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Structural tendon changes in patients with acromegaly: assessment of Achilles tendon with sonoelastography.

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Abstract

Aims: To describe the sonoelastographic appearance of the Achilles tendon in acromegalic patients and to determine whether the blood concentrations of growth hormone (GH) and insulin-like growth factor (IGF-1) are associated with the various sonographic elasticity types of Achilles tendons. **Material and methods:** Eighty-four Achilles tendons of 42 acromegaly patients and 84 Achilles tendons of 42 healthy volunteers were assessed with sonoelastography. The tendons were classified into two main types according to the elasticity features: type 1 blue/green (hard tissue) and type 2 yellow/red within green (intermediate–soft tissue). Two subtypes of these types were also defined. According to the definition, the elasticity of the tissue was in a spectrum ranging from hard to soft as the type progressed from 1a to 2b. **Results:** The mean thickness of Achilles tendons in patients with acromegaly was significantly higher compared with healthy Achilles tendons (5.1±0.7 mm vs. 4.4±0.5, $p<0.001$), and patients with active disease had thicker Achilles tendons (5.5±0.8 mm vs. 4.8±0.5 mm in inactive disease, $p=0.003$). A significantly higher proportion of acromegaly patients had type 2 sonoelastographic appearance of the Achilles tendon (124/252 third; 49.2% vs. 81/252 third; 32.1%, $p=0.0001$). Activity status of acromegaly and GH/IGF-I levels were similar in patients with different types of elasticity ($p>0.05$). **Conclusions:** Sonoelastography revealed structural changes in the tendinous tissue of patients with acromegaly, but it was not sensitive enough to reflect changes in the serum levels of GH/IGF-1.

Keywords: Achilles tendon, acromegaly, growth hormone, insulin-like growth factor 1, sonoelastography

Introduction

The tendons are decisive in the transmission of force from muscle to the skeleton and locomotive and supportive functions are dependent on these anatomical structures. Tendons are composed of fibroblast-like cells of mesenchymal origin (tenocytes) embedded in a three-dimensional network of extra-cellular matrix (ECM). The major components of ECM are collagen, which predominantly consists of collagen type I (> 95%) and other types of collagens (types III and V), proteoglycans, fibronectin, and elastin [1].

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Growth hormone (GH) and insulin-like growth factor 1 (IGF-I) have an important role in the regulation of protein synthesis, including collagen and myofibrillar protein, the two most abundant proteins in musculoskeletal tissues. The effects of these two endocrine mediators on connective tissue structure are more pronounced during maturation, e.g., in bone growth [2]. However, the effect of GH/IGF-I on connective tissue, especially at chronically altered levels, remains uncertain in adulthood.

Sonoelastography is a recently developed ultrasound-based method that enables the qualitative visual or quantitative measurements of the mechanical properties of tissue [3-4]. It is based on the principle of tissue deformability upon the application of pressure [3-4]. As musculoskeletal disorders are associated with abnormalities in the muscle and tendon biomechanics, sonoelastography may have potential clinical applications in the musculoskeletal system. The Achilles tendon has provided most of the clinical data available so far for musculoskeletal applications [5-7].

In the present study, we attempted to describe the elastographic appearance of the Achilles tendon in acromegalic patients and to determine whether the blood concentrations of GH and IGF-I were associated with the various sonographic elasticity types of Achilles tendons reflecting the stiffness of this tissue.

Material and methods

This single-center, prospective case–control study was undertaken in the endocrinology department of a tertiary referral center between January 2011 and December 2014. Our local ethics committee for human studies approved the protocol, and all participants provided their informed consent. Patients aged over 65 years or those with a history of diabetes, familial hypercholesterolemia, fluoroquinolone therapy, glucocorticoid therapies within the last 12 months (either orally or locally administered), collagenopathies, seropositive or seronegative enthesopathies, or anti-tumor drug administration were excluded. Forty-two acromegaly patients and 42 healthy volunteers as a control group were included in the study. For each patient we recorded the duration of disease, therapies received, and serum GH/IGF-I concentrations measured within one month of the sonoelastography. The criteria used to define inactive disease were nadir GH < l μg/L after 75 g oral glucose tolerance test and normal age-matched IGF- I levels.

Serum GH was assessed by electrochemiluminescence immunoassay (hGH kit, Roche, Mannheim, Germany). The sensitivity of the method was 0.03 ng/mL. Serum total IGF-1 was assessed by immunometric chemiluminescence assay (IMMULITE 2000, SIEMENS, Gwynedd, United Kingdom). The detection limit for IGF-1 was 20 ng/mL, and no hook effects were observed at levels up to 100,000 ng/mL. Age-adjusted reference ranges were used for the evaluation of IGF-I levels.

All patients and controls underwent a sonoelastography of the Achilles tendon. Sonographic examinations were performed with a real-time sonoelastographic scanner (HI Vision Avius, Hitachi Medical, Japan) and a linear–array transducer with a frequency of 5 MHz–13 MHz by the same experienced radiologist. Each Achilles tendon was examined in axial and longitudinal planes in the prone position and with the foot hanging over the examination bed in a relaxed position [8]. During the sonographic examination, the thickness of the tendon was determined by measuring the anteroposterior diameter in the transverse view at the level of the medial malleolus. The Achilles tendon was divided into the following three parts: proximal third (musculotendinous junction), middle third (2 cm–6 cm above the insertion of the calcaneus), and distal third (insertion at the calcaneus) [7], 252 third of tendons being analysed in every group. The anteroposterior diameters of each third were measured in transverse scans. Real-time sonoelastographic images of the tendon were obtained in the longitudinal plane while the transducer was perpendicular to the tendon to avoid tissue shifting. The local strain was calculated under slight compression and decompression applied using the free-hand technique. The optimal strain was assessed according to the visual indicator for the compression seen on the video screen, which indicates the average strain in the region of interest between two frames. The tissue elasticity distribution was calculated in real-time, and the results were represented on a color map superimposed on the B-mod image. The spectrum of color ranged from blue (hard) to red (soft), and it represented the relative stiffness of the tissue.

At least three real-time sonoelastography images of each tendon third were assessed, and the most representative one was chosen for image interpretation. The definition of elastographic patterns were assessed by two radiologists in consensus. The elasticity of the tendons was divided into two main types: type 1 blue/green (hard tissue) and type 2 yellow/red within green (intermediate–soft tissue), both categories with two subtypes detailed in table I. According to the definition, the elasticity of the tissue was in a spectrum ranging from hard to soft as the type progresses from 1a to 2b.

Statistical analysis

Statistical analysis was performed by SPSS software (Statistical Package for the Social Sciences, version 18.0, SPSS Inc., Chicago, IL). Continuous data are shown as mean ± standard deviation (SD). Normality in the variables was assessed by the Kolmogorov–Smirnov test.

Table I. Elasticity types and appearance in sonoelastography

Types	Tendon Patterns
Type 1 (Hard tissue)	Blue/Green
Type 1a	Blue predominance (blue areas more than 50%)
Type 1b	Green predominance (green areas more than 50%)
Type 2 (Intermediate–Soft tissue)	Yellow/Red within green
Type 2a	Small yellow/Red areas within green predominance (green areas more than 75%)
Type 2b	Small green areas within yellow-red predominance (yellow/red areas more than 25%)

Differences in the continuous variables were assessed by Student's t-test. The difference in the proportions was evaluated using an independent X^2 test. $P<0.05$ was considered statistically significant.

Results

Eighty-four Achilles tendons of 42 acromegaly patients (Male/Female: 15/27, Mean age±SD: 46.7±12.5) and 84 Achilles tendons of 42 control subjects (Male/Female: 15/27, Mean age±SD: 46.9±10.8) were evaluated. Age and sex distributions were similar between the two groups ($p>0.05$). Thirty patients underwent surgery and 12 patients received long-acting somatostatin analogs as primary therapy. Thirteen patients did not require any medical therapy after surgery because remission could be maintained without adjuvant treatment. The median (min–max) concentrations for GH were 1.42 (0.1–50) ng/mL and for IGF-1 287.5 (67.8–1358) ng/mL. According to nadir GH after the oral glucose tolerance test and age-matched IGF-I levels, acromegaly patients were further divided into two subgroups with either active (21 patients) or inactive (21 patients) disease.

The mean thickness of Achilles tendons in patients with acromegaly was significantly higher than that of healthy Achilles tendons (5.1±0.7 mm *vs*. 4.4±0.5, $p<0.001$). The patients with active disease had thicker Achilles tendons (5.5±0.8 mm) than those with inactive disease (4.8±0.5 mm) ($p=0.003$**).** In patients with acromegaly and the controls, gray scale ultrasound images of the Achilles tendon showed a parallel contour with homogeneous and fibrillar echotexture. In acromegaly patients the elastographic images showed almost half of the Achilles tendons as type 2 (124/252 third; 49.2%) and the remaining as type 1 (128/252 third; 50.8%). In healthy individuals, 81 out of the 252 third (32.1%) were

Table II. Sonoelastography findings in healthy Achilles tendon sections.

Patterns	Proximal Achilles (n=84)	Middle Achilles (n=84)	Distal Achilles (n=84)
Type 1a	39 (46.4)	7 (8.3)	-
Type 1b	44 (52.4)	61 (72.6)	20 (23.8)
Type 2a	1 (1.2)	16 (19.1)	51 (60.7)
Type 2b	-	-	13 (15.5)

n – number; results are expressed in n (%).

Table III. Sonoelastography findings in the Achilles tendon sections of 42 acromegaly patients (Active: 21, Inactive: 21) according to the activity status of the disease.

Patterns		Proximal Achilles (n=42)	Middle Achilles (n=42)	Distal Achilles (n=42)
Active disease	Type 1a	21 (50)	2 (4.8)	-
	Type 1b	20 (47.6)	14 (33.3)	10 (23.8)
	Type 2a	1 (2.4)	25 (59.5)	28 (66.7)
	Type 2b	-	1 (2.4)	4 (9.5)
Inactive disease	Type 1a	18 (42.9)	1 (2.4)	-
	Type 1b	23 (54.8)	7 (16.7)	12 (28.6)
	Type 2a	1 (2.4)	33 (78.6)	29 (69)
	Type 2b	-	1 (2.4)	1 (2.4)

All values expressed as n (%). Active disease vs. inactive disease: $p>0.05$ for all the parameters compared. Each of the two groups with active and inactive disease status has126 thirds.

Table IV. Serum growth hormone/Insulin-like growth factor-I levels according to the elasticity types of Achilles tendons in sonoelastography.

	Total number (84)	Serum GH level	Serum IGF-I level
Based on the softest section in each tendon	Type 1a (n=0)	-	-
	Type 1b (n=10)	2.4 (0.1–20.7)	256.3 (102–1232)
	Type 2a (n=67)	1.4 (0.1–50)	321 (67.8–1358)
	Type 2b (n=7)	0.4 (0.1–20.8)	260 (102–708)
Based on the hardest section in each tendon	Type 1a (n=39)	0.8 (0.1–50)	260 (67.8–1358)
	Type 1b (n=45)	1.5 (0.1–50)	315 (91.4–1358)

Among the three sections, the softest and the hardest ones were used to classify the corresponding tendon. GH: Growth hormone, ng/mL, IGF-I: Insulin-like growth factor I, ng/mL. Expressed as median (min–max). No statistically significant difference was found between the groups ($p>0.05$).

Table V. Mean thickness of the Achilles tendons in patients with acromegaly according to the elasticity type in sonoelastography.

Patterns	Thickness (Mean ± SD)	P value
Type 1a (n=42)	1.9±0.9	<0.001 1a vs. 1b /2a /2b
Type 1b (n=86)	3.4±1.6	<0.05 1b vs. 2a / 2b
Type 2a (n=117)	5.1±0.9	>0.05 2a vs. 2b
Type 2b (n=7)	4.7±0.3	

type 2, and 171 out of the 252 third were type 1 (67.9%). Accordingly a significantly higher proportion of acromegaly patients had type 2 sonoelastographic appearance of the Achilles tendon (p=0.0001). The sonoelastographic findings of healthy individuals and acromegaly patients are shown in Tables II and III. The difference between subgroups based on the activity status of acromegaly was statistically insignificant (p>0.05). GH and IGF-1 levels were similar in patients with different sonoelastographic appearances of their Achilles tendons (Table IV). The thickness of Achilles tendons in patients with acromegaly increased as the type progressed from 1a to 2b (Table V).

Discussions

Our study showed that acromegaly patients had thicker tendons that were abnormally softened during sonoelastography. Moreover, Achilles tendon thickening was more prominent in acromegaly patients with active disease status. These findings merit discussion because they may shed light on the effects of abnormalities in GH/IGF-1 axis on the tendinous tissue.

GH and IGF-1 are important for locomotor and supportive functions. They may mediate the adaptation of matrix tissue to a mechanical load. In animal tendons, IGF-1 has a dose-dependent effect on collagen production and cell proliferation, and similar effects are observed after GH administration in muscles and tendons of sheep, rats, and pigs [9-12]. Several human studies have provided information on this topic [13-15]. In patients with acromegaly, high blood concentrations of GH and IGF-I were associated with increased thickness and content of collagen-rich tissue [13,14]. Doessing et al explored the effects of recombinant human growth hormone (rhGH) in healthy subjects to test the hypothesis that GH promotes matrix collagen synthesis in musculotendinous tissue [15]. They found that rhGH caused an increase in matrix collagen synthesis in skeletal muscle and tendon but had no effect on myofibrillar protein synthesis [15]. The authors reported an increase in both systemic IGF-I and tissue IGF-I mRNA, thus suggesting IGF-I signaling [15].

As observed in our study, Achilles tendon thickening can be explained by the abovementioned results [9-15]. However, what could be the reason for the abnormal softening of tendons in acromegaly patients despite the increased collagen content? Two studies provide an indirect answer. In the first study, Nielsen et al showed that mice with low GH/IGF-I signaling due to a defective GH receptor (GHR−/− mice) were found to have less tightly packed collagen fibrils (volume fraction) and a smaller than average collagen fibril diameter. A doubling of GH and IGF-I levels did not change the collagen fibril volume fraction, but it did affect the diameter distribution as it presented more small fibrils than in the control mice [16]. In the second study, Doessing et al found quantitative collagen morphology in the tendons of acromegaly patients [17]. They found a tendency toward a smaller tendon collagen fibril area and a higher concentration of small collagen fibrils in acromegalic patients than in GH-deficient patients. This finding suggests that a chronically high level of GH/IGF1, with a concomitant increase in collagen expression and in absolute collagen synthesis, correlates with a smaller collagen fibril area in the tendon [17].

The recent introduction of sonoelastography into commercially available ultrasound systems has promoted research activity toward the potential clinical applications of this novel method in the musculoskeletal system [5-7,18]. In fact, sonoelastography features have been found to be correlated with histopathological findings as well [19]. In a relevant study, 13 Achilles tendons in 10 cadavers were examined with sonoelastography, and biopsy specimens from these tendons were evaluated histologically [19]. Sonoelastography grading was as follows: Grade 1 indicated blue (hardest) to green (hard); grade 2, yellow (soft); and grade 3, red (softest). All 11 tendon thirds of the histologically normal tendons were verified as normal (grade 1) and sonoelastography revealed 14 of the 14 (100%) tendon thirds with histologic degeneration (grade 2 or 3) [19].

What are the clinical implications of our findings? Sonoelastographic parameters were not correlated with GH/IGF-1 levels in our series. Therefore, we consider that sonoelastography is not sensitive enough to have the potential to be adjunctively used for the followup of acromegaly. Moreover, we cannot draw a clear-cut conclusion because of several limitations to our study. Firstly, a histopathological examination of the biopsy

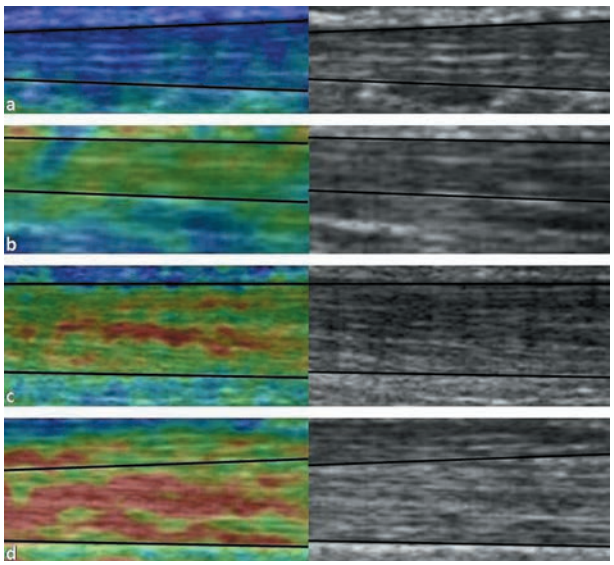

Fig 1. Elastographic color spectrum of healthy Achilles tendons: a) Hardest tendon, type 1a (proximal third of tendon), in a 46-year-old man. b) Hard tendon, type 1b (middle third of tendon), in a 47-year-old man. c) Intermediate–soft stiffness tendon, type 2a (middle third of tendon), in a 40-year-old man. d) Intermediate–soft stiffness tendon, type 2b (distal third of tendon), in a 39-year-old man.

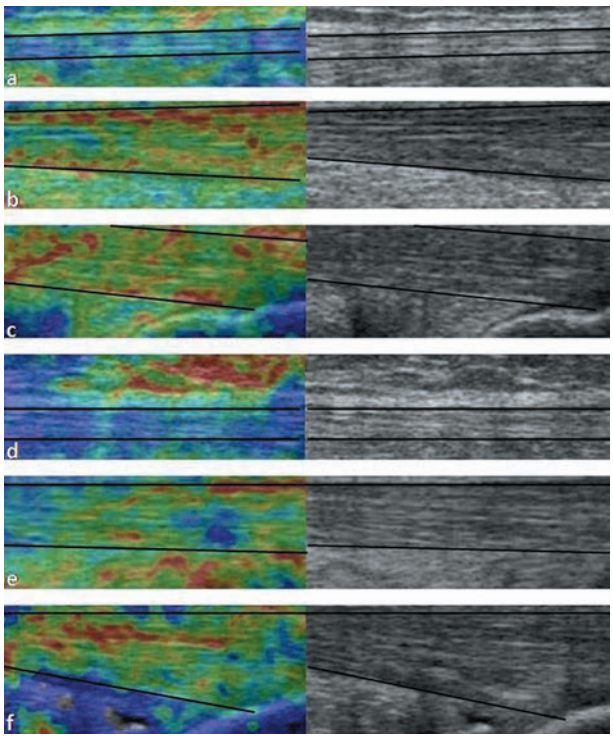

Fig 2. Elastographic color spectrum of tendons in a 41-year-old man with acromegaly. a, d) Type 1a (proximal third of right and left Achilles tendons); b, e) Type 2a (middle third of right and left Achilles tendons); c, f) Type 2a (distal third of right and left Achilles tendons).

samples from the tendons, a prospective followup of our patient group with sonoelastography, and an evaluation of the hormonal status and range of motion would clearly have allowed for a more complete understanding of the role of sonoelastography in acromegaly patients. Secondly, we used a qualitative sonoelastography technique that is prone to inter-reader and intrascan variability. The interpretation of the color scale through a quantitative post-processing method would significantly improve the power of our study, as it would yield an objective measure of tendon stiffness. Thirdly, confirming our findings with a sophisticated method, such as magnetic resonance elastography or shear-wave elastography, would be more effective. Lastly, we did not use the standoff/jelly technique as the tendons were under the subcutaneous fat tissue but not superficial. This technique is recommended for patients with a thin subcutaneous tissue, but the number of these patients was limited. To the best of our knowledge, our study is the first to reveal the structural tendon changes in acromegaly patients using sonoelastography despite the limitations stated above.

Conclusions

Subclinical abnormalities occur in the tendinous tissue of acromegaly patients, and sonoelastography reveals these structural changes. Further studies are required to uncover the relevant pathophysiologic mechanisms and to determine whether sonoelastography can be potentially used to predict the forthcoming locomotor and supportive dysfunction related to an abnormal tendon structure.

Conflict of interest: none

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