Ultrasonic Imaging

INFRARED THERMOGRAPHY, INTRATENDON VASCULAR RESISTANCE AND ECHOTEXTURE IN ATHLETES WITH PATELLAR TENDINOPATHY: A CROSS-SECTIONAL STUDY

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Complete List of Authors:	Molina-Payá, Francisco J.; CEU Cardenal Herrera University - Elche Campus, Department of Physiotherapy, Health Science Faculty Ríos-Díaz, José; Universidad Pontificia Comillas Escuela Universitaria de Enfermería y Fisioterapia San Juan de Dios, Campus San Rafael Carrasco-Martínez, Francisco; Privace Practice. Clínica de fisioterapia F&C Martínez-Payá, Jacinto J.; University of Murcia, Physiotherapy Department, Faculty of Medicine
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Abstract:	Background: Ultrasonographic signs of tendinopathies are an increase in thickness, loss of alignment in collagen fibers and the presence of neovascularization. Nevertheless, analysis of intratendinous vascular resistance (IVR) can be more useful for understanding the physiological state of the tissue. Objective: To show thermal, echotextural and Doppler signal differences in athletes with patellar tendinopathy and controls. Methods: Twenty-six athletes with patellar tendinopathy (PT) participants (30.1 yrs; SD= 9.0 yrs) and 27 asymptomatic athletes (23.3 yrs; SD= 5.38 yrs) were evaluated with thermographic and Doppler ultrasonography (DS). Area of Doppler signals (DS), echotextural parameters (echointensity and echovariation) and IVR were determined by image analysis. The statistical analysis was performed by Bayesian methods and the results were showed by Bayes Factor (BF10: probability of alternative hypothesis over null hypothesis), and Credibility intervals (CrI) of the effect. Results: The absolute differences of temperature (TD) were clearly greater (BF10= 19) in the tendinopathy group (patients) than in controls. Regarding temperature differences between the affected and healthy limb, strong evidence was found (BF10= 14) for a higher temperature (effect= 0.53 ºC; 95% CrI=0.15 to 0.95 ºC) and very strong for reduced IVR compared (BF10= 71) (effect= -0.67; 95% CrI=-1.10 to -0.25). The differences in area of DS (BF10= 266) and EV (BF10= 266) were higher in tendinopathy group. TD showed a moderate positive correlation with VISA-P scores (tau-B=.29; 95% CrI=.04 to .51) and strong correlation with IVR (r=-.553; 95%CrI=-.75 to -.18).

	Conclusion: Athletes with patellar tendinopathy showed a more pronounced thermal difference, a larger area of Doppler signal, a lower IVR and a moderately higher echovariaton than controls. The correlation between temperature changes and IVR might be related with the coexistence of degenerative and inflammatory process in PT.

1. INTRODUCTION

Patellar tendinopathy (PT) is characterized by localized pain in the patellar tendon, and is related to load, which increases with the mechanical demand on the knee extensors, especially in activities that load and release energy on the patella [1]. It is a common pathology of the knee with a high occurrence in both athlete and non-athlete populations [2]. Athletes with PT can suffer from uncomfortable symptoms accompanied by decreased function with a negative effect on their quality of life [2], deterioration of their physical performance [3], and even the need to end their sporting career prematurely [4]. It is difficult to determine the exact frequency of PT in athletes because this type of sports injury is often underreported. Its incidence is higher in jumping sports and in sports that require repetitive loading of the patellar tendon, but its prevalence among elite athletes in different sports has been estimated to be around 14% [3].

The diagnosis of PT is challenging, as there is currently no gold standard diagnostic technique [5]. In clinical practice, it is diagnosed by antecedents, knee examination, and palpation of the tendon and its attachments [6]. However, the physiology of tendon pain is not yet fully understood [7].

Diagnosis can be confirmed by imaging tests, commonly ultrasonography and magnetic resonance imaging, which detect abnormalities in the structure of the patellar tendon, such as increased

thickness, loss of alignment in collagen fibers and the presence of neovascularization [8]. More specifically, morphological, and echo-textural analysis of the tendon using ultrasound image analysis has proven to be reliable [9] and useful for assessing changes in tendon structure [10]. In addition, the possibility of assessing tendon vascular perfusion by using analysis of intratendinous vascular resistance (IVR) based on Doppler ultrasound image analysis and the resistance index (RI) formula has recently been demonstrated [11]. The ultrasound system expresses numerically the tissue resistance to flow originating from the microvascular bed distal to the measurement site [12]. Low vascular resistance is associated with high perfusion of the distal bed and therefore with a situation compatible with the presence of inflammation [13,14].

However, although it is generally considered that the presence of an intratendinous Doppler signal is associated with a sign of tendon abnormality [15] and its absence with healthy tendons [16], its presence has also been detected in asymptomatic subjects [17], leading to debate about its usefulness. Nevertheless, analysis of IVR can be more useful for understanding the physiological state of the tissue[18].

In this context, some studies have also considered infrared thermography (IRT) as a suitable technique for the diagnosis of PT. IRT is an inexpensive, reliable, non-invasive, and accurate imaging technique that provides information on thermal, metabolic, and

vascular conditions of the human body that can be used to interpret pathophysiological changes [19,20]. The use of IRT in musculoskeletal pathology is based on the thermal symmetry between both sides of the body, so that the presence of thermal asymmetry is indicative of some type of anomaly, especially in the knee when the difference is greater than 0.5 ºC [21,22].

Yet there are no studies that have evaluated and related tendon echo-textural changes, IVR, thermal alterations, and functionality in athletes with PT. The information offered by all these imaging techniques can help us better understand the pathological process of PT from both a structural and physiological point of view and establish a better therapeutic strategy. Therefore, the objectives of this study were 1) to show the prevalence of abnormalities in thermal, vascular, and echo-textural parameters between athletes with PT and asymptomatic athletes; 2) to analyze the evidence of changes in thermal, vascular and eco-textural parameters in athletes with PT between symptomatic and asymptomatic tendons from a Bayesian perspective, and 3) to explore the Bayesian plausibility of relationships between symptomatology and thermal asymmetry with respect to vascular and echo-textural parameters.

2. MATERIALS AND METHODS

2.1. Sample size

Given the exploratory aim of this study, a sample of 27 athletes with chronic unilateral tendinopathy and a control group of asymptomatic athletes (n=27) were recruited in a non-randomized, intentional, and consecutive manner.

2.2. Study design and participants

The study included 26 patients (athletes with PT) (mean age= 30.1 years; SD= 9.0 years; range= 18-50 years) of whom 21 were male (81%), and a control group of 27 asymptomatic athletes (mean age= 23.3 years; SD= 5.38 years; range=19-42 years) of whom 12 (44%) were male. Of the 27 symptomatic athletes, 26 of them had symptoms between 3 and 60 months (0.3 to 5 years). One participant was excluded because he presented an evolution of 120 months (10 years), twice or more than the rest of the participants. The exclusion was made before implementing the statistical analyses. All the participants were federated athletes in different sports.

All the participants were recruited voluntarily from a private physical therapy center (Clinica F&C Fisioterapia Avanzada y Neurorehabilitación, Huelma, Spain) in July 2019. All participants were informed of the study's objectives and signed an informed consent

document. The study was approved by the Ethics Committee of the Catholic University of Murcia (CE111803).

Demographic and clinical characteristics (sex, age, and time of evolution) were recorded. Knee functionality was assessed with the Victorian Institute of Sport Patellar Tendon Assessment Questionnaire (VISA-P) [23] which is simple and practical, with good clinimetric properties [24] and widely used in PT research.

Participants were divided into two groups: 1) Twenty-seven athletes suffered from unilateral PT which had been diagnosed with clinical criteria, an evolution time of more than three months and a VISA-P score of less than 100; 2) Twenty-seven healthy volunteer athletes that had been recruited (not matched) under the inclusion criteria of no previous PT and a VISA-P score of 100.

2.3. Data sources, measurements, and outcomes

Thermographic analysis of patellar tendon

To minimize the influence of extrinsic individual factors on vascular and thermographic recordings, all participants were asked not to exercise, not to drink alcohol, coffee, or energy drinks in the previous 12 hours, and not to smoke in the previous 6 hours. They were asked not to apply creams or lotions on their legs to avoid alterations in skin emissivity, as well as to report the use of drugs or treatments and to try to avoid altering their rest and mealtimes [25].

The IRT images were recorded with an OPTRIS PI 450 IRT camera coupled to Optris PI Connect Software (Germany). The IRT camera has a Noise Equivalent Temperature Difference <40 mK with 38º x 29º FOV, a wide temperature range of −20°C to +100°C, a spectrum range of 7.5–13 µm, a focal plane array sensor size of 382 × 288 pixels, an emissivity set at 0.98 and a measurement uncertainty of ± 2% of the overall temperature reading. The capture frame size was 55.4 × 40.63 cm (1.5 mm/px).

The participant was acclimatized in an isolated room (3.86 x 3.47 m^2), with no temperature source, and at a mean temperature of (23.7 ± 0.9ºC) and a relative humidity of (49 ± 5%) for 15 min without clothing on the lower limbs [25]. The participant was seated on a hydraulic stretcher with his/her feet on a step to ensure no contact with the ground and the camera was positioned perpendicular to the subject for a more accurate reading [25].

For determination of the regions of interest (ROI) of the patellar tendon, the method previously described by [26] was used by superimposing ROI of the patellar tendon using skin markers, of a thermal image on another previous thermal image without thermal contamination due to the contact of the manipulation of skin markers, avoiding distorsion of the results caused by contact with the patient´s skin, avoiding distortion of the results caused by contact with the patient's skin (**figure 1**).

FIGURE 1 ABOUT HERE

The average temperature of each tendon was obtained and subsequently the temperature difference of both. A difference greater than 0.5 ºC between the affected side and the healthy side is compatible with tendon pathology [21,22].

Quantification of intratendinous vascular resistance with Doppler Ultrasonography

IVR was determined using the RI from power Doppler (PD) ultrasonography records.

$$RI = \frac{peak\ systolic\ velocity - diastolic\ velocity}{peak\ systolic\ velocity}$$

The patient was placed in supine position with knees extended and both knees were evaluated. The scan was performed with a Telemed SmartUS ultrasound apparatus (Vilnius, Lithuania) with a 7-15 MHz linear transducer (L15-7L40H-5). The setting parameters were the same for all patients and care was taken not to apply pressure with the transducer. The Power Doppler settings were Doppler frequency of 6.7 MHz and a PRF of 0.7 kHz, with the lowest wall filter and a Doppler gain just below the level of random noise production.

Since the use of spectral Doppler mode for quantification of intratendinous vessels can be difficult due to the high number and low diameter of the signals, the patellar tendon was scanned using Power Doppler mode in the longitudinal plane to localize the maximum

intratendinous Doppler signal and the 4-second video was stored for further processing and analysis. The image processing and analysis method to obtain the IVR has been described previously [18]. The method was based on the measurement of pixel colour intensity on the intratendon Doppler signals, considering this data as a flow velocity. In this procedure, the intensity of the intratendon Doppler signals of the image with the highest Doppler signal of the cardiac cycle was considered as the peak systolic velocity, and of the image with the lowest signal as the end-diastolic velocity. The measurements obtained were transferred to the RI formula, obtaining a value of the IVR (**Figure 2**). In tendons where no Doppler signal was recorded the RI was equal to 1, which represents normality in musculoskeletal tissue [13,27].

FIGURE 2 ABOUT HERE

Textural analysis of patellar tendon

The textural analysis of the tendon was performed on B-mode ultrasound images, both in longitudinal and transverse sections. The resulting bitmaps had a resolution of 660x 556 pixels (16.5 px/mm) with 256 grey levels and were stored as *.TIFF* files without compression or losses [28]. Image processing and analysis was performed by the same researcher using ImageJ (v. 1.53) software. The ROI was selected using the ROI Manager application for ImageJ, which were 165 x 42 px

for the longitudinal section and 247 x 42 px for the transverse section on an 8-bit grey scale, by a researcher blinded to the diagnosis.

In the transverse section, the ROI was placed in the center of the tendon. In both cases the ROIs were prevented from crossing the paratenon as shown in **figure 3**.

FIGURE 3 ABOUT HERE

Echointensity (EI) and echovariaton (EV) were assessed. EV can be interpreted as a textural parameter [29] and was determined by the relation between the standard deviation and the mean pixel intensity obtained from the histogram. In quantitative terms, a symptomatic tendon is characterized by reduced echogenicity and increased variance [30,31].This methodology has good reliability and reproducibility [9].

EV =σ/μ • 100 where σ is the standard deviation of the image intensities, and μ is the mean value of the intensity in each ROI.

For convenience we will refer to both EI and EV as echo-textural parameters to differentiate them from other ultrasound variables.

2.4. Statistical methods

Data analysis was performed with the statistical package R v.3.6 and JASP Team (2020). JASP (Version 0.14.1) [Computer software], and Morey, R. D., & Rouder, J. N. (2015). Bayes Factor (Version 0.9.11-3) [Computer software].

Data were summarized using mean and standard deviation, range and quartiles for continuous variables and absolute and relative frequencies for categorical variables.

1) For comparing the presence of abnormal thermal asymmetry, Doppler signal, bilateral Doppler signal, pre-abnormal vascular resistance and bilateral abnormal vascular resistance between control and tendinopathy group, the odds ratios (95% interval of confidence) and p-values (level of significance ≤.05) were obtained with logistic regression analysis [32].

2) For comparisons of thermographic, vascular, and textural ultrasonographic differences between patients and controls, independent sample means were compared by Bayesian statistics.

Since no previous evidence is available, the prior was described in a conservative manner by a Cauchy distribution centered around zero and with a width parameter of 0.707. This corresponds to a probability of 50% that the effect size lies between -0.707 and 0.707. [33] In addition, since differences between the parameters of interest, if any, were assumed to be in a certain direction, one-sided contrasts were performed.

We give the Bayes factor (BF_{10}), interpreted as the number of times that the observations are more likely to support the hypothesis of interest than the null hypothesis (in this case no difference). This is a direct indicator of the degree of evidence provided by the observations

and, as only a guide only, could be considered anecdotal (BF 1 to 3), moderate BF (3 to 10), strong (10 to 30), very strong (30 to 100) and extreme (>100). The median effect size and credibility intervals are also provided, and it is assumed that if they contain 0, the differences between groups are not credible[33].

3) Comparisons for symptomatology, and thermographic, vascular and ultrasonographic parameters within the patient group were performed by paired sample means Bayesian analysis. In this case, two analyses were performed, the first with all patients and the second one excluding patients with a bilateral IVR less than one.

4) Finally, to explore the relationship between VISA-P, time of evolution and thermal asymmetry with vascular and ultrasonographic parameters, a Bayesian correlation analysis was performed. Kendall's *tau*-B correlation coefficient was used for contrast with VISA-P (ordinal variable), time of evolution (no normal distribution) and Pearson's *r* coefficient for the contrasts between thermal asymmetry and vascular and ultrasonographic variables. In addition, the credibility interval for the coefficient, the effect size R^2% (interpreted as % of explained variance), and BF10 are shown [34].

3. RESULTS

3.1. Sample characterization and vascular assessment

Both age (mean dif.=6.8 years; 95%CI= 2.7 to 10.9 years; *p*-value= .001) and sex were significantly different (χ^2 =5.97; d.f.=1; *p*-value=.0015) between the two groups and will therefore be entered as covariates in the models that require them.

The mean VISA-P score was 67.3 (S.D.=17.9) with a range from 29.0 to 94.0 points (median= 68.5; interquartile range= 56.3-81.5) and the mean time of evolution was 22.3 months (SD=16.9; range=3-60; median =13.5; IQR=12.0-34.5 months).

At the time of the study, practicing sports was 3.2 times higher in the tendinopathy group than in the asymptomatic group (*p*-value= .055). About 90% of the participants in both groups had a dominant right leg. Within the tendinopathy group, no relationship was found between the presence of tendinopathy and dominance (approximately 46% of the affected tendons were in the dominant leg).

In 73% of cases the maximum temperature was recorded in the affected leg, although only 46% of the differences were abnormal (>0.5 ºC). However, an abnormal difference was found to be 7.3 times more likely among the tendinopathy group than among the asymptomatic group (*p*-value= .004).

In the tendinopathy group, Doppler signals were found in 85% of the affected tendons, while Doppler signals also appeared in 50% of the unaffected tendons. Doppler signals also appeared in 30% of the tendons of asymptomatic subjects. However, the presence of Doppler signals in the tendons of patients (regardless of whether the tendon is affected or not) was 28.5 times more frequent (*p*-value <.001) than in asymptomatic subjects; and the presence of bilateral Doppler signal was 20.8 (*p*-value <.001) more likely in patients than in the control group.

Finally, a regard the vascular resistance, it was found that abnormal RI was 25.3 times more likely in the tendinopathy group than in the asymptomatic group. In addition, bilateral RI was found in 33% of the patients.

3.2. Thermographic, vascular, and textural ultrasonographic differences between patients and controls

This section shows an analysis of the differences in absolute values between knees (side by side) for each of the groups (**Table 2**).

The absolute differences of temperature (ºC) were clearly greater (BF_{10}= 19) in patients than in controls. The differences in area of Doppler signals were also larger (BF_{10}= 266) which can be taken as an extreme degree of evidence.

The differences in maximum systolic velocity (m/s), final diastolic velocity (m/s) and vascular resistance were smaller in patients than in the controls, with a moderate to strong degree of evidence (BF_{10} =17 for vascular resistance).

Regarding the differences in echo-textural parameters, only the changes in EV (both longitudinal and transversal section) showed higher values in the patients, especially in the transverse section (BF_{10}=11). This means that the EV between the injured and healthy tendon was higher than in the controls.

3.3. Thermographic, vascular, and textural ultrasonographic differences between injured and healthy tendons

This section shows the differences between injured and uninjured tendons (in patients' sample) (**Table 3**).

Regarding temperature differences between the affected and healthy limb, strong evidence was found (BF_{10}= 14) for a higher temperature in the affected tendon (effect= 0.53 ºC; 95% CrI=0.15 to 0.95 ºC). Similarly, there was very strong evidence (BF10= 71) in favor of reduced vascular resistance in affected tendons compared to healthy tendons (effect= -0.67; 95% CrI=-1.10 to -0.25).

As regards Doppler signals characteristics, there was moderate evidence (BF10= 5.2) for increased Doppler signals area in the affected tendons (effect=0.44 mm^2; 95%ICr=0.0 to 0.84), and strong evidence

for great maximum systolic velocity (BF_{10}= 17) and final diastolic velocity (BF_{10}= 18).

Since the presence of abnormal bilateral vascular resistance may affect the interpretation of thermal differences, a sensitivity analysis of these same variables was performed excluding cases with bilateral IVR (n=18), the results of which are shown in **Table 4**.

In this case, the trends found were maintained, although the effect of the difference in temperatures and areas was slightly reduced. However, vascular resistance (together with systolic and diastolic velocities), as expected, showed more pronounced differences, with extreme evidence (BF_{10}= 395) in favor of a lower temperature in affected tendons (effect =-1.04; 95%ICr=-1.7 to -0.45).

The echo-textural variables (EI and EV) also showed no evidence of differences between pathological and healthy tendons in either longitudinal or cross-sectional ultrasound slices.

3.4. Relationships between clinical variables and thermographic, vascular, and textural ultrasonographic parameters

Finally, this section shows the relationships between the clinical variables VISA-P and time of evolution with the physiological parameters in the sample of injured tendons (note that in these cases only the values of the injured tendons were analyzed).

In addition, the correlations between temperature differences between the affected and healthy limbs compared with the differences in vascular and ultrasound variables are shown in **Table 5**.

VISA-P scores only showed a positive correlation (*tau*-B=.29; 95% CrI=.04 to .51) with the thermal difference between limbs, although with a moderate degree of evidence.

Time of evolution was not related with any of the thermal, vascular or ultrasound variables.

The third aim was to determine the relationship between thermal asymmetry and the rest of the vascular and echo-textural variables (**table 5**).

Strong evidence of correlation was found between thermal asymmetry and the variables maximum systolic velocity **(figure 4)** (BF_{10}=128; r=.63; 95%CrI=.29 to .80), final diastolic velocity (BF_{10}=60; r=.59; 95%CrI=.24 to .78) and vascular resistance (BF_{10}=25; r=-.553; 95%CrI=-.75 to -.18). No correlations were found between thermal asymmetry and differences in EI and EV.

4. DISCUSION

In this study, the use of different imaging techniques has made it possible to analyse the patellar tendon of athletes from a structural point of view, using EI and VE, and physiological, using IRT and IVR

analysis, providing information on degenerative and inflammatory processes respectively.

4.1. Technical issues

Quantitative analysis in thermal imaging depends on the selection of the ROI [26,35]. Previous studies analyzing patellar tendons included the entire knee area [19,36]. By selecting a ROI of the entire knee, specificity is lost since other structures that may alter the result are included. In this study a previously designed protocol[26] was used for the specific selection of the patellar tendon in which excellent intraobserver and interobserver reliability was found for the variables of ROI position and size, and mean temperature. All the lower limits of the intraclass correlation coefficient were below 0.84 with no bias [26].

The DS is quantified by the automatic calculation of RI by the ultrasound scanner, measuring one vessel at a time [37]. At intratendinous level, the high number of vessels of small size makes it difficult to calculate the IVR. Some authors have tried to solve this problem by calculating the mean RI of the three largest vessels [13,27]. In the present study, to average the vascular resistance of all the intratendinous vessels, a previously developed methodology based on pixel color intensity was used, with excellent reliability [18].

4.2. Differences between patients and controls

The skin temperature distribution of a healthy human body shows contralateral symmetry, so that thermal asymmetry may be an indicator of abnormality [38]. When the thermal difference between the knees of patients and the control group was compared, the difference was observed to be greater in the patient group. This result agrees with those of Seixas (2013), who analyzed the knee temperature of twenty male volleyball players, and found that even mild tendinopathy can affect the skin temperature of the affected knee when compared with a control group with no history of tendon pathology.

Intratendinous Doppler signals were present in 85% of the affected tendons of the patients and 50% of the unaffected ones. It should be noted that 30% of asymptomatic subjects also had Doppler signals, analogous that reported by other authors (29%) in asymptomatic athletes [39]. These data may suggest that the presence of intratendinous Doppler signals is not always a pathological sign [40]. This idea is reinforced by findings that intratendinous Doppler signal can increase with exercise, even in asymptomatic patients [41]. In addition, intratendinous flow may appear as part of the normal adaptive physiological response to loading [42].

In our study, the presence of Doppler signals in patients' tendons was found approximately 29 times more frequently than in asymptomatic subjects, so we cannot completely rule out this variable as one of the

signs present in tendon pathology [15]. The greater presence of bilateral Doppler signal in patients reinforces the idea that this parameter is a sign to be considered, as there is a greater probability of also finding similar deterioration in the contralateral tendon [43].

In our study, IVR was calculated by image analysis of intratendinous Doppler signal and using the RI formula. As with the RI, a low value is associated with low peripheral resistance and high perfusion of the distal bed and hence a situation compatible with the inflammatory process [14,27]. A low vascular resistance was 25.3 times more likely to be found in patients than in the control group, and there was strong evidence (BF10 = 71) for a lower vascular resistance in affected tendons than in healthy ones. This would have added to the higher temperature observed in the affected tendons, indicating a probable inflammatory process, in line with the current trend to consider tendinopathy as a pathophysiological process in which degenerative and inflammatory state coexist [44,45].

As regards echo-textural parameters, EI has been extensively studied in the musculoskeletal system [9,46], while very few authors have analyzed the EV, focusing mainly on muscle and nervous tissue [47,48]. This is even though EV could provide more information on the structural characteristics of the tissue than EI, which only provides the mean intensity. When analyzing changes in EV among patients' tendons, higher values were observed than in controls, reflecting

changes in the ultrastructural pattern of the tendon, probably caused by a degenerative process. Although the presence of hypoechogenic regions and thickening in symptomatic tendons has been described [5], we found no differences in EI between groups, probably because the hypoechogenic regions may be found also in asymptomatic patellar tendons.

4.3. Differences between injured and healthy tendons

In the group of athletes with PT, symptomatic patellar tendons displayed thermal asymmetry, as defined by other authors [19,38]. This thermal asymmetry was shown as a warm pattern of the affected tendon (effect = 0.53 ºC; 95% CrI = 0.15-0.95 ºC), although only 46% of the differences were greater than 0.5ºC, which, despite the low percentage, was 7.3 times more likely to be found in the patients than in the control group. This same warm pattern has also been observed in other studies [19,36] and in other types of tendinopathies [50].

Two patients had an affected tendon with a cold pattern compared with the contralateral tendon. Some authors have attributed this cold pattern to chronic PT [19,20]. In the results analyzed, we found no relationship between chronicity and temperature, vascular or echo-texture variables, so we could discard this hypothesis. Moreover, this association does not correspond with current views that propose the coexistence of the inflammatory and degenerative process in tendinopathies [51,44]. Along these lines, there is a possibility that the

asymptomatic contralateral tendon will present an inflammatory process, with an increase in temperature, thus diminishing the thermal difference with the affected tendon that may even show a higher temperature, the symptomatic tendon showing hypothermia. This hypothesis is reinforced by the observation that tendons in the control group with a thermal asymmetry greater than 0.5 ºC appear despite being asymptomatic. In this same line, Liu et al. (2020) also found 28% of asymptomatic collegiate athletes with a thermal asymmetry greater than 0.5 ºC).

To avoid the possibility that an asymptomatic inflammatory process of the contralateral tendon might alter the thermal difference, a sensitivity analysis of the same variables was performed, excluding cases with a bilateral anomalous vascular resistance, observing a similar trend in the results. Another proposed hypothesis is that this cold pattern is due to an activation of the sympathetic nervous system, which decreases microcirculation and local perfusion [52]. The temperature of the skin overlying the patellar tendon may directly reflect underlying vascular disturbance and tissue metabolism, and therefore could translate into a cold pattern, although it is unknown whether these effects occur before the tendon becomes painful and at what level they become clinically relevant [53].

We observed that, unlike between-group comparisons, there was only moderate evidence (BF10 = 5.2) of an increase in Doppler signal area

in affected tendons. In contrast, there was strong evidence (BF10 = 71) of abnormal vascular resistance in affected tendons compared to healthy tendons, suggesting that analysis of IVR may provide more clinical information than Doppler area quantification.

Although higher EV values were found in the tendons of patients than in those of the control group, these changes were not reflected when comparing the affected tendons of the patients with the asymptomatic contralateral tendon, and no changes in EI or EV were evident. This finding is probably because the contralateral asymptomatic tendon may also be undergoing degenerative echo-textural changes even though it is asymptomatic.

It must be borne in mind that it is not always possible to establish a direct relationship between pathological imaging and symptomatology [54], since pathological images have been found in asymptomatic subjects, and symptomatic tendons in normal images [55]. Some abnormal images may even be due to different factors, such as an adaptation of the collagen fibers to the activity performed [56].

4.4. Relationships between clinical variables and thermographic, vascular, and textural ultrasonographic parameter

The thermal difference between patients' knees showed a correlation with the VISA-P score, a finding in line with the consulted literature, where a positive correlation was found between pain and thermal

abnormalities [19]. Seixas et al. (2013) also reported that increased skin temperature in the knee of subjects affected by tendinopathy could correlate with a decrease in the VISA-P scale score. In our study was a moderate degree of evidence of correlation, so it should be remembered that symptomatology is not always accompanied by changes in physiological parameters or in the imaging techniques themselves [54].

It has been revealed that the presence of intratendinous hypervascularization does not necessarily correspond with pain or clinical observations [57]. This relationship was not found in our study, either.

Contrary to what we observed in the analysis of thermal asymmetry, the lack of an association between the analyzed variables of echo-textural and knee pain and functionality (VISA-P) suggests that the use of EI and EV may not be the best biomarkers to characterize PT symptomatology.

Thermal asymmetry and abnormal IVR can be considered signs of an inflammatory process and as expected, strong evidence of a negative correlation between them was found in this study. In contrast, no relationship was found between these variables and time of evolution, probably because during the evolution of the pathology, the inflammatory process may be present to a greater or lesser extent [44]. Nor was a correlation found between thermal asymmetry and

differences in EI and EV, although it must be borne in mind that structural changes are analyzed with ultrasound, while physiological changes are studied with IRT [58].

4.5. Limitations of the study

Although the methodology used in this study is highly reliable, it has several limitations. First, both age and sex were significantly different between the two groups., and these variables can be considered as influencing factors in the thermographic recordings performed [25]. To minimize the influence of these variables, the analysis was performed on the thermal differences between knees and the statistical analysis was adjusted by age and sex.

The type of sport practiced by the participants was also not considered, which could affect to a greater or lesser extent, the structural or physiological characteristics of the patellar tendon [55,56]. Analysis of the influence of the type of sport on the thermal, echo-textural, and vascular variables analyzed in this study could provide information on the impact of the type of sport on the patellar tendon.

Due to the complexity and time required in performing these image analyses, their application in daily clinical practice may be difficult. It would be interesting to conduct future research, in collaboration with engineers and manufacturers, to incorporate these image analysis tools into ultrasound devices.

5. CONCLUSION

As conclusions, athletes with PT showed a more pronounced thermal difference, a larger area of Doppler signal, a lower IVR and a moderately higher EV than controls

In the patient group, the affected tendon had a higher temperature, a lower IVR and a moderately higher Doppler signal area compared with the contralateral asymptomatic tendon, while no differences were observed between EI and EV parameters.

A relationship was found between a greater temperature difference between patellar tendons and increased pain and decreased functionality of the patient's knee, as measured by the VISA-P scale.

Athletes with tendinopathy showed a correlation between thermal asymmetry and low IVR, inflammatory signs that might be related with the coexistence of the degenerative and inflammatory process in PT, reinforcing the latest findings in tendon pathophysiology, which in the future could lead to new strategies for imaging evaluation and treatment.

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Table 1. Frequencies and Odds Ratios for sociodemographic variables and vascular parameters.

Variable	Tendinopathy (n=26)		Healthy (n=27)			
	n	%	n	%	OR (95%IC)	*p*-value
Sex (male)	21	80.8	12	44.4	5.4 (20.4 to 1.5)	**.006**
Sport practice	20	76.9	14	51.9	3.2 (0.87 to 11.2)	.055
Dominant leg (right)	24	92.3	24	88.9	1.5 (9.8 to 0.23)	.669
Injured leg (right)	12	46.2	--		--	--
Injured leg is dominant leg	12	46.2	--		--	--
Difference of temperature (abnormal)	12	46.2	3	11.1	7.3 (1.4 to 39.2)	**.004**
Knee with maximum temperature (injured)	19	73.1	--		--	--
Doppler signal (tendons)	24	92.3	8	29.6	28.5 (5.4 to 150.2	**<.001**
Doppler signal bilateral	11	42.3	1	3.7	19.1 (2.2 to 162.2)	**<.001**
Doppler signal injured leg	22	84.6	--		--	--
Doppler signal non-injured leg	13	50.0	--		--	--
Vascular resistance (abnormal)	22	84.6	5	18.5	28.9 (5.2 to 140.0)	**<.001**
Vascular resistance bilateral (abnormal)	8	30.8	0	0.0	--	--

The reference categories marked in parentheses have been used to calculate the odds ratios (OR).

Table 2. Thermographic, vascular and ultrasonographic differences between patients and controls.

Variable	Group	Mean	SD	Min	Q1	Median	Q3	Max	BF_{10}	Effect size (median)	Lower 95% CrI	Upper 95% CrI	Evidence
Differences in temperature	Control	0.24	0.20	0.0	0.10	0.20	0.35	0.80	19	-0.72	-1.28	-0.19	Strong*
	Tendinopathy	0.49	0.36	0.0	0.20	0.40	0.78	1.5					
Differences in Doppler area	Control	0.42	0.85	0.0	0.00	0.00	0.30	3.40	266	-1.01	-1.56	-0.43	Extreme*
	Tendinopathy	2.1	1.91	0.0	0.83	1.7	2.7	9.2					
Differences in maximum systolic velocity	Control	5.6	9.52	0.0	0.0	0.00	11.1	23.2	4.3	-0.54	-1.08	-0.09	Moderate*
	Tendinopathy	12.0	11.08	0.0	1.0	12.7	22.9	25.5					
Differences in final diastolic velocity	Control	3.7	7.95	0.0	0.0	0.0	0.0	20.9	18	-0.71	-1.28	-0.19	Strong*
	Tendinopathy	11.1	10.18	0.0	0.4	19.2	20.3	23.5					
Differences in vascular resistance	Control	0.17	0.36	0.0	0.0	0.0	0.0	1.00	17	-0.71	-1.27	-0.19	Strong*
	Tendinopathy	0.49	0.42	0.0	0.0	0.8	0.9	1.0					
Differences in longitudinal echointensity	Control	6.4	4.58	0.0	2.6	6.7	8.7	18.8	0.7	0.30	0.02	0.77	--
	Tendinopathy	5.1	4.29	0.3	1.6	4.0	7.4	15.5					
Differences in longitudinal echovariaton	Control	3.7	3.35	0.0	0.9	3.3	4.5	12.7	5.6	-0.569	-1.12	-0.10	Moderate*
	Tendinopathy	7.7	7.99	0.0	1.9	5.3	10.9	34.0					
Differences in transversal	Control	3.8	2.54	0.1	1.9	3.2	5.4	10.1	0.3	-0.18	-0.60	-0.01	--

echointensity	Tendinopathy	4.0	3.64	0.3	1.6	2.4	5.9	15.6				
Differences in transversal echovariaton	Control	3.5	2.70	0.2	1.5	2.9	5.8	9.4	11	-0.657	-1.21	-0.15	Strong*
	Tendinopathy	7.2	6.38	0.0	2.6	6.0	8.5	25.8				

SD: standard deviation. Min: minimum. Q1: 1st quartile. Q3: 3rd quartile. Max: maximum. BF_{10}: Bayes factor as the number of times in favor of the evidence of the alternative hypothesis over the null hypothesis. 95% CrI: 95% credible interval. *: a priori unilateral contrast. It can be considered that when the credibility intervals continue to have an effect of 0, there is no evidence of differences between groups.

Table 3. Thermographic, vascular and ultrasonographic differences between symptomatic and non-symptomatic tendons.

Variables (n=26)	Mean	SD	Min	Q1	Median	Q3	Max	BF_{10}	Effect size (median)	Lower 95%CrI	Upper 95%CrI	Evidence
Temperature symptomatic leg (ºC)	31.1	1.66	27.9	30.1	31.3	32.7	33.8	14	0.53	0.15	0.95	Strong*
Temperature non-symptomatic leg (ºC)	30.8	1.71	27.7	29.9	31.1	32.0	33.9					
Area symptomatic leg (mm^2)	2.57	2.41	0.00	1.03	1.95	3.55	9.20	5.2	0.44	0.09	0.84	Moderate*
Area non-symptomatic leg (mm^2)	1.34	2.12	0.00	0.00	0.20	1.78	9.10					
Maximum systolic velocity symptomatic leg	19.6	8.62	0.0	21.9	22.4	23.7	26.9	17	0.55	0.16	0.97	Strong*
Maximum systolic velocity non-symptomatic leg	11.0	11.27	0.0	0.0	10.0	22.1	23.6					
Final diastolic velocity symptomatic leg	16.5	8.3	0.0	19.0	20.2	20.6	23.5	18	0.55	0.16	0.97	Strong*
Final diastolic velocity non-symptomatic leg	8.6	10.3	0.0	0.0	0.0	19.6	21.7					
Vascular resistance symptomatic leg	0.31	0.35	0.05	0.10	0.15	0.30	1.00	71	-0.67	-1.10	-0.25	Very strong*
Vascular resistance non-symptomatic leg	0.69	0.44	0.05	0.13	1.00	1.00	1.00					
Longitudinal echointensity symptomatic leg	22.5	8.48	5.9	17.6	22.9	28.8	36.6	0.27	-0.13	-0.50	0.24	--
Longitudinal echointensity non-symptomatic leg	23.4	6.67	10.2	18.4	24.2	27.7	34.7					
Longitudinal echovariaton symptomatic leg	34.1	10.28	19.4	26.1	31.9	41.6	53.5	0.23	0.085	-0.28	0.45	--
Longitudinal echovariaton non-symptomatic leg	33.1	9.32	17.8	28.3	30.8	36.6	52.3					

Variables (n=26)	Mean	SD	Min	Q1	Median	Q3	Max	BF_{10}	Effect size (median)	Lower 95%CrI	Upper 95%CrI	Evidence
Transversal echointensity symptomatic leg	35.5	7.70	13.6	31.7	36.6	42.1	45.3	0.21	-0.021	-0.38	0.34	--
Transversal echointensity non-symptomatic leg	35.7	7.11	23.4	29.4	35.1	43.1	47.1					
Echovariaton symptomatic leg	31.0	7.65	22.3	25.4	28.3	34.6	48.9	0.27	0.14	-0.23	0.51	--
Transversal echovariaton non-symptomatic leg	29.5	8.73	20.4	23.5	26.4	32.0	53.3					

SD: standard deviation. Min: minimum. Q1: 1st quartile. Q3: 3rd quartile. Max: maximum. BF_{10}: Bayes factor as the number of times in favor of the evidence of the alternative hypothesis over the null hypothesis. 95% CrI: 95% credible interval. *: a priori unilateral contrast. It can be considered that when the credibility intervals continue to have an effect of 0, there is no evidence of differences between limbs.

Table 4. Thermographic, vascular and ultrasonographic differences between symptomatic tendons and non-symptomatic tendons (excluding bilateral RI).

Variables (n=18)	Mean	SD	Min	Q1	Median	Q3	Max	BF_{10}	Effect size (median)	Lower 95%CrI	Upper 95%CrI	Evidence
Temperature symptomatic leg (ºC)	31.2	1.73	27.9	30.1	31.1	32.7	33.8	7.2	0.56	0.12	1.06	Moderate *
Temperature non-symptomatic leg (ºC)	30.8	1.84	27.7	29.9	30.9	31.9	33.9					
Area symptomatic leg (mm^2)	1.81	2.16	0.00	0.50	1.45	2.30	9.20	8.1	0.57	0.12	1.08	Moderate *
Area non-symptomatic leg (mm^2)	0.27	0.55	0.00	0.00	0.00	0.30	1.80					
Maximum systolic velocity symptomatic leg	17.7	9.82	0.0	20.4	22.2	23.6	25.5	16	0.66	0.17	1.18	Strong*
Maximum systolic velocity non-symptomatic leg	5.9	9.87	0.0	0.0	0.0	15.0	23.1					
Final diastolic velocity symptomatic leg	14.7	9.4	0.0	4.8	19.7	20.5	23.5	26	0.71	0.21	1.25	Strong*
Final diastolic velocity non-symptomatic leg	3.3	7.5	0.0	0.0	0.0	0.0	19.6					
Vascular resistance symptomatic leg	0.37	0.40	0.05	0.11	0.16	0.81	1.00	395	-1.04	-1.65	-0.45	Extrema*
Vascular resistance non-symptomatic leg	0.95	0.19	0.18	1.00	1.00	1.00	1.00					
Longitudinal echointensity symptomatic leg	21.3	7.78	5.9	17.6	22.0	27.9	32.1	0.48	-0.29	-0.72	-0.02	--
Longitudinal echointensity non-symptomatic leg	23.4	6.66	10.2	20.4	24.2	27.6	34.7					
Longitudinal echovariaton symptomatic leg	34.8	9.72	22.7	28.1	32.9	40.6	53.0	0.24	0.02	-0.41	0.04	--
Longitudinal echovariaton non-symptomatic leg	34.5	9.12	19.0	28.3	33.9	38.1	52.3					
Transversal echointensity symptomatic leg	35.6	5.83	25.0	31.7	36.0	40.2	45.0	0.25	0.05	-0.38	0.48	--
Transversal echointensity non-symptomatic leg	35.4	7.28	23.4	29.4	35.5	43.1	46.1					
Transversal echovariaton symptomatic leg	30.5	7.39	22.3	25.1	27.7	34.6	48.9	0.25	0.06	-0.37	0.49	--
Transversal echovariaton non-symptomatic leg	29.7	8.78	21.5	24.3	26.8	30.4	53.3					

SD: standard deviation. Min: minimum. Q1: 1st quartile. Q3: 3rd quartile. Max: maximum. BF_{10}: Bayes factor as the number of times in favor of the evidence of the alternative hypothesis over the null hypothesis. 95% CrI: 95% credible interval. *: a priori unilateral contrast. It can be considered that when the credibility intervals continue to have an effect of 0, there is no evidence of differences between limbs.

Table 5. Correlations between clinical variables and thermal asymmetry with vascular and ultrasonographic parameters.

Variables (for symptomatic tendons n=26)	*R*	Lower 95%CrI	Upper 95%CrI	R^2%	BF_{10}	Evidence
		VISA-P Kendall's tau-B				
Evolution Time	-.18	-.41	-.02	3%	1.0	Anecdotal*
Temperature difference	.29	.04	.51	8%	3.8	Moderate*
Doppler area	-.15	-.39	-.01	2%	0.7	--
Maximum systolic velocity	-.17	-.41	-.01	3%	0.9	--
Final diastolic velocity	.00	-.29	-.01	0%	0.3	--
Vascular resistance	-.13	-.38	-.01	2%	0.6	--
Longitudinal echointensity	.12	.01	.37	1%	0.6	--
Longitudinal echovariation	-.13	-.38	-.01	2%	0.6	--
Transversal echointensity	.02	.01	.30	0%	0.3	
Transversal echovariation	-.19	-.43	-.02	4%	1.1	Anecdotal*
		Evolution Time Kendall's tau-B				
Temperature difference	-.14	-.38	-.01	2%	0.7	--
Doppler area	.04	-.22	.29	0%	0.3	--
Maximum systolic velocity	.11	-.16	.35	1%	0.3	--
Final diastolic velocity	.18	-.09	.41	3%	0.6	--
Vascular resistance	-.02	-.27	.23	.1%	0.3	--
Longitudinal echointensity	-.07	-.32	.19	.5%	0.3	--
Longitudinal echovariation	-.08	-.33	.18	.7%	0.3	--
Transversal echointensity	.08	-.18	.32	.7%	0.3	--
Transversal echovariation	.09	-.17	.33	.8%	0.3	--
		*Difference of temperature Pearson's r***				
Differences Doppler area	.35	.04	.63	12%	1.9	Anecdotal*
Differences maximum systolic velocity	.63	.29	.80	39%	128	Extreme*
Differences final diastolic velocity	.59	.24	.78	35%	60	Very Strong*
Differences vascular resistance	-.55	-.75	-.18	30%	25	Strong*
Differences longitudinal echointensity	.14	.01	.50	2%	0.5	--
Differences longitudinal echovariation	.04	-.34	.40	0%	0.2	--
Differences transversal echointensity	.07	-.31	.42	0%	0.3	--

Differences transversal echovariation	.15	-.24	.48	2%	0.3	--

R: coefficient of correlation (Pearson's *r* or Kendall's *tau*-B). R^2%: % of explained variance, size effect. BF10: Bayes factor as the number of times in favor of the evidence of the alternative hypothesis over the null hypothesis. 95% CrI: 95% credible interval. *: a priori unilateral contrast. **Differences refer to symptomatic and non-symptomatic. It can be considered that when the credibility intervals continue to have an effect of 0, there is no evidence of differences between limbs.

Figure 1. Temperature measurement methodology by IRT. First thermogram (T1) with references points (a), location of the metallic markers in the second thermogram (T2) (b), location of the ROI (c) and superposition of the ROIs on the first thermogram (T1) (d).

Figure 2. Quantification of intratendinous vascular resistance using color pixel quantization from segmentation of systolic peak (a) and final diastolic peak (b) and applying resistance index formula.

Figure 3. ROI selection in the patellar tendon for echotexture analysis. a) Longitudinal section; b) cross section.

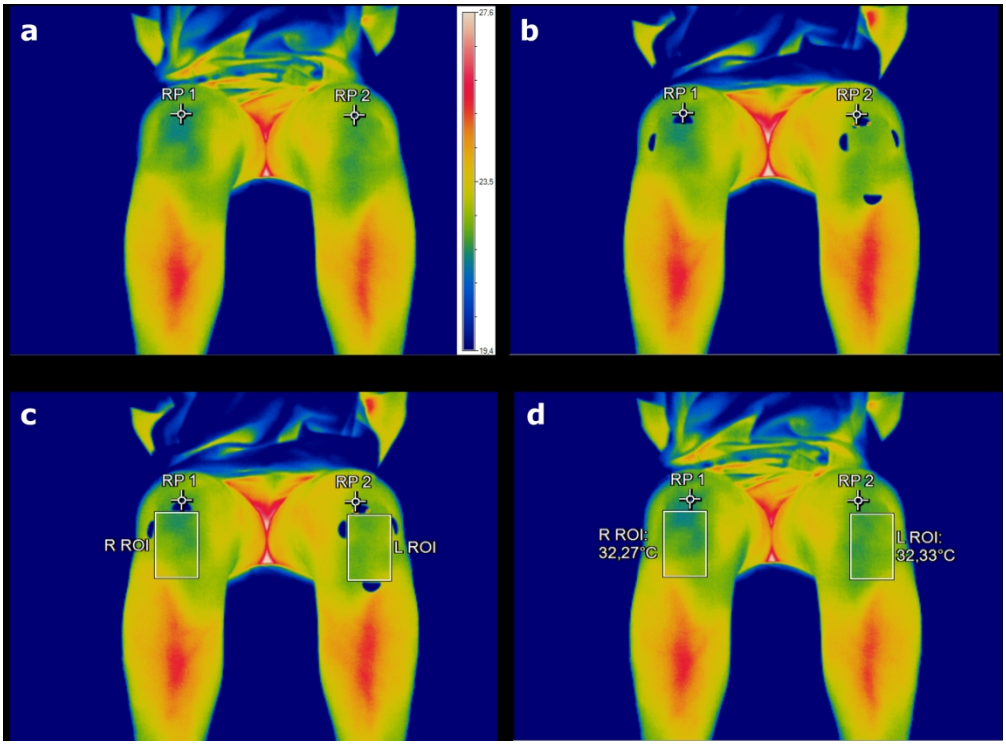

Figure 1. Temperature measurement methodology by IRT. First thermogram (T1) with references points (a), location of the metallic markers in the second thermogram (T2) (b), location of the ROI (c) and superposition of the ROIs on the first thermogram (T1) (d).

252x185mm (300 x 300 DPI)

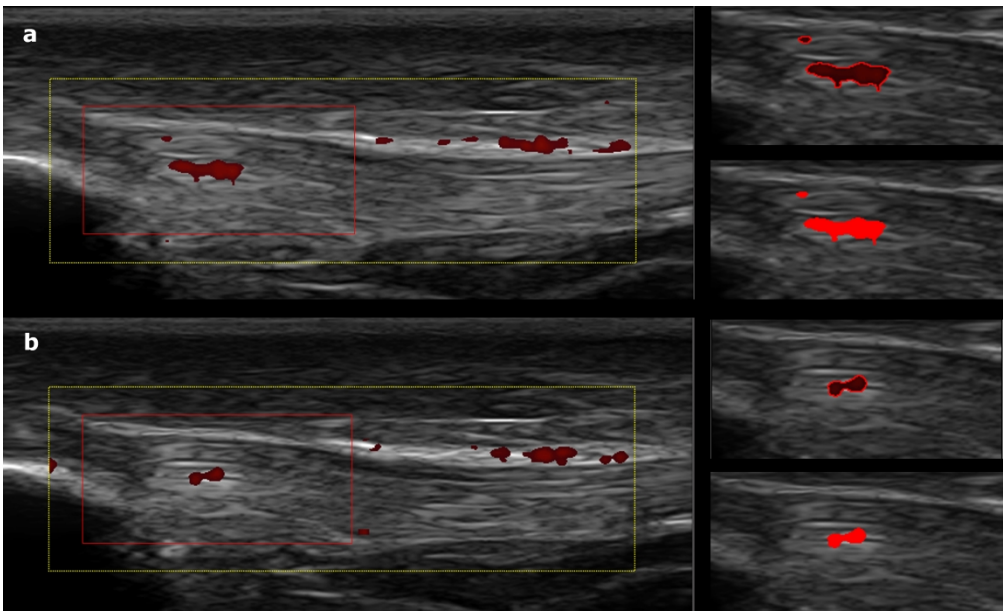

Figure 2. Quantification of intratendinous vascular resistance using color pixel quantization from segmentation of systolic peak (a) and final diastolic peak (b) and applying resistance index formula.

315x190mm (300 x 300 DPI)

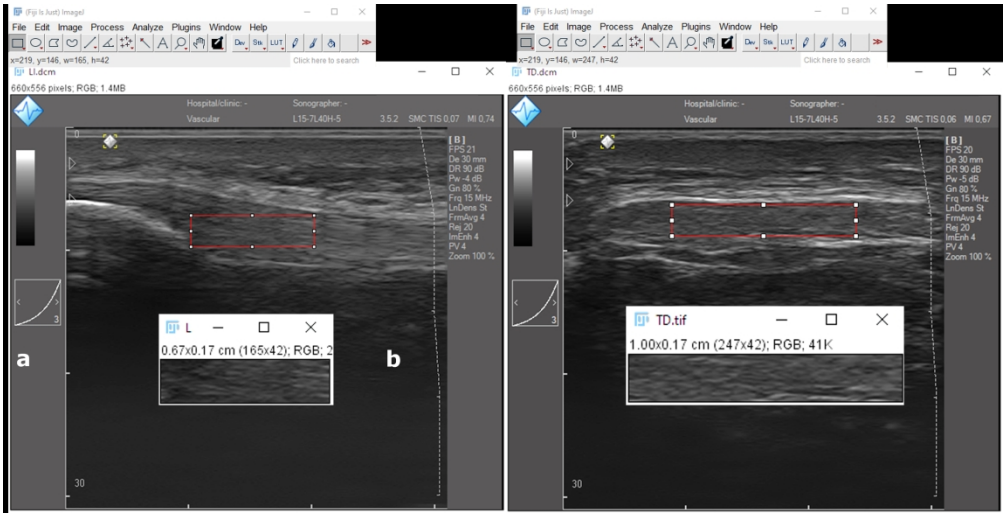

Figure 3. ROI selection in the patellar tendon for echotexture analysis. a) Longitudinal section; b) cross section.

322x163mm (300 x 300 DPI)

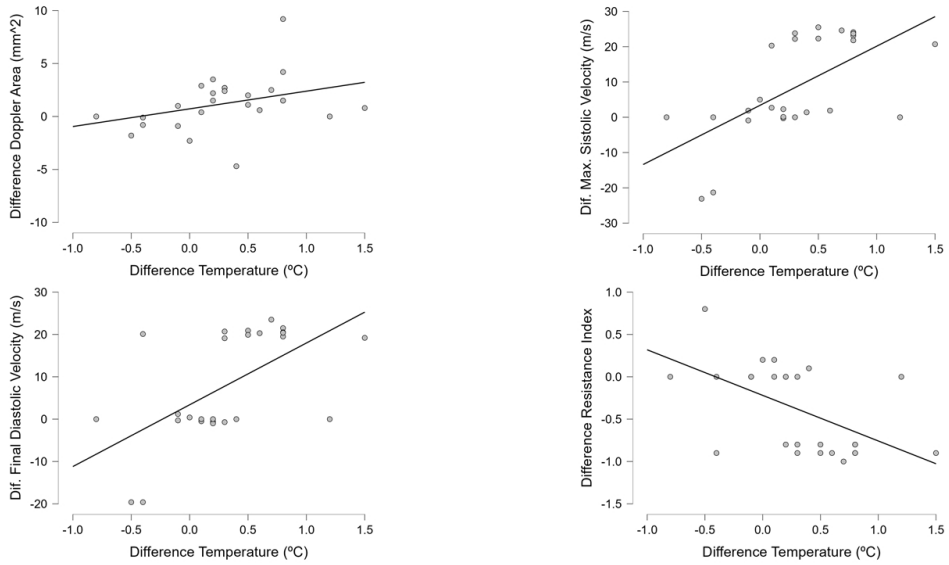

Figure 4. Correlations between differences in temperature and differences in vascular parameters.

338x190mm (96 x 96 DPI)