

## Low-intensity tensile loading increases intratendinous glucose uptake in the Achilles tendon

J. Bojsen-Møller,<sup>1</sup> K. K. Kalliokoski,<sup>2</sup> M. Seppänen,<sup>2</sup> M. Kjaer,<sup>1</sup> and S. P. Magnusson<sup>1</sup>

<sup>1</sup>*Institute of Sports Medicine, Bispebjerg Hospital, Copenhagen, Denmark; and*

<sup>2</sup>*Turku PET Centre, University of Turku and Turku Central Hospital, Turku, Finland*

Submitted 4 January 2006; accepted in final form 12 March 2006

**Bojsen-Møller, J., K. K. Kalliokoski, M. Seppänen, M. Kjaer, and S. P. Magnusson.** Low-intensity tensile loading increases intratendinous glucose uptake in the Achilles tendon. *J Appl Physiol* 101: 196–201, 2006. First published March 30, 2006; doi:10.1152/jappphysiol.00004.2006.—The metabolic activity of tendinous tissues has traditionally been considered to be of limited magnitude. However, recent studies have suggested that glucose uptake increases in the force-transmitting tissues as a response to contractile loading, which in turn indicates an elevated tissue metabolism. The purpose of the present study was to investigate whether such a mechanism could be observed for the human Achilles tendon following tensile loading. Six subjects participated in the study. Unilateral Achilles tendon loading was applied by 25-min intermittent voluntary plantar flexor contractions. A radioactive tracer ( $[^{18}\text{F}]\text{-2-fluoro-2-deoxy-D-glucose}$ ) was administered during muscle action, and glucose uptake was measured by use of PET. Regions of interest were defined on the PET images corresponding to the cross section of Achilles tendon at two longitudinally separated sites (insertion and free tendon). Glucose uptake index was determined within respective regions of interest for the active and resting leg. Tendon force during voluntary contractions was  $\sim 13\%$  of maximal voluntary contraction force. Tendon loading induced an elevated glucose uptake index compared with that of the contralateral resting tendon in the region of tendon insertion ( $0.13 \pm 0.05$  vs.  $0.09 \pm 0.02$ ;  $P < 0.05$ ) and at the free tendon ( $0.12 \pm 0.01$  vs.  $0.08 \pm 0.02$ ;  $P < 0.05$ ). The present data suggest that tissue metabolism is elevated in the human Achilles tendon in response to low-intensity loading.

tendon metabolism; tendon mechanical function; connective tissue; imaging

TENDONS HAVE TRADITIONALLY been described as rigid and coherent structures that transmit contractile force in a linear manner to produce joint movement. However, it has become increasingly apparent that tendons undergo considerable deformation during loading and thereby contribute to locomotion by storing and releasing energy (10, 24, 35, 44). Moreover, the general perception has until recent years been that tendons exhibit a limited ability to react to mechanical loading by structural adaptation. Nonetheless, recent studies have demonstrated that tendinous tissues respond to both acute and habitual contractile loading, as indicated by either increased glucose uptake, oxygen consumption, and/or collagen synthesis of intra- or peritendinous tissues (8, 30–34). Furthermore, *in vivo* investigations of the mechanical properties of human tendon tissue indirectly suggest that tendons may adapt with respect to tissue “quality” in response to changes in loading patterns of

considerable magnitude, i.e., high-intensity exercise or disuse (26–28, 39–41). Collectively, these data suggest that the human tendon is far more dynamically responsive to loading than previously assumed.

Positron emission tomography (PET) enables three-dimensional depiction of glucose uptake and offers a minimally invasive method for investigating tissue metabolism. Several studies have successfully used PET to demonstrate exercise-induced increases in glucose uptake in heart and skeletal muscle (13, 14, 22, 37, 38); however, few studies have applied PET scanning to tendinous tissues (18, 21). Elevated glucose uptake was recently demonstrated in the quadriceps and patellar tendons after dynamic exercise (21), and concurrently it was suggested that exercise induced an increase in Achilles tendon glucose uptake (18). However, in the latter study, no resting values were obtained, and it therefore remains unknown whether the Achilles tendon responds metabolically to loading.

The Achilles tendon and its associated aponeuroses is a complex collagenous structure that undergoes substantial loading and deformation during human locomotion (12, 16, 35, 42). The tendon receives direct muscular insertion from three separately activated muscle compartments, and due to this complexity the mechanical function *in vivo* of the triceps surae muscle-tendon complex is poorly understood. Nonetheless, advances in this field would increase the understanding of Achilles tendon injury and exercise mechanisms. Interestingly, it was recently suggested that the Achilles tendon may undergo nonuniform loading due to heterogeneous activation of single muscles (2, 3, 7), although to what extent the three separate muscle compartments contribute to the loading of the common free tendon remains unanswered.

Therefore, the purpose of the present study was to apply PET to investigate 1) whether Achilles tendon glucose uptake increases in response to contractile loading and 2) whether selective activation of one of the muscles of the triceps surae (supposedly inducing nonuniform tendon loading) results in a different pattern of intratendinous glucose uptake compared with that during uniform tendon loading (voluntary contraction). We hypothesized 1) that glucose uptake increases in tendinous tissues after low-intensity loading and 2) that activating a single muscle of the triceps surae yields a heterogeneous pattern of glucose uptake intensity across the tendon to indicate that the Achilles tendon may undergo nonuniform loading during *in vivo* muscular contraction.

Address for reprint requests and other correspondence: J. Bojsen-Møller, Inst. of Sports Medicine Copenhagen, Bispebjerg bakke, 23, bygn 8, Bispebjerg Hospital, 2400 Copenhagen NV, Denmark (e-mail: jbm01@bbh.hosp.dk).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

## MATERIALS AND METHODS

**Subjects.** Six healthy men participated in the study (age,  $21 \pm 1$  yr; height,  $177 \pm 6$  cm; body weight,  $74.5 \pm 6.9$  kg). Subjects were recruited among recreationally active students. Before inclusion, all subjects filled out a questionnaire regarding medical history, and subjects were excluded if current lower extremity or lower back pain was reported or in case of present or recent leg injury. The ethical Committee of the Hospital District of the South-Western Finland approved the experimental procedures, and the study was performed in accordance with the Declaration of Helsinki. The purpose, procedures, and associated risks were explained to the subjects, and written, informed consent was obtained.

**Experimental protocol.** Each subject participated in two experimental sessions separated by at least 4 days. Between sessions, a disparate tendon-loading pattern was achieved by applying percutaneous stimulation of a single triceps surae muscle compartment (lateral gastrocnemius muscle) in *session 1* and voluntary contraction in *session 2*. Apart from the contraction mode, the protocol was similar in the two sessions. After insertion of catheters into the antecubital veins in both arms, the subjects were situated in a seated position with the hip joints slightly abducted and the knee joint of the experimental leg extended (Fig. 1). A small amount of saline was continuously administered to keep catheters clear. The total amount of saline infused during the 4 h of experiments was  $<500$  ml. The left ankle joint was in a neutral position ( $\sim 90^\circ$  between the sole of the foot and the tibia), and the forefoot was resting on a force transducer (I-KON, Chattanooga Group, Oxfordshire, UK) that was rigidly attached to the experimental bench. The subjects were supported with firm padding, and the backrest was adjusted such that no ankle joint movement was possible during plantar flexor contractions. The force sampling apparatus enabled audiovisual feedback to the subjects during voluntary contractions.

In *session 1*, two self-adhering surface electrical stimulation electrodes [StimTrode, model ST32D (32-mm RND), Axelgaard Manufacturing] were attached to the skin over the lateral gastrocnemius muscle (interelectrode distance of  $\sim 45$  mm). The electrodes were attached to an electrical stimulation apparatus (Elpha II 3000 Danmeter, Biofina). Each subject was familiarized to percutaneous stimulation by several submaximal electrical stimuli (range: 1–20 mA) over a period of 5–10 min. During all stimulation, subjects were in control of the stimulator and were instantly able to discontinue stimulation if intolerable pain or discomfort occurred. The current was progressively increased until a clear muscle fiber contraction was visualized. The subjects were encouraged to increase stimulation intensity to a bearable maximum considering the duration of the stimulation. The corresponding contractile force was registered. The following parameters were used during the electrical stimulation of the muscle: current amplitude of  $\sim 20$ –30 mA, 50 Hz, impulse time of 300 ms, ascending

time of 1.5 s, total stimulation time of 3 s, descending time of 1.5 s, rest time of 3 s. After 10 min of stimulation, 240 MBq of [ $^{18}$ F]-2-fluoro-2-deoxy-D-glucose in 2 ml of saline were infused, and stimulation was continued for an additional 25 min. All subjects were able to sustain 35 min of stimulation without decreasing stimulation intensity. The contractile force was registered every 5 min during the contraction protocol.

For the voluntary contraction protocol, a “target force” was determined based on the force obtained from the stimulation experiment in an attempt to have the lateral gastrocnemius muscle exert a force of similar magnitude in the two experimental situations. The voluntary target force was estimated based on the physiological cross-sectional areas of the lateral gastrocnemius (taken from the literature) relative to that of the entire triceps surae (15). The magnitude of force during voluntary contraction averaged  $\sim 13\%$  of plantar flexor maximal voluntary contraction (MVC) and was performed as 3-s contractions separated by 3 s of rest. To mimic the stimulated contraction protocol, voluntary contractions were controlled by audiovisual signals to the subject such that one signal was given to initiate contraction, one signal was given when a target force was reached, and one signal was given to terminate contraction. The low contraction intensity meant that all subjects were able to perform the voluntary task for the required time duration. Tendon force was estimated based on measurement of the distance from the lateral malleolus and the Achilles tendon and point of force application under the foot.

**PET image acquisition and processing.** Immediately after the muscle contractions were terminated, subjects were relocated to the PET scanner (Siemens ECAT HR+, Knoxville, TN) while care was taken to minimize any muscular contraction. The lower legs were divided into three segments starting from the heel and covering up to the fossa popliteus. Each segment was scanned in 4-min time frames, and this was repeated three times (21) (Fig. 2). After all emission scans, transmission scans for attenuation correction were performed for the same three areas. All data sets were corrected for attenuation, and they were reconstructed iteratively [6 iterations; 16 ordered subsets; Hann filter;  $128 \times 128$  matrix; zoom of either 2 (tendon region) or 1.8 (muscle region)]. Standard random, dead time, and scatter corrections of the manufacturer were employed. The axial and in-plane resolution of the reconstructed images was  $\sim 5$ -mm full width at half maximum. A venous blood sample for blood radioactivity determination was taken 50 min after the tracer injection and used for the calculation of glucose uptake index as described below.

Additionally, a MRI scan was performed of the lower legs of each subject (Fig. 3). This enabled a reference for determining the regions of interest (ROIs) that were defined during subsequent analysis.

Based on clinical observations of where tendon pain and injury often occur, two ROIs were defined for each tendon: 1) the “calcaneus region” just proximal to the insertion of the Achilles tendon on the

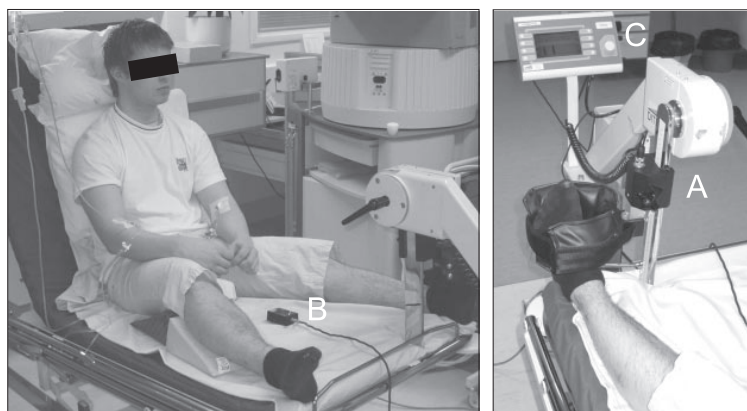
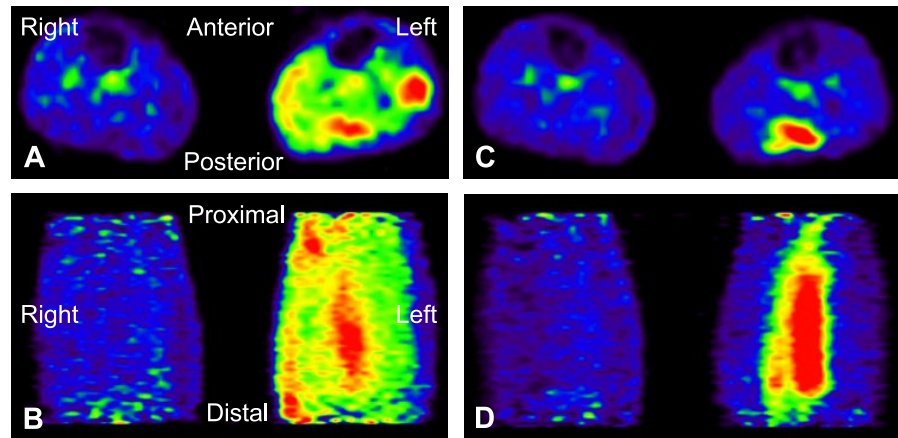


Fig. 1. Experimental setup. On both experimental days, subjects were in a seated position with the knee joint of the experimental leg extended and the ankle joint in neutral position ( $\sim 90^\circ$  between the sole of the foot and the tibia). The forefoot was resting on a force transducer (A). The subjects were supported with firm padding, and the backrest was adjusted such that no ankle joint movement was possible during plantar flexor contractions. The sampling apparatus enabled audiovisual (B and C) feedback to the subjects during voluntary contractions.

Fig. 2. Representative PET images from 1 subject obtained subsequent to unilateral contractile activity of the plantar flexor muscles. *A* and *C*: axial scans at  $\sim 50\%$  of the gastrocnemius muscle belly length. *B* and *D*: frontal plane scans of the proximal region of the lower legs. *A* and *B*: unilateral voluntary contraction of the plantar flexor muscles (right leg in image is the subject's left leg). *C* and *D*: unilateral selective stimulation of the m. gastrocnemius lateralis (*C* and *D* are oriented as *A* and *B*).



calcaneus and 2) the “free tendon” area  $\sim 3\text{--}4$  cm proximal to insertion where the tendon cross section exhibits a more rounded shape and where no muscle fibers insert on the tendon structure (Fig. 3). The ROIs were constructed by combining three adjacent scanning slices (each  $\sim 2.4$  mm thick), and therefore the ROIs should be regarded as 3D cross-sectional tendon volumes with a longitudinal thickness of  $\sim 7.2$  mm. During subsequent analysis, the glucose uptake index in the respective ROIs was calculated by dividing the tissue radioactivity with blood radioactivity (21, 45). Furthermore, the voxel values of glucose uptake index (a voxel is  $\sim 16$  mm<sup>3</sup>,  $2.6 \times 2.6$  mm in plane multiplied with the slice thickness of  $\sim 2.4$  mm) were extracted from the PET images, and the relative dispersion (equal to heterogeneity) of these values was calculated as relative dispersion =  $SD \times 100\% / \text{mean}$  for each ROI in each subject. In addition, the skewness and kurtosis of the distribution of voxel values were calculated.

**Statistical analysis.** Wilcoxon matched-pairs tests were used for comparing the glucose uptake index between resting and loaded

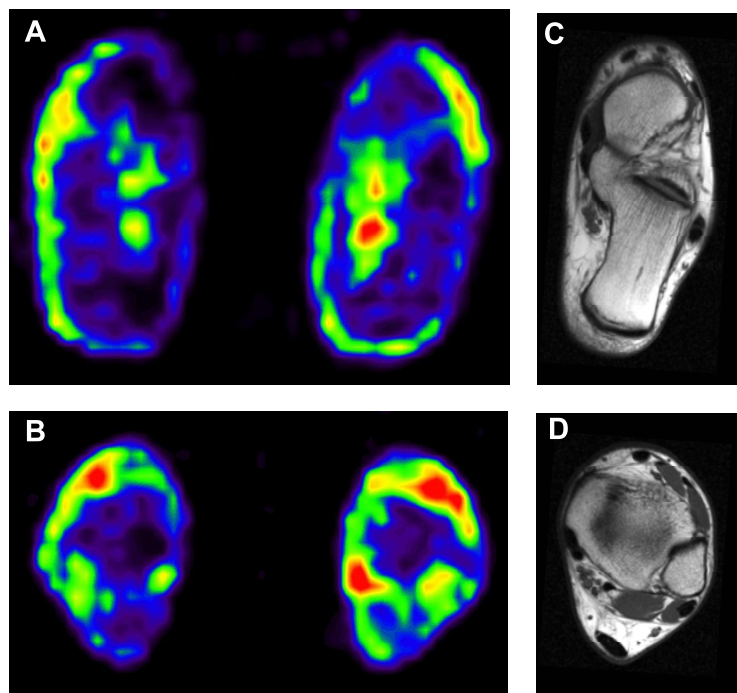
tendons and the glucose uptake heterogeneity of loaded tendons between conditions (voluntary vs. stimulated contraction). An  $\alpha$  level of  $<0.05$  was considered significant.

## RESULTS

The estimated tendon force during voluntary plantar flexor contractions was  $655 \pm 89$  N, which corresponded to  $\sim 13\%$  of MVC force. In the stimulation experiment, the stimulation intensity of  $24 \pm 4$  mA yielded an estimated tendon force of  $75 \pm 14$  N or  $\sim 2\%$  of MVC.

At the level of the calcaneal insertion, the voluntary activation resulted in an elevated glucose uptake index compared with that of the contralateral resting side ( $0.13 \pm 0.05$  vs.  $0.09 \pm 0.02$ ;  $P < 0.05$ ; Fig. 4). Similarly, at the level of the free tendon, the voluntary activation resulted in an elevated

Fig. 3. Representative PET images from 1 subject after unilateral voluntary contraction at the level of the calcaneus (*A*) and at the more proximal tendon level (*B*). Images are oriented as described in Fig. 2. *C* and *D*: corresponding magnetic resonance (MR) images in which the Achilles tendon can be identified by its dark color [MR image (*C*) is for technical reasons slightly more proximal than the PET image]. Glucose uptake is elevated in and/or around the tendon that has been loaded compared with the tendon of the resting leg. Note the wide crescent-shaped insertion area that corresponds to the most distal Achilles tendon inserting at the calcaneus.



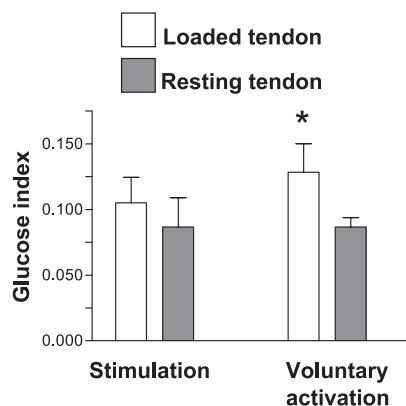


Fig. 4. Glucose uptake at the tendon insertion on the calcaneus. Glucose uptake is elevated in the tendon that has been loaded by voluntary contraction compared with the resting tendon. For the stimulation experiment, no difference is observed between loaded and unloaded tendon. \*Significantly different from resting tendon ( $P < 0.05$ ).

glucose uptake index compared with the contralateral resting tendon ( $0.12 \pm 0.01$  vs.  $0.08 \pm 0.02$ ;  $P < 0.05$ ; Fig. 5).

No increase in glucose uptake index was observed subsequent to electrical stimulation of the lateral gastrocnemius compared with the resting side. Glucose uptake heterogeneity was not different in any of the two anatomical tendon regions between electrically stimulated and voluntary contraction conditions. Skewness and kurtosis of voxel value distributions did not differ between the conditions.

## DISCUSSION

The main finding of the present study was that the Achilles tendon tissues exhibit elevated glucose uptake as a response to tendon loading. The tissue loading was generated by 25 min of voluntary intermittent isometric muscle contraction at low contraction intensity ( $\sim 13\%$  MVC). An increase in intra- and peritendinous glucose uptake suggests the occurrence of intratendinous metabolic activity as a response to contractile loading.

**Intratendinous metabolism.** Previous studies have demonstrated that cardiac and skeletal muscle tissue display large increases in tissue metabolism in response to exercise (14, 37, 38), which plausibly corresponds to the high density and nature of the cells in these tissues. Conversely, tendons have traditionally been regarded as metabolically inactive due to the limited occurrence of cells within tendinous tissues. Nonetheless, previous studies have demonstrated the presence of oxidative and glycolytic enzymes in tendons (20, 29). These enzymes are associated with the fibroblasts, which are the principal cells within tendinous tissues. The present experimental setup does not allow for a mechanistic explanation for the observed increased glucose uptake after loading. Nonetheless, previous *in vitro* studies have demonstrated that loading produces deformation of tendon, and hereby also of the embedded fibroblast cells and their nuclei (4, 43), and that fibroblasts respond to strain via mechanotransduction pathways to remodel the extracellular matrix (1, 5, 6, 46). It seems plausible that the observed glucose uptake in the present study may be associated to this plethora of intracellular processes that is elicited by fibroblast deformation.

Recent *in vivo* work in human models has demonstrated an increased oxygen uptake and blood flow in and/or around force-transmitting tissues as a response to mechanical loading (8, 9), and enhanced metabolic activity and collagen synthesis has been demonstrated in the peritendinous tissue of the Achilles tendon subsequent to acute loading (30–34). Attempts to examine human *in vivo* tendinous metabolism in response to loading have included near-infrared spectroscopy (9), microdialysis (33), and tendon biopsy (36), of which the latter two include considerably invasive procedures. PET scanning offers a minimally invasive technique to study physiological processes in the human body, and two recent studies have applied PET to investigate glucose uptake in tendinous tissues subjected to loading. Kalliokoski et al. (21) demonstrated an elevated tissue metabolism in the patellar tendon following fatiguing dynamic knee extensor contractions. Concomitantly, Hannukainen et al. (18) measured glucose uptake in the Achilles tendon tissues following bicycle exercise, and although resting levels of glucose uptake were not acquired the data indicated an elevation in glucose uptake as a response to tendon load. In the present study, glucose uptake was compared with resting levels of the contralateral tendon, and thus the present data extend the findings of Hannukainen et al. with respect to load-induced increase in Achilles tendon metabolism. Moreover, tendon load was not measured in the two previous PET studies since dynamic pedaling or kicking muscle actions were applied, and, hence, the present data add to previous results by showing that tendinous tissues respond with an increase in glucose uptake following isometric tendon loading at a low force level of  $\sim 13\%$  of MVC.

The voluntary contraction intensity of the present study was chosen to match that of the stimulated contractions (relative to muscle volume; see above), resulting in an estimated tendon load during voluntary contractions of  $\sim 650$  N or  $\sim 0.9$  times body weight. For technical reasons, few studies have directly measured Achilles tendon force during daily movement tasks: using an optic-fiber technique, Finni et al. (12) observed peak tendon forces of  $\sim 1,400$  N during walking, although large variation was observed between subjects. Computer models have been used to estimate that peak Achilles tendon forces reaches three to four times body weight during walking (16),

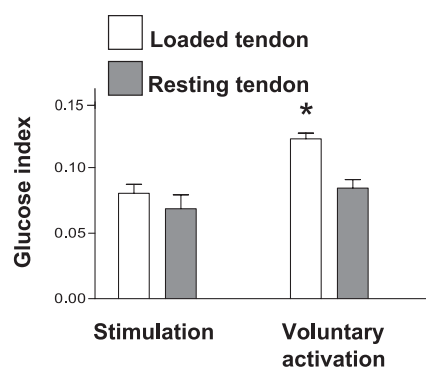


Fig. 5. Intratendinous glucose uptake at free-tendon level ( $\sim 3$ – $4$  cm proximal to the insertion). Glucose uptake is elevated in the tendon that has been loaded by voluntary contraction compared with the resting tendon. For the stimulation experiment, no difference is observed between loaded and unloaded tendon. \*Significantly different from resting tendon ( $P < 0.05$ ).

whereas Komi (23) recorded far greater peak Achilles tendon forces using a buckle transducer implant during running and jumping (3–10 kN). One study used the tendon buckle transducer to record tendon forces that were comparable to those of the present study during cycling ( $n = 1$ ; Ref. 17). It seems that the mechanical loading of the tendon in the present study is similar to or just below that associated with walking or cycling. The present data, therefore, suggest that even low-intensity loading, which may be associated with some rehabilitation protocols, elicits a certain metabolic response that may involve regenerative processes within the tendinous tissues.

**Nonuniform tendon deformation.** The Achilles and patellar tendons are force transmitters for the triceps surae muscle and the quadriceps muscle and, as such, are highly involved in human locomotion. The two tendons exhibit vastly distinct mechanical function: the patellar tendon has both a bony origin and insertion, whereas the Achilles tendon originates from contractile tissue and inserts onto bone. Although variation in tissue mechanical properties have been observed within the patellar tendon (19), which may suggest nonuniform loading, it seems plausible that the Achilles tendon to a greater extent may be subjected to nonuniform loading compared with the patellar tendon if the associated single muscle compartments are heterogeneously activated. Nonuniform activation would, in theory, result in uneven tendon deformation, and previous results have suggested that such a mechanism occurs in the Achilles tendon-aponeurosis complex (3, 7, 11). These recent findings are based on cadaver experiments or in vivo methods such as magnetic resonance scanning and ultrasonography. Importantly, these observations were performed at the aponeurosis level and are therefore solely indirect evidence of uneven tendon deformation. One in vivo study has directly observed nonuniform Achilles tendon deformation (35). In this study, a syringe needle (0.5-mm diameter) was (for other purposes) inserted transversely into the Achilles tendon, after which the subjects performed maximal voluntary plantar flexor contractions. In a subgroup of the subjects, the needle was permanently distorted on retraction. Because direct in vivo assessment of Achilles tendon deformation is presently not feasible, the secondary goal of the present study was to investigate whether regional difference in glucose uptake (i.e., heterogeneity) could be observed following different loading paradigms of the tendon. Tendon cross-sectional or longitudinal difference in glucose uptake between voluntary contraction and selective stimulation of a single compartment would indicate nonuniform stress distribution within the tendon. Due to limited scanning resources and safety issues regarding the infusion of the radioactive tracer, a model was chosen where voluntary contraction was compared with selective stimulation of one single muscle compartment of the triceps surae. The lateral gastrocnemius was chosen based on pilot experiments that demonstrated greater glucose uptake index within the tendon compared with selective stimulation of other triceps surae compartments. Nonetheless, when six subjects were included, no difference was observed between the resting tendon and the loaded tendon subsequent to lateral gastrocnemius stimulation, and therefore the previous notion of nonuniform tendon loading and/or associated heterogeneous tendon deformation is not supported by the present data. However, it should be noted that the stimulation of the lateral gastrocnemius yielded a limited stress on the associated force-transmitting tendon structures,

and although the loading was of a certain time duration, the present stimulation protocol may not have been a sufficient stimulus to be detected with the current scanner resolution. The axial and in-plane scanner resolution in the present study was ~4–5 mm, which becomes a limiting factor when the target of study (the Achilles tendon) has a diameter of ~8–12 mm and a cross-sectional area of ~100–120 mm<sup>2</sup> (25, 35). Future technical developments will likely increase scanner resolution, which potentially will allow explication of the mechanisms of tendon function in more detail than presently feasible. Furthermore, additional experiments applying greater stimulation intensity and/or additional stimulated muscle mass are required to address the question.

In conclusion, the present data demonstrate that glucose uptake in the Achilles tendon tissues increases during mechanical loading at low contraction intensity. This observation was made at the site of tendon insertion as well as in more proximal areas. An increased glucose uptake within the tendon indicates that tendinous tissues are metabolically active and likely exhibit greater ability to structural adaptation and remodeling in response to loading stimuli than previously appreciated. No conclusions regarding nonuniform tendon deformation were feasible with the present experimental approach; however, the PET scanning technique combined with other in vivo methods such as MRI and ultrasonography seem promising tools to investigate the distinct mechanical and structural function of human muscle-tendon complexes and their response to loading stimuli.

#### GRANTS

The present work was supported by the Academy of Finland (grants 206970 and 204240), by the Danish Research Agency, and by the Council for Sports Research appointed by the Danish Minister of Culture.

#### REFERENCES

1. **Almekinders LC, Banes AJ, and Ballenger CA.** Effects of repetitive motion on human fibroblasts. *Med Sci Sports Exerc* 25: 603–607, 1993.
2. **Arndt A, Bruggemann GP, Koebeke J, and Segesser B.** Asymmetrical loading of the human triceps surae: I. Mediolateral force differences in the Achilles tendon. *Foot Ankle Int* 20: 444–449, 1999.
3. **Arndt AN, Komi PV, Bruggemann GP, and Lukkariniemi J.** Individual muscle contributions to the in vivo achilles tendon force. *Clin Biomech (Bristol, Avon)* 13: 532–541, 1998.
4. **Arnoczky SP, Lavagnino M, Whallon JH, and Hoonjan A.** In situ cell nucleus deformation in tendons under tensile load; a morphological analysis using confocal laser microscopy. *J Orthop Res* 20: 29–35, 2002.
5. **Arnoczky SP, Tian T, Lavagnino M, Gardner K, Schuler P, and Morse P.** Activation of stress-activated protein kinases (SAPK) in tendon cells following cyclic strain: the effects of strain frequency, strain magnitude, and cytosolic calcium. *J Orthop Res* 20: 947–952, 2002.
6. **Banes AJ, Tsuzaki M, Hu P, Brigman B, Brown T, Almekinders L, Lawrence WT, and Fischer T.** PDGF-BB, IGF-I and mechanical load stimulate DNA synthesis in avian tendon fibroblasts in vitro. *J Biomech* 28: 1505–1513, 1995.
7. **Bojsen-Moller J, Hansen P, Aagaard P, Svantesson U, Kjaer M, and Magnusson SP.** Differential displacement of the human soleus and medial gastrocnemius aponeuroses during isometric plantar flexor contractions in vivo. *J Appl Physiol* 97: 1908–1914, 2004.
8. **Boushel R, Langberg H, Green S, Skovgaard D, Bulow J, and Kjaer M.** Blood flow and oxygenation in peritendinous tissue and calf muscle during dynamic exercise in humans. *J Physiol* 524: 305–313, 2000.
9. **Boushel R, Langberg H, Olesen J, Nowak M, Simonsen L, Bulow J, and Kjaer M.** Regional blood flow during exercise in humans measured by near-infrared spectroscopy and indocyanine green. *J Appl Physiol* 89: 1868–1878, 2000.
10. **Cavagna GA.** Storage and utilization of elastic energy in skeletal muscle. *Exerc Sport Sci Rev* 5: 89–129, 1977.

11. Finni T, Hodgson JA, Lai AM, Edgerton VR, and Sinha S. Nonuniform strain of human soleus aponeurosis-tendon complex during submaximal voluntary contractions in vivo. *J Appl Physiol* 95: 829–837, 2003.
12. Finni T, Komi PV, and Lukkariniemi J. Achilles tendon loading during walking: application of a novel optic fiber technique. *Eur J Appl Physiol Occup Physiol* 77: 289–291, 1998.
13. Fujimoto T, Itoh M, Tashiro M, Yamaguchi K, Kubota K, and Ohmori H. Glucose uptake by individual skeletal muscles during running using whole-body positron emission tomography. *Eur J Appl Physiol* 83: 297–302, 2000.
14. Fujimoto T, Kempainen J, Kalliokoski KK, Nuutila P, Ito M, and Knuuti J. Skeletal muscle glucose uptake response to exercise in trained and untrained men. *Med Sci Sports Exerc* 35: 777–783, 2003.
15. Fukunaga T, Roy RR, Shellock FG, Hodgson JA, Day MK, Lee PL, Kwong-Fu H, and Edgerton VR. Physiological cross-sectional area of human leg muscles based on magnetic resonance imaging. *J Orthop Res* 10: 928–934, 1992.
16. Giddings VL, Beaupre GS, Whalen RT, and Carter DR. Calcaneal loading during walking and running. *Med Sci Sports Exerc* 32: 627–634, 2000.
17. Gregor RJ, Komi PV, and Jarvinen M. Achilles tendon forces during cycling. *Int J Sports Med* 8, Suppl 1: 9–14, 1987.
18. Hannukainen J, Kalliokoski KK, Nuutila P, Fujimoto T, Kempainen J, Viljanen T, Laaksonen MS, Parkkola R, Knuuti J, and Kjaer M. In vivo measurements of glucose uptake in human Achilles tendon during different exercise intensities. *Int J Sports Med* 26: 727–731, 2005.
19. Haraldsson BT, Aagaard P, Krogsgaard M, Alkjaer T, Kjaer M, and Magnusson SP. Region-specific mechanical properties of the human patella tendon. *J Appl Physiol* 98: 1006–1012, 2005.
20. Jozsa L, Balint JB, Reffy A, and Demel Z. Histochemical and ultrastructural study of adult human tendon. *Acta Histochem* 65: 250–257, 1979.
21. Kalliokoski KK, Langberg H, Ryberg AK, Scheede-Bergdahl C, Doessing S, Kjaer A, Boushel R, and Kjaer M. The effect of dynamic knee-extension exercise on patellar tendon and quadriceps femoris muscle glucose uptake in humans studied by positron emission tomography. *J Appl Physiol* 99: 1189–1192, 2005.
22. Kempainen J, Fujimoto T, Kalliokoski KK, Viljanen T, Nuutila P, and Knuuti J. Myocardial and skeletal muscle glucose uptake during exercise in humans. *J Physiol* 542: 403–412, 2002.
23. Komi PV. Relevance of in vivo force measurements to human biomechanics. *J Biomech* 23, Suppl 1: 23–34, 1990.
24. Komi PV and Bosco C. Utilization of stored elastic energy in leg extensor muscles by men and women. *Med Sci Sports* 10: 261–265, 1978.
25. Kongsgaard M, Aagaard P, Kjaer M, and Magnusson SP. Structural Achilles tendon properties in athletes subjected to different exercise modes and in Achilles tendon rupture patients. *J Appl Physiol* 99: 1965–1971, 2005.
26. Kubo K, Akima H, Kouzaki M, Ito M, Kawakami Y, Kanehisa H, and Fukunaga T. Changes in the elastic properties of tendon structures following 20 days bed-rest in humans. *Eur J Appl Physiol* 83: 463–468, 2000.
27. Kubo K, Kanehisa H, and Fukunaga T. Effects of different duration isometric contractions on tendon elasticity in human quadriceps muscles. *J Physiol* 536: 649–655, 2001.
28. Kubo K, Kanehisa H, and Fukunaga T. Effects of resistance and stretching training programmes on the viscoelastic properties of human tendon structures in vivo. *J Physiol* 538: 219–226, 2002.
29. Kvist M, Jozsa L, Jarvinen MJ, and Kvist H. Chronic Achilles paratenonitis in athletes: a histological and histochemical study. *Pathology* 19: 1–11, 1987.
30. Langberg H, Bulow J, and Kjaer M. Blood flow in the peritendinous space of the human Achilles tendon during exercise. *Acta Physiol Scand* 163: 149–153, 1998.
31. Langberg H, Bulow J, and Kjaer M. Standardized intermittent static exercise increases peritendinous blood flow in human leg. *Clin Physiol* 19: 89–93, 1999.
32. Langberg H, Rosendal L, and Kjaer M. Training-induced changes in peritendinous type I collagen turnover determined by microdialysis in humans. *J Physiol* 534: 297–302, 2001.
33. Langberg H, Skovgaard D, Karamouzis M, Bulow J, and Kjaer M. Metabolism and inflammatory mediators in the peritendinous space measured by microdialysis during intermittent isometric exercise in humans. *J Physiol* 515: 919–927, 1999.
34. Langberg H, Skovgaard D, Petersen LJ, Bulow J, and Kjaer M. Type I collagen synthesis and degradation in peritendinous tissue after exercise determined by microdialysis in humans. *J Physiol* 521: 299–306, 1999.
35. Magnusson SP, Hansen P, Aagaard P, Brond J, Dyhre-Poulsen P, Bojsen-Møller J, and Kjaer M. Differential strain patterns of the human gastrocnemius aponeurosis and free tendon in vivo. *Acta Physiol Scand* 177: 185–195, 2003.
36. Miller BF, Olesen JL, Hansen M, Dossing S, Cramer RM, Welling RJ, Langberg H, Flyvbjerg A, Kjaer M, Babraj JA, Smith K, and Rennie MJ. Coordinated collagen and muscle protein synthesis in human patella tendon and quadriceps muscle after exercise. *J Physiol* 567: 1021–1033, 2005.
37. Nuutila P, Peltoniemi P, Oikonen V, Larmola K, Kempainen J, Takala T, Sipila H, Oksanen A, Ruotsalainen U, Bolli GB, and Yki-Jarvinen H. Enhanced stimulation of glucose uptake by insulin increases exercise-stimulated glucose uptake in skeletal muscle in humans: studies using [<sup>15</sup>O]O<sub>2</sub>, [<sup>15</sup>O]H<sub>2</sub>O, [<sup>18</sup>F]fluoro-deoxy-glucose, and positron emission tomography. *Diabetes* 49: 1084–1091, 2000.
38. Peltoniemi P, Yki-Jarvinen H, Laine H, Oikonen V, Ronnema A, Kalliokoski K, Raitakari O, Knuuti MJ, and Nuutila P. Evidence for spatial heterogeneity in insulin- and exercise-induced increases in glucose uptake: studies in normal subjects and patients with Type 1 diabetes. *J Clin Endocrinol Metab* 86: 5525–5533, 2001.
39. Reeves ND, Maganaris CN, Ferretti G, and Narici MV. Influence of 90-day simulated microgravity on human tendon mechanical properties and the effect of resistive countermeasures. *J Appl Physiol* 98: 2278–2286, 2005.
40. Reeves ND, Maganaris CN, and Narici MV. Effect of strength training on human patella tendon mechanical properties of older individuals. *J Physiol* 548: 971–981, 2003.
41. Reeves ND, Narici MV, and Maganaris CN. Strength training alters the viscoelastic properties of tendons in elderly humans. *Muscle Nerve* 28: 74–81, 2003.
42. Scott SH and Winter DA. Internal forces of chronic running injury sites. *Med Sci Sports Exerc* 22: 357–369, 1990.
43. Screen HR, Lee DA, Bader DL, and Shelton JC. Development of a technique to determine strains in tendons using the cell nuclei. *Biorheology* 40: 361–368, 2003.
44. Voigt M, Simonsen EB, Dyhre-Poulsen P, and Klausen K. Mechanical and muscular factors influencing the performance in maximal vertical jumping after different prestretch loads. *J Biomech* 28: 293–307, 1995.
45. Yokoyama I, Inoue Y, Moritan T, Ohtomo K, and Nagai R. Simple quantification of skeletal muscle glucose utilization by static <sup>18</sup>F-FDG PET. *J Nucl Med* 44: 1592–1598, 2003.
46. Zeichen J, van Griensven M, and Bosch U. The proliferative response of isolated human tendon fibroblasts to cyclic biaxial mechanical strain. *Am J Sports Med* 28: 888–892, 2000.