Mechanical properties of tendon and aponeurosis of human gastrocnemius muscle in vivo

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Muramatsu, Tadashi, Tetsuro Muraoka, Daisuke Takeshita, Yasuo Kawakami, Yuichi Hirano, and Tetsuo Fukunaga. Mechanical properties of tendon and aponeurosis of human gastrocnemius muscle in vivo. J Appl Physiol 90: 1671–1678, 2001.—Load-strain characteristics of tendinous tissues (Achilles tendon and aponeurosis) were determined in vivo for human medial gastrocnemius (MG) muscle. Seven male subjects exerted isometric plantar flexion torque while the elongation of tendinous tissues of MG was determined from the tendinous movements by using ultrasonography. The maximal strain of the Achilles tendon and aponeurosis, estimated separately from the elongation data, was $5.1 \pm 1.1$ and $5.9 \pm 1.6\%$, respectively. There was no significant difference in strain between the Achilles tendon and aponeurosis. In addition, no significant difference in strain was observed between the proximal and distal regions of the aponeurosis. The results indicate that tendinous tissues of the MG are homogenously stretched along their lengths by muscle contraction, which has functional implications for the operation of the human MG muscle-tendon unit in vivo.

elements; ultrasonography

SKELETAL MUSCLE IS COMPOSED of contractile (muscle fibers) and elastic components. The elastic component is passively stretched by external force (3, 39) and interacts with the contractile component (14, 21). The elastic component also functions as elastic energy storage (1, 3) and mechanical buffer (13). Anatomically, the elastic component is composed of tendinous tissues (tendon and aponeurosis); epi-, peri-, and endomysium; sarcolemma; and endosarcomeric structures (41). Among these, the tendinous tissues have been shown to act as a primary elastic component (41).

During an isometric contraction, the tendinous tissue is stretched while muscle fibers shorten (13, 17, 36). Several studies have reported that elastic strain of the tendon and aponeurosis differs: the aponeurosis has often been found to bear more strain than the tendon (7, 10, 28). Even the homogeneity of strain within the aponeurosis has been questioned (39, 41). Another study, however, reported that the mechanical properties of the tendon and aponeurosis were similar (36). Lack of consensus in the difference between the tendon and aponeurosis has made it difficult to understand the relative contributions of the elements of tendinous tissues as an elastic component. Knowledge of the strain distribution along the tendon and aponeurosis is necessary to understand structure and function of the tendinous tissues and to make more accurate models of the muscle-tendon unit.

Most studies on the mechanical properties of the elastic component have been based on animal experiments (10, 23, 28, 33, 39, 40) or human cadavers (4, 6). The results of these studies might not be directly applicable to humans in vivo because of the differences in species (23) and/or the effects of sterilization or preservation (38).

The interaction between contractile and elastic components is important in the performance of human physical activities. This is especially important in muscles with long tendons such as the gastrocnemius muscle in humans, because, for the same strain, the absolute elongation is greater when the tendon is longer. Although recent studies reported mechanical properties of the human tendinous tissues in vivo by using ultrasonography (18, 19, 25, 26, 29, 30), few of those estimated strain of the tendon and aponeurosis separately (30). In addition, no study to date has estimated strain distribution along the human aponeurosis in vivo.

The purposes of this study were 1) to measure and compare the strain of the human tendon and aponeurosis in vivo and 2) to examine the strain distribution along the aponeurosis. We studied the tendon (Achilles tendon) and aponeurosis of the human medial gastrocnemius (MG) muscle. We regarded the Achilles tendon to extend from muscle-tendon junction of MG to the insertion onto the calcaneus bone (12, 24). Our hypotheses were 1) that Achilles tendon and MG aponeurosis

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are both extensible, and hence strain by muscle force production, 2) that MG aponeurosis strains more than Achilles tendon, and 3) that heterogeneity of the strain along the aponeurosis exists.

METHODS

Subjects

The subjects of this study were seven healthy men [age 26.1 ± 3.3 yr, height 171.3 ± 8.2 cm, weight 71.9 ± 8.6 (SD) kg]. The purpose and procedures were explained to the subjects before their consent to participate in the study was obtained.

Joint Position Settings and Torque Measurements

An electric myometer (model Myoret RZ-450, Asics, Tokyo, Japan) was used to fix the ankle joint and to measure plantar flexion torque. Each subject lay prone on a bed, with the left foot firmly secured to the footplate of the myometer with two straps. The center of rotation of the myometer was visually aligned with the center of rotation of the ankle joint. The foot was fixed at neutral anatomic position, where the sole of the foot was at 90° to the tibia (Fig. 1). Torque signals were analog-to-digital converted at a sampling rate of 1 kHz (MacLab, AD Instrument) for further analysis.

After a warm-up session with three submaximal contraction and maximum voluntary contraction (MVC) measurements, the subjects were instructed to exert isometric torque from relaxation to 90% MVC with a visual aid of the developed torque on an oscilloscope with a ramp increase in torque at 10% MVC/s. At this rate of force increment, we assumed that viscous properties of tendinous tissues are not substantial (5, 15, 22, 28). The measurement was repeated three times for each site (P1–P3; see Measurement of the Tendinous Tissue Displacement) with at least 3 min of rest between trials. Average values over three measurements were adopted. In our pilot experiments on seven subjects (the 6 subjects of the present study plus 1 additional male subject), electromyograms of the MG, lateral gastrocnemius, soleus, and tibialis anterior muscles were taken during isometric contractions from rest to 90% MVC. The mean electromyographic activity of each muscle was linearly related to the exerted torque ($r > 0.9$). Therefore, we assumed that there was a consistent relative contribution of MG to plantar flexion torque at different torque levels. The electromyographic activity of tibialis anterior during contractions of plantar flexors was negligible, and we conclude that the effect of cocontraction was minimal.

In addition to the above main experiment, additional measurements under the following two conditions were conducted for all subjects.

Passive condition. The subjects were asked to completely relax their leg muscles. Then, the ankle was passively moved at 5°/s with the range of motion between 75 and 135° of the included angle of the ankle. After five cycles as preconditioning (9, 27), we collected data during the sixth and seventh cycles in the plantar flexion phase. This condition was executed to determine the effect of the ankle joint movements on the tendinous tissue movements.

Plantar flexed condition. During the main experiments with the ankle angle set at 90°, small but significant passive torque was detected. Therefore, we could not exclude the possibility that the tendinous tissue had already been elongated at a resting state. As a result, the estimated strain might have been underestimated. To address this concern, we executed an additional experiment in which the ankle angle was set at 105°, where the passive torque was virtually zero (34). The detected torque at 105° was 0.085 Nm (0.09% of the maximal torque at 105°) on average. Subjects were instructed to perform ramp isometric torque development from complete relaxation to ~10 Nm. Three trials were carried out. We determined the strain that had been generated in the resting state at 90° by passive lengthening of the muscle.

Measurement of the Tendinous Tissue Displacement

The muscle tested in this study was the medial gastrocnemius muscle (MG). The ultrasonic apparatus (model SSD-2000, Aloka, Tokyo, Japan) was used with an electronic linear array probe of 7.5-MHz wave frequency. The scanning
head of ultrasonic apparatus was coated with water-soluble transmission gel, which provided acoustic contact without depressing the dermal surface. The width and depth resolution of ultrasonography with this probe were 0.67 and 0.4 mm, respectively. The precision and linearity of the image using ultrasonography have been confirmed by Kawakami et al. (21), who compared the distance between pins struck on an acoustic standoff and the distance between the pins in the reconstructed image.

The probe was longitudinally attached to the dermal surface by an adhesive tape, which restrained the probe from sliding (11, 26, 30), over the mediolateral center of MG. To evaluate the elongation of the Achilles tendon and the aponeurosis separately, the movements of the following three points were recorded by ultrasonography: the myotendinous junction of MG (P1) and the central (P2) and proximal (P3) intersection made by the fascicles and aponeurosis of MG (Fig. 2). We carefully chose these points, confirming that they showed clear echoes throughout the muscle contraction and that P2 was almost midpoint between P1 and P3. We recorded P2 on the aponeurosis to clarify the position-dependent length change along the aponeurosis. The portion P0–P1 is the Achilles tendon. Its proximal part is a composite of the MG tendon and soleus aponeurosis (12). Therefore, MG tendon and soleus aponeurosis act in fusion, and the force produced by the soleus muscle might have affected the behavior of Achilles tendon. This concern will be discussed later (see DISCUSSION). The order of experiments among P1–P3 was randomized. Thus the trials were carried out under the three conditions (main condition plus 2 additional conditions) and the three places (P1–P3) of the probe. The trial was repeated when the subjects could not exert designated torque.

In all conditions, each point moved slowly enough for the investigator to scan each point successfully (Fig. 3). The ultrasound images were transferred to a personal computer (model Powerbook G3, Apple, Tokyo, Japan) at 30 Hz for measurements of the displacement of the three points by using the public domain National Institute of Health (NIH) Image program (written by Wayne Rasband at the NIH and available from http://rsb.info.nih.gov).

Measurement of the Ankle Joint Movements

Changes of the joint angle affect tendinous tissue movements (11, 21, 31). Even in the “isometric” tests using myometers, the joint angle might change as a result of the mechanical compliance of the myometers and/or the subjects' limb deviation from the myometers, especially in the joint that produces large torque as during plantar flexion. In that case, the displacement of the tendinous tissue was attributed not only to the mechanical force exerted on the tissue but also to the joint movements and hence length change of the whole muscle-tendon complex. We would then overestimate the tendinous tissue movements as a result of not compensating for the effect of the ankle joint movements.

To evaluate the ankle joint movements, we videotaped the ankle in a left sagittal view at 200 frames/s (MEMRECAM C2S, Nac, Tokyo, Japan). Retroreflective markers were attached to the subjects at the lateral epicondyle of femur (EF), lateral malleolus (LM), calcaneal tuber (CT), lateral side of the head of the fifth metatarsal, and upper and lower part of the footplate of the myometer. The angle made by the EF, LM, and CT was measured as an ankle angle.

Video data from these retroreflective markers was processed by using standard planar calibration techniques to determine the sagittal plane kinematic motion. The myometer, ultrasonography, and the high-speed video camera were synchronized with recordings of a clock timer for subsequent analyses. The recorded (30 Hz) ultrasound images and videotaped (200 Hz) joint kinematics, closest to the time of torque production at the designated level (0–90% MVC), were used for analyses.

Correction of the Tendinous Tissue Displacements

To calculate the tendinous tissue strain accurately, we executed two types of corrections in obtaining the displacement of each point. For correction 1, from the first additional measurement (passive condition), we obtained the relationship between the ankle angle change and the displacement of each point. On the basis of this relationship, we subtracted the displacement due to the plantar flexion movements during isometric contraction. For correction 2, from the second...
additional measurement (plantar flexed condition), we correct for the displacement of each point (P1–P3) that had occurred in the main condition at 90° by the presence of passive torque. This was done by measuring the positional changes in each point during a ramp isometric contraction at 105°, up to the torque passively recorded at 90°. The target torque level was set to 3.2 ± 0.9 Nm, corresponding to 3.0 ± 0.8 Nm at 90°, after taking into consideration the changes in moment arm length by joint rotation (35). Then, we added the calculated displacement to each points’ displacement data at each torque (10%–90% MVC). In this way, we double corrected the displacement data to reduce errors included in the measurements.

Estimation of the Strain of the Tendinous Tissue

The Achilles tendon length at rest was defined as the distance between P1 and the osteotendinous junction (P0) detected by ultrasonography. The position of the probe was marked on the dermal surface to determine the distance between P0, P1, P2, and P3. We estimated the length of each segment at rest by measuring the distance between respective points, taking into consideration the positions of the probe. The elongation of each segment was calculated by subtracting the distal point’s displacement from the proximal one. The strain of each segment was obtained by dividing the elongation by the length of the segment at rest.

Reproducibility

We evaluated the reproducibility of the displacement data of P1, P2, and P3 through three procedures: 1) interday reproducibility, which was tested for three subjects on 2 separate days; 2) reproducibility of three trials for all subjects in the main experiments; and 3) reproducibility of the digitizing the ultrasonic image; images for all trials were digitized for three times. The reproducibility was evaluated on the basis of a coefficient of variation (CV) (SD/mean) (19, 31).

Statistics

Values are presented as means ± SD. A two-way ANOVA with repeated measures was used to analyze the effects of 1) torque and positions (P1–P3) on the displacement, 2) torque and segments (Achilles tendon and aponeurosis) on the strain, and 3) torque and segments in the aponeurosis (distal and proximal) on the strain. To evaluate reproducibility of measurements, paired t-test and a one-way ANOVA with repeated measures were used to analyze the interday reproducibility and the effect of positions (P1–P3) on the CV of the displacements of three trials, respectively. F ratios were considered significant at P < 0.05. Significant differences among means at P < 0.05 were detected by using Tukey-Kramer’s post hoc tests.

RESULTS

As for the interday reproducibility, the mean CV of the displacements of P1, P2, and P3 was, on the average, 8.5, 5.3, and 5.7%. Paired t-test showed no significant difference of the displacement at nine torque levels between 2 days for each of three subjects. The mean CV of the displacements of P1, P2, and P3 among three trials of all subjects in the main experiments was 8.8, 5.3, and 5.7%, respectively. Significant differences of the CV between P1 and the other two points were found. The mean CV of three digitizing for the same image was 1.6%.

Displacements of the three points (P1–P3) at nine torque levels at 90° are shown in Fig. 4. Differences of points and torque levels both affected the displacements significantly. Post hoc tests showed the significant differences between P1 and the other two points.

Figure 5 shows the strain of the Achilles tendon and the aponeurosis (P1–P3) at nine torque levels at 90°. In the Achilles tendon and the aponeurosis, strain did not significantly change above 30 and 60% MVC, respectively. In addition, significant difference in strain was not found between the Achilles tendon and aponeurosis. There was no significant difference in strain between the distal (P1–P2) and the proximal (P2–P3) aponeurosis (Fig. 6).

A typical recording of the tendinous displacement, torque, and ankle angle change for one subject is shown in Fig. 7.

Figures 8 and 9 show the effect of the two corrections on the torque-strain relationship in Achilles tendon and aponeurosis, respectively. The effect of the correction 1 (i.e., passive condition) is apparent in Fig. 8. Without the correction 1, the Achilles tendon strain was substantially overestimated, especially when the exerted torque was large. In Fig. 9, the effect of correction 2 (i.e., plantar flexed condition) is clearly shown. Without correction 2, the aponeurosis strain was underestimated in all torque levels. Mean ankle angle change from rest to 90% MVC was 7.4 ± 1.8°.

DISCUSSION

This is, to the best of our knowledge, the first study that measured the strain of human Achilles tendon and aponeurosis separately and the strain distribution along the aponeurosis in vivo. Most of the previous
studies have focused on animals or human cadavers (4, 6, 10, 23, 28, 33, 39, 40). The data from human in vivo studies are important for understanding the tendinous behavior during actual human movements.

We made three hypotheses as follows: 1) Achilles tendon and MG aponeurosis both possess elasticity and hence strain by muscle force production, 2) MG aponeurosis strains more than Achilles tendon, and 3) heterogeneity of the strain along the MG aponeurosis exists. Among those, hypothesis 1 was supported, based on the fact that both the Achilles tendon and MG aponeurosis strained substantially. However, hypotheses 2 and 3 were not supported because distinct heterogeneity of the strain along the tendinous tissue was not found.

In the present study, maximal torque level during testing was 90% of the initially measured MVC. Trials demanding large force exertions were executed at least nine times (3 trials for 3 positions), and it was impossible for subjects to repeatedly exert 100% MVC. Therefore, the maximum strain measured in this study might have been underestimated. However, considering that the strain did not significantly increase above 30 and 60% of MVC for the tendon and aponeurosis, respectively, the underestimation should not be considerable.

Each point (P1–P3) moved in the identical plane during force exertion because each point was continuously in focus (21). In addition, the probe was put on the midlongitudinal line (21). Therefore, it would be reasonable to suppose that underestimation of the displacement did not occur.

Reported values of strain of the tendon are 2% for frog semitendinosus and gastrocnemius muscles at P0 (28, 39), 2.5% for human tibialis anterior muscle at P0 (30), 4% for wallaby gastrocnemius muscle during hopping (2), and 5% for human Achilles tendon during running (2). Alexander and Ker (2) suggested that leg tendons of many mammals are stretched by ~5% during running. Similarly, Shadwick (37) reported that tendons were elastically stretched up to 5%. As for the aponeurosis, reported strain values at P0 are wide ranging: 2% for frog gastrocnemius (39), 3.5% for rat gastrocnemius (16), 7% for human tibialis anterior muscle (30), 8% for frog semitendinosus (27), and 14.3% for rat MG (41). This variation may have resulted from not only the difference of the species or muscles but also the difference of the definition of 0% strain, which will be discussed later. Our results (~5% for the Achilles tendon and ~6% for the aponeurosis) are in line with those studies, although very few data are presently available for the strain of both the tendon and aponeurosis of humans in vivo.

In the present study, there was no significant difference in the maximum strain between the Achilles tendon and aponeurosis. This supports previous studies (33, 36, 39), but some studies have shown the contrary [2 vs. 8% (28), 3~4 vs. 9~10% (10), 2.5 vs. 7% (30) (tendon strain vs. aponeurosis strain)]. Scott and Loeb (36) reported that the organization of the collagen in the aponeurosis was similar to that of the tendon. On the other hand, Delgado-Lezama et al. (7) reported that the stiffness of the tendon was larger than that of the aponeurosis, and they attributed the difference to the difference in the mass per unit length. Proximal part of the outer tendon (Achilles tendon) was composed of not only the MG tendon but also the soleus aponeurosis (12), both of which are tightly connected with each other. Therefore, this part is pulled by both the MG and soleus muscle. That is, the tension applied on this part should be clearly larger than that on the MG aponeurosis. The result that the strain was not different between the two regions is presumably because of the difference in the cross-sectional area of the Achilles tendon.
tendon and aponeurosis. The tendon of the MG gradually changes its shape, from columnar part around the calcaneus to a sheetlike structure before becoming the distal aponeurosis (12). In other words, stress in the Achilles tendon and aponeurosis might have been similar because the intrinsic material was the same (36). Further studies would clarify this point, and actual tendon and aponeurosis stress would be determined by combining the present findings with accurate muscle force estimation.

Heterogeneity of the strain along the aponeurosis was not seen in the present study. This was an unexpected result, because Zuurbier et al. (41) reported higher compliance of the distal compared with the proximal aponeurosis. However, van Bavel et al. (40) reported that aponeurosis thickness systematically decreased as it extended from the muscle-tendon junction to the muscle belly, which paralleled changes in

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Fig. 7. Actual recording of the displacement, torque, and ankle angle change for 1 subject. The probe was placed at P2 in this trial.

Fig. 8. Effects of corrections on Achilles tendon strain. ○, No correction; □, correction 1; △, correction 2; ●, corrections 1 and 2. Correction 1 was expected to correct for the effect of the ankle joint movements on the tendinous tissue movements. Correction 2 was executed to correct for the initial lengthening at rest of the tendinous tissues.

Fig. 9. Effects of corrections on aponeurosis strain. ○, No correction; □, correction 1; △, correction 2; ●, corrections 1 and 2.
the forces exerted on the aponeurosis. This resulted in constant strain over the whole aponeurosis. The present result is in accordance with this finding.

If there is heterogeneity of the strain along the tendinous tissue (tendon and aponeurosis), the region with larger strain could be damaged more easily, because it is closer to its failure limit of the stress-strain relationship. The result that there was not distinct heterogeneity of the strain along the tendinous tissue is, therefore, physiologically reasonable.

Both in the tendon and aponeurosis, the strain substantially changed from 0 to 10% MVC, which would correspond to the toe region (32). Dunn and Silver (8) stated that the crimp in collagen fibrils from rat tail tendon was straightened by strains of ~3%. In light of this finding, in the present study, the crimp of the collagen fibrils in the tendon and aponeurosis might have been straightened by ~20% MVC. In the present study, however, the strain did not increase significantly with the torque increment above 30% MVC for the Achilles tendon and above 60% MVC for the aponeurosis (Fig. 5). The lack of significant increase of the strain partly resides in the large individual variability in the load-strain relationship. Intersubject variability of the elastic properties of the tendinous tissues has recently been studied (25, 26). Another possibility might be that the lack of statistical significance was due to the methodological errors in this study, which might be associated with the CVs in the ultrasonic measurements. Specifically, the CVs of the displacement of P1 was significantly large, which means that scanning of the muscle-tendon junction is more difficult than that of intersection made by aponeurosis and fascicles. Current studies are underway to elucidate the effects of these factors.

Two types of corrections were used to reduce errors in estimation of the strain in the present study. The effect of correction 1 (passive condition) on the Achilles tendon strain and the effect of correction 2 (plantar flexed condition) on the aponeurosis strain were substantial (Figs. 8 and 9, respectively). In any human isometric tests, the joint angle has been assumed to be constant without direct monitoring the joint angle. This assumption might induce errors if the measured variables were angle dependent. The present study showed the necessity of monitoring the joint angle in strain tests based on the isometric contractions.

Other factors that could make the comparison of the strain value among the studies difficult are the lack of agreement on a definition of 0% strain (28, 41). In most of the material tests of the tendinous tissue, small passive preload was applied to the tendon before testing (28). In the human tests in vivo, the estimation of the exact force applied on the tendinous tissue is very difficult. We assumed that the tension of the tendinous tissue was negligible when the detected torque was zero. At the 105° angle in relaxed state, the detected torque was negligible; therefore, we defined that the strain of the tendinous tissue in this position was 0%.

The tendinous tissues of the gastrocnemius muscles have been shown to be the site of elastic energy storage that enhances the efficiency of locomotion (3, 39) and a mechanical buffer to protect the muscle fibers during eccentric contractions (13). Significant strain of both Achilles tendon and aponeurosis, as observed in this study, would reflect the contribution of the whole tendinous tissues as an elastic component that could favor movement performance.

In summary, during isometric contraction of the human medial gastrocnemius muscle in vivo, we found that 1) both Achilles tendon and aponeurosis strained substantially, 2) strain of Achilles tendon and aponeurosis was similar, and 3) there was no difference in strain between proximal and distal part of the aponeurosis. These findings are particularly important for understanding the mechanical functions of the human gastrocnemius muscle-tendon unit in vivo and for more accurate muscle modeling in future studies.

REFERENCES


