry wt	0.49±0.27*	0.73±0.14
Hydroxylysyl pyridinoline, mmol/mol collagen	898±172*	645±183
Lysyl pyridinoline, mmol/mol collagen	49±38†	16±8
Pentosidine, mmol/mol collagen	73±13†	11±2

Values are means ± SD. *,†Significantly different from the values of YM ($P < 0.05$ and $P < 0.01$, respectively).

has also been suggested that tendon compliance and electromechanical delay increase with aging (80), which would reduce efficient transfer of contractile force and therefore amplify the age-associated decline in muscle function. However, to what extent the mechanical properties of human tendon change with aging remains unclear. Data based on animal models are disjointed as they suggest that aging reduces (27, 90, 91), augments (34, 69, 83), or leaves the mechanical properties unchanged (39). Similarly, data based on human in vivo models are inconclusive. These studies show that, with aging, the tendon becomes more compliant (43, 52, 61, 63, 70), less compliant (47–49), or remains unchanged (18, 43). In contrast, data on isolated human cadaver preparations consistently suggest that aging does not influence the mechanical properties (31, 40, 41), which is in agreement with the present data and that of Carroll et al. (18) on human patellar tendon in vivo.

The present mechanical data diverge from those of others based on the human in vivo model with one exception (18), and this may be related to methodological differences and study design. It has been shown that strength training and habitual loading pattern may result in tendon hypertrophy (6, 21, 46), which would influence the mechanical properties of the tendon (80). In contrast to previous in vivo studies (18, 43, 47, 52, 61, 63, 70), we have taken this aspect into account by comparing age groups with similar activity levels. However, it cannot be ruled out that prior life-long training history, including exercise mode and intensity, which was unaccounted for in the study, may have influenced the data. Furthermore, it has been shown that the ultrasonography-based method of obtaining patellar tendon deformation requires that the movement of the tibia also has to be considered (37, 71), which was achieved in the present study and in the study by Carroll et al. (18). Notably, the present data and that of Carroll et al. (18) cannot demonstrate any age-associated difference in mechanical properties of the patellar tendon. Moreover, several studies have investigated the effect of aging on the mechanical properties based on a composite measure of deformation that includes both that of the tendon and aponeurosis, rather than the tendon per se (43, 52, 61, 63, 70). This makes direct comparisons of results difficult since the free tendon and aponeurosis have dissimilar mechanical properties and because the stiffness of the aponeurosis can be modulated during contraction (30, 57). It should also be recognized that the present data and that of others are based on cross-sectional designs with inherent limitations, including the striking variations in tendon mechanical properties between subjects (54); therefore, a type II error cannot be ruled out.

The densities of mature lysyl oxidase-derived intermolecular covalent cross-links, such as HP and LP, gradually increase during tendon tissue maturation, and it is commonly believed that these cross-links are the chief contributors to the function and mechanical properties of the tendon (7, 9, 12, 76). In animal models, it has been shown that there is a positive

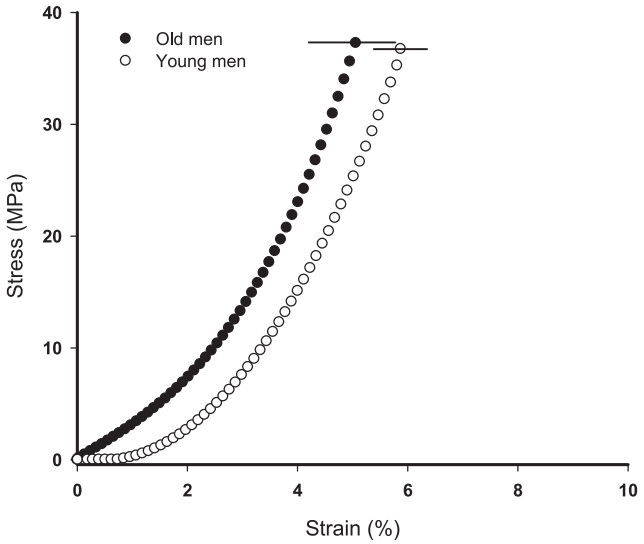

Fig. 2. The patellar tendon stress-strain relationship based on common force. Values are means ± SD of all subjects. There were no differences between OM and YM with respect to tendon strain, stress, or Young's modulus based on average tendon cross-sectional area ($P > 0.05$).

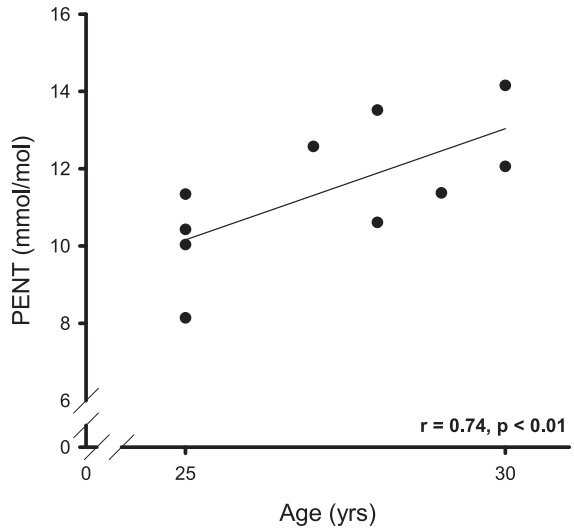

Fig. 3. Pentosidine was positively related to age in YM ($r = 0.74$, $P < 0.01$).

relationship between HP cross-link density and mechanical strength of the healing medial collateral ligament tissue and anterior cruciate ligament graft (32, 68) and furthermore that changes in HP and LP density will result in altered mechanical properties in collagenous tissue (7, 72). However, there are only sparse human data on mature cross-link density in tendon and how these are influenced by aging (11). A small but significant age-related increase of LP has been demonstrated in the supraspinatus tendon in vitro (11). The present data on LP and HP densities of YM correspond well with previously reported data in a similar population (45). However, the present data also demonstrate that HP and LP densities of the human patellar tendon were ~40% and threefold higher, respectively, in OM than in YM, suggesting a rather marked age-associated elevation in these enzymatic cross-links. It is noteworthy that, despite the rather robust difference in both HP and LP, there was no difference in the mechanical properties of the tendon. However, it should be noted that fibril length (22, 77), fibril diameter (74), and proteoglycans and glycosaminoglycans (44, 73, 75) have all been implicated in contributing to the tendon mechanical properties, and the relative contribution of these factors and that of mature cross-links remains elusive.

To our knowledge, these are the first data showing that AGE is markedly increased in the human patellar tendon of OM vs. in that of YM. AGE cross-links, including pentosidine, are formed when lysine amino acid residues in the collagen triple helix come into contact with glucose (7, 62), and the accumulation of these cross-links is known to accelerate with aging (7, 44) and disease processes such as diabetes (16, 28, 62), atherosclerosis (88), Alzheimer (20, 64), and renal failure (89). AGE products are also used as markers of tissue turnover (11). It has been shown that the difference between young and older individuals in AGE cross-link density in collagen is ~2-fold in human skeletal muscle (38), 5-fold in human bone (81), 7-fold in human tendon (present data), 9-fold in human ligaments (19), and 33-fold in human cadaver cartilage (86), demonstrating the tissue-specific turnover. The fact that pentosidine density is sevenfold greater in tendon of OM than in YM (Table 5), coupled with the fact that pentosidine appears to be related to age in a narrow age span (Fig. 3), firmly demonstrates the positive relationship between AGE cross-linking of human patellar tendon collagen and aging ex vivo. These data corroborate and extend those previously reported on cadaver tissue (11, 84). From a functional standpoint, an elevated AGE cross-link density has been suggested to result in increased tensile stress and tendon stiffness in animal models (4, 5, 8, 34, 78, 79, 85). However, in the present study, both the stiffness (Fig. 1) and the Young's modulus (Fig. 2) of the tendon did not differ between OM and YM despite the sevenfold difference in pentosidine, suggesting that factors other than AGE may also play a major role in determining the mechanical properties of human tendon.

In the present study, the total collagen concentration of the patellar tendon was ~34% lower in OM than in YM (Table 5), and this age-linked reduction is in accordance with that found in the canine patellar tendon (39) and rat tail tendon (90). It is possible that the lower collagen concentration with aging may represent the reduced size and/or density of collagen fibrils that is known to occur with aging (26, 65, 74, 76, 82). It was recently reported that MRI signal intensity of the patellar tendon was reduced with aging (18), which may be a function of the reduction in collagen concentration observed in the present study. Unfortunately, the size of the obtained biopsy in the present study precluded transmission electron microscopy analysis for fibril size and density. The lower collagen concentration in OM may be an age-related change in the tendon per se and/or a function of reduced tendon loading due to an age-related loss of muscle mass/strength. Interestingly, the average whole tendon CSA did not differ between OM and YM (Table 2), which in light of the reduced collagen concentration may be related to increased amounts of other extracellular matrix components, such a proteoglycans and glycosaminoglycans. Alternatively, the retained tendon CSA in OM may result from tendon intrafibrillar fat that can accumulate with aging (1, 2, 25). It has previously been demonstrated that the Achilles tendon CSA is larger in postmenopausal women than in young women (55), but it should be noted that female hormones may significantly impact collagen synthesis (36, 56). This potential sex-dependent factor is the reason that we have undertaken the present study in men only.

Albeit speculative, it is possible that the elevated enzymatic and/or nonenzymatic cross-link density in OM vs. that shown in YM served to maintain tendon stiffness and Young's modulus despite the diminished collagen concentration. Such maintenance of tendon stiffness would serve to maintain effective transfer of muscle force despite a lower absolute muscle size (Table 2) and strength (Table 3). In this context, it should be noted that the present data were obtained in moderately physically active individuals, and future studies will need to address the effect of training per se in elderly.

In conclusion, results from the present study raise the possibility that the dimensions and mechanical properties of the human patellar tendon in vivo may not differ between OM and YM. On the other hand, the OM group displayed lower collagen concentration, but greater enzymatic (HP and LP) and nonenzymatic (pentosidine) collagen cross-links, than YM. This age-related increase in both enzymatic and nonenzymatic cross-linking compounds may serve to maintain the mechanical properties of tendon with aging.

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