

STIFFENING EFFECT OF HUMAN TENDONS DURING STRAIN CONTROLLED FATIGUE LOADING

Gábor Szebényi^{1,3}, Dénes Faragó^{2,3}, Rita Kiss M.^{2,3}, Károly Pap⁴

¹ Department of Polymer Engineering, Budapest University of Technology and Economics

² Department of Mechatronics, Optics and Mechanical Engineering Informatics, Budapest University of Technology and Economics

³ Cooperation Research Center for Biomechanics, Budapest University of Technology and Economics

⁴ Orthopedics & Traumatology Department, Uzsoki Hospital

szebényi@pt.bme.hu

DOI: 10.17489/2018/2/09

Abstract

The goals of our study were to evaluate the biomechanical differences between five tendons and the changes in biomechanical properties caused by low cycle fatigue loading. Achilles, quadriceps, semitendinosus + gracilis (STG), tibialis anterior (TA) and the peroneus longus (PL) were harvested from 8 donors. The grafts were removed and placed in a radio-cryoprotectant solution and slowly cooled and stored at -78 °C. The load was defined as a sinusoid function, the starting values are assigned to a peak load of 250 N and a minimum load of 0 N. Data was recorded in the 2th, 4th, 8th, 16th, 32nd, 64th, 128th, 256th, 512th and 1000th cycle. In the given cycles the whole measured waveform was registered. Young modulus of elasticity was calculated. To compare the biomechanical behavior of the different tendons the Young's modulus values were evaluated in the 64th, 128th, 256th, 512th and 1000th cycles. While in case of PL and STG tendons the change is apparently linear in the investigated range, in case of the Achilles, quadriceps and TA tendons there is a region where a significant change in modulus occurs.

Keywords: human tendon; fatigue; Young's modulus; mechanical properties

Introduction

The human tendons are one of the most important parts of our limbs, these help to fix and to move the bones together. The tendons are subjected to a continuous fatigue load during the movement of the body.^{1,2} In the human body, the knee joint is the largest and the most complicated articulation, furthermore it owns the highest incidence of tendons injury. Several articles have been dealt with the mechanical

study of the knee joint.^{1,3,4} After the cyclic tensile loading of the tendons, it can be observed that breaking force and the modulus of elasticity exceeding the pre-load state can be measured, primarily due to the fibrous structure and the orientation of these fibers in the load-bearing orientation.^{2,5,6} Fatigue of the first cross-band of the knee joint represent an increasing proportion in orthopaedic deformations. As a result the surrounding ligaments and muscles become unstable, surgical treatment will be necessary.^{1,4,7,8}

The efficiency of each new surgical method must be demonstrated by tests performed on *in vitro* specimens before introduction. The tendons that are potentially usable to replace the first cross-band of the knee joint should be tested.^{1,3,9} The following tendons are good candidates for substitution: achilles, quadriceps, semitendinosus + gracilis, tibialis anterior, peroneus longus. The question can also be posed as follows: how should tendons be preserved for the *in vitro* examination of different surgical techniques in order that their mechanical properties resemble the characteristics of live tendons the most.²⁴

Fitzgerald stated that the mechanical properties of bones and ligaments change significantly 5 hours post mortem this particularly applies to tensile strengths.^{10,11} Linde and Sorensen measured significant decrease in the Young's modulus 24 hours post mortem by compressive tests on trabecular bones extracted from the tibia.¹²

There are two recommended methods for storage to identify strength characteristics: cooling and freezing. Both methods of storage can be dry or wet. In the case of storage by freezing, the mechanical properties of human and animal ligaments do not change significantly even after 100 days of storage. One of the best conservative solution media for this method is the radio-cryoprotectant solution, which is slowly cooled to -78 degrees Celsius.^{3,10,11,12}

The frozen ligaments must be melted correctly before the test. This is done with one of the controlled melting methods so that the mechanical properties of the tendon do not change. Poor melting process can damage the structure.^{13,14}

The most optimal defrost method after freezing with the radio-cryoprotectant solution is to melt the tendons at 37 degrees Celsius for 20 minutes.^{13,15}

In this study, we investigated the stiffness change of human tendons during strain controlled fatigue loading. Five different tendons were tested as *in vitro* samples, which have been thawed after freezing. We have investigated the Young's modulus change in several selected cycles. Our goal was to investigate how the stiffness of the tendons change with the progressing fatigue loading in case of the different tested samples.

Materials and methods

In the course of our experiments, potential replacements of the first cross-band of the knee joint were tested: achilles, peroneus longus (PL), quadriceps, semitendinosus + gracilis (STG) and tibialis anterior (TA) were studied with a constant elongation amplitude sinusoidal loading during a fatigue test. We have collected the five types of grafts from 8-8 human cadaver tendons within 24 hours post mortem.

The TA, STG, PL tendons were harvested from the musculotendinous junction. All soft tissue including the paratenon was removed from around the tendons. The mid-thirds of the quadriceps and the Achilles tendons were used.

We used only the free ends of the grafts, because of the measurement difficulties, All tendons were visually and mechanically screened for degenerative changes. There was no previous history or evidence of injury or disease of the tendons in the patient's documentation. We have removed and placed the grafts in a radio-cryoprotectant solution and slowly cooled and stored them at -78 °C.

Before the test the ligaments were thawed at 37 °C for 20 minutes and then advanced for 30 seconds before testing. Using a freezer clamp structure, 1000 cycles of 2 Hz frequency sinusoid waveform fatigue were performed on the ligaments using Instron 8872 computer-con-

trolled servo hydraulic tester equipped with an Instron FastTrack 8800 control/data acquisition unit at the accredited material testing laboratory of the BME Biomechanical Co-operation Research Center.

Before starting the load, we have waited for freezing to fix the ligaments. Because of the poor thermal conductivity of the tissue, the freezing effect of the measuring length in the clamps was minimal. Before and during the test, the temperature of the samples was checked using a Flir A325sc infrared (IR) camera. The load was defined as a sinusoid function, the starting values are assigned to a peak load of 250 N and a minimum load of 0 N. Data was recorded with the Instron Fasttrack 8800 data acquisition device in the 2th, 4th, 8th, 16th, 32nd, 64th, 128th, 256th, 512th and 1000th cycle. In the given cycles the whole measured waveform was registered.

An example test setup of the fatigue test is presented in *Figure 1*.



Figure 1. Test setup of the fatigue test

Results and discussion

To compare the biomechanical behavior of the different tendons the Young's modulus values were evaluated in the 64th, 128th, 256th, 512th and 1000th cycles. An example curve set used for the modulus evaluation is presented in *Figure 2*. In the graph it can be seen that as the number of cycles increases, the maximum force increases, resulting in an increase in the inclination of the sinus curve. The shape of the curves is also worth investigating: while the loading strain excitation waveform is a pure sinusoid, in case of the recorded load curves show an asymmetric shape. The positive peak is a standard sinusoid, but the negative peak corresponding to the unloading of the tendon in each cycle is deformed. This can be caused by the viscoelastic behavior of the tendons: the effect of unloading is damped by the soft material with high loss factor.

Comparing the different tendons in the same cycles (*Figure 3 and Figure 4*), similar behavior can be observed showing that the basically identical structure results in also similar mechanical behavior.

The evaluated Young's moduli in function of the elapsed cycles is presented in *Figure 5 and Table 1*. On the summary chart, it can be seen that the orientation of the fibers is clearly present during the fatigue tests. While in case of PL and STG tendons the change is apparently linear in the investigated range, in case of the Achilles, quadriceps and TA tendons there is a region where a significant change in modulus occurs. In case of the Achilles and the quadriceps tendons, which have the largest cross-section this significant increase appears earlier, between the 128th and 256th cycles.

In case of the TA tendon this change occurs later, between the 512th and 1000th cycle. This can be probably explained by the differences in the cross-sections of the tested tendons.

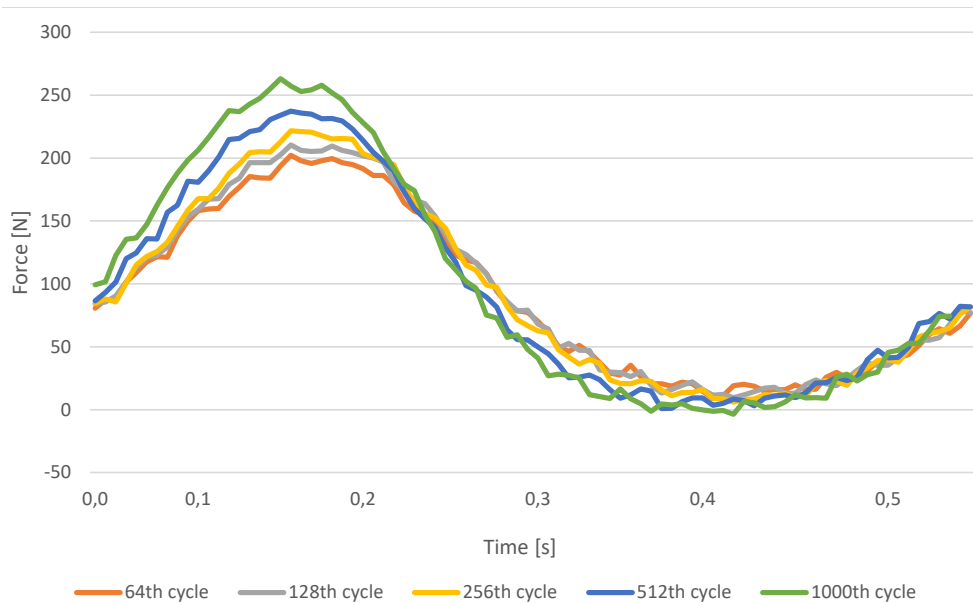


Figure 2. Sample sinus curves of an Achilles tendon in the examined cycles

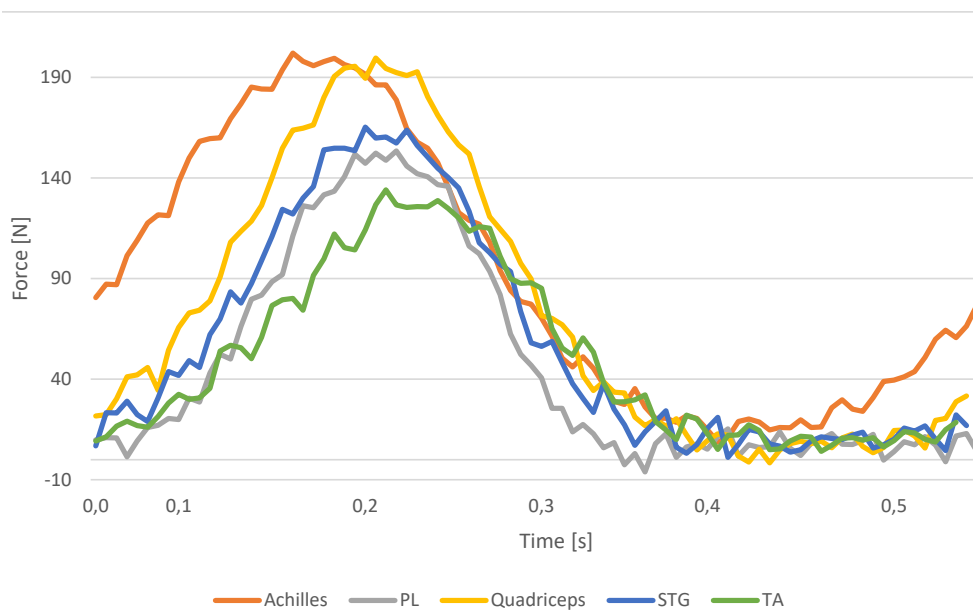


Figure 3. Behavior of different tendons in the 64th cycle

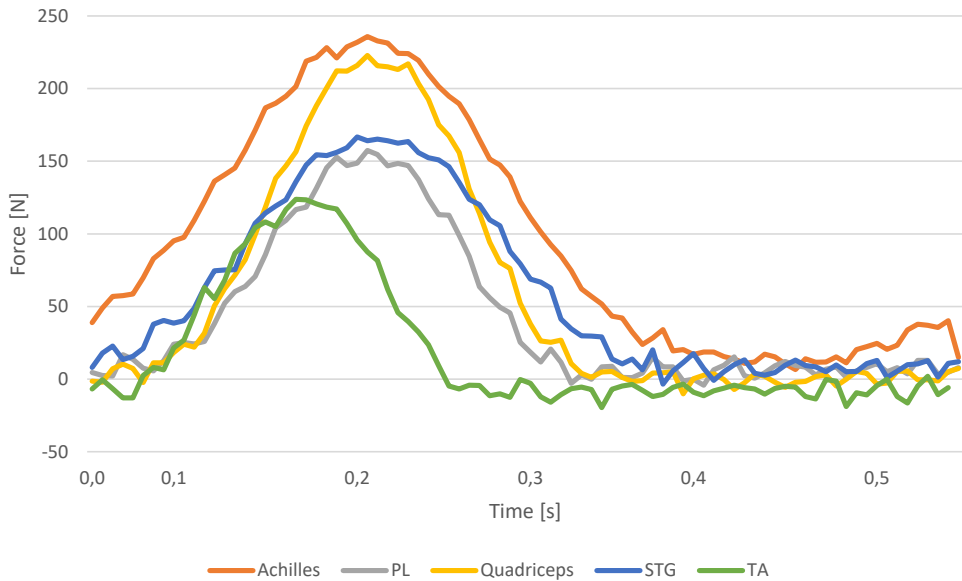


Figure 4. Behavior of different tendons in the 1000th cycle

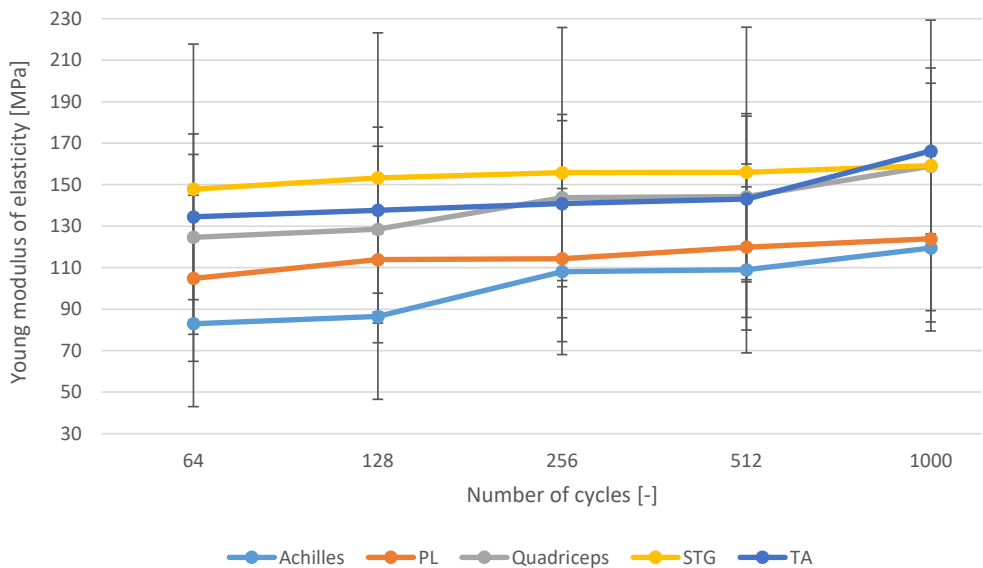


Figure 5. Average Young modulus of elasticity evolution in different cycles for all types of tendons

Tendon type	Achilles	Perosneus longus	Quadiceps	Semitendinosus + Gracilis	Tibialis ant
Cycle number	Young's modulus of elasticity [MPa]				
64	36,1	106,0	124,2	195,1	175,9
	88,9	131,3	108,5	297,6	147,6
	155,8	88,0	57,7	87,3	135,2
	96,7	126,0	125,9	118,2	159,1
	64,1	110,5	102,7	134,1	60,9
	32,3	67,6	185,6	88,2	173,3
	89,1	105,2	167,6	114,2	94,1
	100,6	104,4	124,9	147,9	129,9
Average	82,9	104,9	124,6	147,8	134,5
Deviation	39,6	20,2	39,2	69,8	39,8
128	34,2	130,3	128,9	219,8	131,0
	107,8	142,5	119,3	289,6	158,8
	154,3	102,2	62,8	87,8	193,1
	112,4	114,0	115,9	124,1	169,2
	75,1	118,5	96,5	140,0	76,7
	34,3	75,6	169,2	83,3	122,1
	82,9	115,2	206,5	127,7	111,5
	90,8	112,5	129,2	154,0	139,0
AVERAGE	86,5	113,8	128,5	153,3	137,7
DEVIATION	40,3	19,7	43,6	69,5	36,3
256	43,5	110,2	134,1	220,7	183,2
	120,5	135,8	111,4	293,5	184,7
	173,2	104,3	64,9	85,0	140,6
	120,5	112,4	141,2	127,8	164,8
	173,2	108,5	110,1	136,6	73,4
	120,3	113,8	208,1	95,4	123,6
	77,3	109,8	235,4	126,5	116,6
	36,5	119,8	145,2	160,8	139,9
Average	108,1	114,3	143,8	155,8	140,9
Deviation	52,4	9,8	54,8	69,5	37,3
512	33,8	104,5	151,2	223,0	197,3
	141,1	137,8	139,0	287,9	152,5
	213,8	123,1	109,3	96,2	196,6
	132,9	110,7	153,9	127,2	161,1
	89,4	123,5	154,5	123,4	72,5
	40,3	138,5	150,8	100,1	119,4

Tendon type	Achilles	Perosneus longus	Quadriceps	Semitendinosus + Gracilis	Tibialis ant
Cycle number	Young's modulus of elasticity [MPa]				
	97,2	110,2	140,9	125,0	102,8
	123,1	111,2	154,2	165,2	142,8
Average	109,0	119,9	144,2	156,0	143,1
Deviation	58,2	13,0	15,4	67,1	43,7
1000	30,2	132,9	157,5	212,9	158,4
	165,8	137,5	114,8	285,6	178,0
	202,8	116,3	82,0	86,4	270,6
	148,1	114,0	169,3	133,9	228,4
	70,2	117,6	137,1	128,9	68,6
	94,8	130,8	349,7	123,8	121,1
	123,6	119,2	134,9	142,5	137,0
	120,2	122,8	125,9	160,2	168,2
Average	119,5	123,9	158,9	159,3	166,3
Deviation	54,7	8,7	81,5	62,4	62,5

Table 1. Young modulus of elasticity values in selected cycles for all types of tendons

In case of the wider and thicker tendons with a higher cross-section there are more fiber bundles which can be oriented simultaneously and therefore more easily, providing a more pronounced effect in the increase of the Young's modulus of elasticity.

Further investigations with higher cycle number fatigue tests performed on the STG and PL tendons could further support our assumptions. These tests are currently in progress and will be published in a forthcoming article.

Conclusions

It can be stated that the Young modulus of elasticity of all tendons increased during the cycles by 20-30 MPa, due to the orientation of the fibers. The most significant increase in Achilles was between 128th and 256th cycles, the Quadriceps, PL and STG tendons are increasing continuously, the TA showed the largest modulus increase between 512th and 1000th cycles. These results, extended by microscopic investigations and longer fatigue tests can serve as a basis for the description of the long term fatigue behavior and morphological changes of human tendons.

REFERENCES

1. *Szakály F, Bojtár I, Szabéni G.* Numerical modelling of human ligaments. *Biomechanica Hungarica IX/1.*
2. *Gunther T.* A térdízület biomechanikájának változása az unikompartmentális térdprotézis beültetése során [dissertation]. Budapest (HUN): Semmelweis Univ. 2001.
3. *Szabéni G, Görög P, Török Á, Kiss RM.* Effect of different conservation methods on some mechanical properties of swine bone. *Modelling in Medicine and Biology X 2013: 225-33.*
4. *Hangody Gy, Szabéni G, Abonyi B, Kiss RM, Hangody L, Pap K.* Does a different dose of gamma irradiation have the same effect on five different types of tendon allografts? — a biomechanical study *International Orthopaedics.* Available from: <https://link.springer.com/article/10.1007%2F00264-016-3336-7> 2017. February: 357-65.
5. *Kastelic J, Galeski A, Baer E.* Multicomposite structure of tendon. *Connective Tissue Research 1978;6(1): 11-23.*
6. *Vita RD.* Structural constitutive model for knee Ligaments [dissertation]. Pennsylvania (USA). University of Pittsburgh 2005.
7. *Holzappel GA.* Biomechanics of soft tissue. In: Lemaitre J, editor. *Handbook of materials behaviour models 1.* New York: Academic Press 2000: 1057-75.
8. *Xie F, Yang L, Guo L, Wang Z, Dai G.* A Study on construction three-dimensional nonlinear finite element model and stress distribution analysis of anterior cruciate ligament. *Journal of Biomechanical Engineering 2009;(13112): 121007.*
9. *Weiss JA, Gardiner JC.* Computational modeling of ligament mechanics. *Critical Reviews in Biomedical Engineering 2001;29(3): 303-71.*
10. *Fitzgerald ER.* Dynamic mechanical measurements during the life to death transition in animal tissues. *Biorheology 1975;12:397-408.*
11. *Fitzgerald ER.* Postmortem transition in the dynamic mechanical properties of bone. *Med Phys 1977;4:49-53.*
12. *Linde F and Sorensen HC.* The effect of different storage methods on the mechanical properties of trabecular bone. *Journal of Biomechanics 1993;26: 1249-52.*
13. *Janina Burk et al.* Freeze-Thaw Cycles Enhance Decellularization of Large Tendons *Tissue Engineering, Part C, Methods* [cited 2014 Apr 1]. Available from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3968887/>; 20(4): 276-84.
14. *Delince P, Ghafil D.* Anterior cruciate ligament tears: conservative or surgical treatment? A critical review of the literature. *Knee Surg Sports Traumatol Arthrosc PubMed 2012;20: 48.*
15. *Kuo CK, Marturano JE, Tuan RS.* Novel strategies in tendon and ligament tissue engineering: advanced biomaterials and regeneration motifs. *Sports Med Arthrosc Rehabil Ther Technol 2010;2: 20.*

This research was supported by The National Research, Development and Innovation Office (OTKA K 116189 and NVKP_16-1-2016-0022). Gábor Szabéni acknowledges the financial support received through János Bolyai Scholarship of the Hungarian Academy of Sciences (BO_170_14).

Gábor Szabéni

Department of Polymer Engineering, Faculty of Mechanical Engineering, Budapest University of Technology and Economics

Műgyetem rkp. 3., T. bldg. III., Budapest, Hungary, H-1111

Tel.: (+36) 1 463-1466