Effects of percutaneous needle electrolysis of the patellar tendon on local and contralateral temperature, measured with infrared thermography

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Abstract
Objectives: To assess the effect of the application of percutaneous needle electrolysis (PNE) technique, when applied to the patellar tendon, on the modification of cutaneous blood flow, of both the intervened knee and the contralateral knee over the course of 30 minutes post-application.

Material and Methods: An experimental nonrandomized study was performed, consisting of two groups of voluntary subjects: an electrolysis group (n = 19) and a control group (n = 14). Thermographic images were taken in the area of both knees at four different time intervals (pre-intervention, immediate post-intervention, fifteen minutes and thirty minutes post-intervention. The subjective feeling of perceived pain was also assessed using the visual analogue scale, as well as the attitude of fear-apperception.

Results: After applying the procedure to the patellar tendon, thermal changes were observed in the electrolysis group with a pattern of non-homogeneous behaviour. Changes were also observed in the contralateral knee with characteristics similar to the intervened knee.

Discussion: Thermal changes that occur in the electrolysis group, in both the intervention and contralateral knee, can only be explained by peripheral and central neurological mechanisms.

Conclusions: Percutaneous needle electrolysis in the patellar tendon produces thermal changes mediated by central mechanisms, both at the skin level in the area of the intervened knee, as well as on the contralateral side. The response patterns obtained, despite being heterogeneous, are conditioned by the pathological state of the tendon and the apprehension displayed during the intervention.

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Efectos de la electrolisis percutánea en el tendón rotuliano sobre la temperatura local y contralateral medida con termografía infrarroja

Resumen

Objetivos: Valorar el efecto que tiene la aplicación de la técnica de electrolisis percutánea musculoesquelética en el tendón rotuliano sobre la modificación del flujo sanguíneo cutáneo de la rodilla intervenida y su contralateral a lo largo de los 30 minutos posteriores a la aplicación. Material y métodos: Se realizó un estudio experimental no aleatorizado con dos grupos de sujetos voluntarios: grupo electrolisis (n = 19) y grupo control (n = 14). Se realizaron tomas de imágenes termográficas del área de ambas rodillas en cuatro tiempos diferentes (antes de la intervención, inmediatamente después de la intervención, a los 15 minutos y a los 30 minutos). También se valoró la sensación subjetiva de dolor percibido mediante la escala analógica visual y la actitud de miedo-aprehensión. Resultados: Tras la aplicación del procedimiento en el tendón rotuliano, se observaron cambios térmicos en el grupo electrolisis con un patrón de comportamiento no homogéneo, también se observaron cambios en la rodilla contralateral a la intervenida de características similares a la rodilla sometida a intervención. Discusión: Los cambios térmicos que se producen en el grupo electrolisis tanto en la rodilla intervenida como en la contralateral sólo pueden explicarse por mecanismos neurológicos periféricos y centrales. Conclusiones: La técnica de la electrolisis percutánea musculoesquelética en el tendón rotuliano produce cambios térmicos mediados por mecanismos centrales, tanto en el área cutánea de la rodilla de aplicación como en la contralateral. Los patrones de respuesta obtenidos, a pesar de ser heterogéneos, están condicionados por el estado patológico del tendón y la aprehensión mostrada durante la intervención.

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Introduction

Percutaneous needle electrolysis (PNE) is an invasive physiotherapy technique consisting of the ultrasound-guided application of a galvanic current, via a solid, filiform needle. This produces an analgesic effect in the neuromusculoskeletal soft tissue, together with a local inflammatory process that enables phagocytosis and the repair of the affected tissue. Several studies have demonstrated good clinical results for electrolysis, however, there are very few publications on the physiological effects of the technique. Consequently, it is necessary to further our knowledge regarding this aspect.

A method for testing the physiological effects of conservative and invasive interventions is taking the measurements of skin temperature. Skin temperature is a reflection of the amount of skin blood flow, which is regulated by the sympathetic nervous system and other local mechanisms. For this reason, the use of this measurement variable may provide information regarding the cutaneous vascular effects produced by PNE and, indirectly, regarding its effect on the nervous system.

Infrared thermography is a tool that enables us to measure and visualize skin temperature in real time. It has been validated by different authors for its musculoskeletal applications. Calin et al. performed a review between 2004 and 2014 on the effectiveness of thermography for the diagnosis and monitoring of knee pathologies (which include osteoarthritis, rheumatoid arthritis, ligament pathology, tendinous pathology and knee arthroplasty), reporting that infrared thermography has a specificity of 89% and a sensitivity of 90%. On the other hand, a study by Hildebrandt et al. obtained an intraclass correlation coefficient (ICC) that indicated a good interexaminer reliability in assessments of the anterior knee region over consecutive days.

Infrared thermography has already been employed by other authors with the objective of registering changes in skin temperature after the application of invasive physiotherapy techniques, such as acupuncture or dry needling of myofascial trigger points. However, no study has related the application of percutaneous electrolysis with variations of skin temperature. These thermal changes enable the assessment of the local effect and the distance of the technique, as well as analysing the pattern of thermal change with time, which can describe the physiological response associated with the intervention.

The main aim of this study was to evaluate the effect of the application of musculoskeletal PNE, applied to the patellar tendon, on the modification of skin blood flow of the intervened knee and the contralateral knee over the course of 30 minutes post application. Specifically, we sought to answer the following questions: a) Do changes of skin temperature occur in the intervened knee at any point during the first 30 minutes post intervention and which do not occur in subjects who do not receive MPNE?; b) What trajectories of thermal change occur in the intervened knees during...
these 30 minutes?; c) What is the percentage of subjects with change at the end of the period in the intervened knee?; d) Is this pattern of change homogenous in the intermediate shorter periods (e.g. between 15 and 30 minutes?; c) Does the amount and pattern of the change differ between the intervened knee and the contralateral knee during these 30 minutes?

Material and methods

Design and study subjects

An experimental, non-randomized study was performed with two groups (experimental and control). The experimental group (EG) received PNE over the patellar tendon of one knee. The control (CG) received no intervention. The measurement of results was performed in the same way for both groups and at the same times (before the intervention [PRE], post-intervention [T0], at 15 [T15] and 30 minutes [T30]) and in the same positions. The Research Ethics Committee (CEI) of the CEU San Pablo University approved the study, which fulfils all the principles established in the Helsinki declaration.

The study subjects were voluntary participants over the age of 18 years, recruited among patients from the physiotherapy clinic “Clínica Fisioterapia Océano” (Móstoles, Madrid, Spain) by physiotherapists working at the clinic. The exclusion criteria were: i) systemic pathologies, and ii) usage of substances or medications that could affect the normal physiological behaviour of the nervous system and therefore cause alterations in the skin blood flow. All subjects signed the corresponding informed consent in order to participate in the study.

Physiotherapy interventions

All subjects, from both the EG and the CG, had the application area sterilised and went through an acclimatisation period, lasting 15 minutes, with the subject standing with bare legs in a room at 22-24 °C. This complies with the criteria for the performance of thermographic studies, according to the guidelines determined by the American Academy of Thermology. Thereafter, the subjects from the EG received a non ultrasound-guided approach (in order to avoid artefacts on the thermal images) with the patient in supine position with 30° knee flexion maintained by a semirigid element at one centimetre from the apex of the patella and with a 45° inclination. The knee selected for the intervention was, preferably, the one with ultrasound signs of tendinous pathology or, in its defect, the one where the patient experienced the most symptoms. In the case of the absence of signs or symptoms in both knees, one knee was randomly selected. Sterile single-use needles were used (Physio Invasiva® needle 0.30 x 0.30 mm) with the Physio Invasiva® device (Grupo PRIM, Madrid, Spain) (fig. 1). Three impacts of 3 mA were applied during 3 seconds, after which the needle was removed.

Patients from the control group did not receive any intervention, except for adopting and remaining in the same positions and for equal time periods as those used by the experimental group before, during, and after the application of PNE.

Measurements performed

The main variable measured was the mean temperature, measured in centigrade, measured pre-intervention (baseline), post-intervention (T0), and after 15 and 30 minutes. This variable was measured in both knees of EG subjects (with and without intervention) and in one knee of CG subjects. The subjective pain intensity perceived by the subject was also measured after the intervention (via the visual analogue scale, from 0 to 10, with 0 representing absence of pain and 10 maximal pain), and the attitude of fear-apprehension was assessed qualitatively (yes or no).

A thermographic Flir E60 camera was employed to take images for measurements (resolution 320 x 240; lens FOL18; serial number 64509645099). After the acclimatisation pre-intervention (baseline) images were taken over the area of the anterior aspect of both knees with the subject in standing position. Immediately after, the intervention was performed (electrolysis or non-intervention) in supine. The time that the subject remained on the plinth was 2 minutes for all groups. Subsequently, three more images were taken in standing, at 0, 15 and 30 minutes post-intervention. During the whole procedure, utmost care was taken during the acclimatisation periods to avoid subjects altering their images by contact or unnecessary movements as the procedure lasts approximately 50 minutes.

All the image series (Pre-0-15-30 minutes) were assessed according to the Glamorgan protocol for medical images, using the Flir tools plus (Quick Report) software, and via collecting the mean temperatures of each area under study in each of the four assessment times (fig. 2). All measure-
ments and their interpretations were performed interchangeably by two physiotherapists with experience in infrared thermography.

Statistical analysis

Three patterns of change were established (i.e. temperature increase, decrease, or no change), both for the overall study period (pre-T30), as well as for the intermediate periods (pre-T0; T0-T15; T15-T30). The patterns were determined based on the magnitude of change from maximum to minimum temperature reached by the control subjects from the beginning until the end of each period. Thus, when one subject reached a temperature change between the beginning and the end of a period that was above the maximum value of the controls, this was classified as a pattern of increase in temperature; if it was lower than the minimum value of the controls, this was classified as a pattern of decrease; and if it did not exceed any of the reference values, this was classified as a pattern of no change.

The knees intervened on were also classified in trajectories based on temperature changes between the preintervention time (baseline) and after 30 minutes. Three models of trajectory were established: knees with an initial decrease at T0, those with an initial increase, and finally those that did not change at T0. Based on these models, subject’s knees were classified in 6 different trajectories, according to whether, during the last 30 minutes, they had a pattern of increase, decrease or no change when compared to the pre-intervention moment.

Descriptive statistics were used for characterising the subjects that received the interventions and the controls, and for determining their limits of normality, change patterns and trajectories. The Pearson’s chi-squared test was used to examine whether there were differences between the change patterns of the intervened knees and the contralateral knees. The Student’s t-test was used for paired samples (or the Wilcoxon test if the former were inappropriate) in order to compare the magnitudes of change during each analysed period.

Results

Study population

Thirty-three voluntary subjects participated in the study, 19 in the EG and 14 in the CG, aged between 20 and 42 years, of whom 4 were women and 29 were men. As shown in table 1, the EG and the CG were comparable regarding their basic characteristics, as well as the basal temperature and the presence of tendinopathy. Furthermore, the table also displays the pain intensity experimented by the EG in the intervened knee (mean 5.1; standard deviation 1.78), frequency of apprehension and values of minimum and maximum temperature change experienced by the control group during, both the overall period, and each of the subperiods. These values act as limits of normality based on which the presence of change was considered in the EG knees.

Trajectories and periods of temperature change in the knee with MPNE

All but one of the knees receiving the intervention experienced a change in temperature above the variation limits experienced by the participants in the control group. In figure 3, we can see the change trajectory for this knee (trajectory 1) between pre-intervention and immediate post-intervention (TO), and between this and T30. The re-

![Table 1](http://www.elsevier.es)

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Experimental group* (n = 19)</th>
<th>Control group* (n = 14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Age (years)</td>
<td>37.0 (4.39)</td>
<td>34.9 (6.36)</td>
</tr>
<tr>
<td>• Sex, n (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males, n = 19 (100%)</td>
<td></td>
<td>Males, n = 10 (71.4%)</td>
</tr>
<tr>
<td>Females, n = 0 (0%)</td>
<td></td>
<td>Females, n = 4 (28.6%)</td>
</tr>
<tr>
<td>• With knee tendinopathy, n (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intervened or control</td>
<td>n = 6 (31.6%)</td>
<td>n = 3 (21.4%)</td>
</tr>
<tr>
<td>Contralateral</td>
<td>n = 2 (10.5%)</td>
<td>-</td>
</tr>
<tr>
<td>• Basal knee temperature</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intervened or control</td>
<td>30.3 (1.42)</td>
<td>30.7 (1.28)</td>
</tr>
<tr>
<td>Contralateral</td>
<td>30.3 (1.47)</td>
<td>-</td>
</tr>
<tr>
<td>• Intensity of pain during application, n (%)</td>
<td>5.1 (1.78)</td>
<td>-</td>
</tr>
<tr>
<td>• With apprehension towards application, n (%)</td>
<td>n = 5 (26.3%)</td>
<td>-</td>
</tr>
<tr>
<td>• Amount of temperature change between periods, Range (minimum; maximum)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Between basal and T30</td>
<td>-</td>
<td>1.5 (-1.3; 0.2)</td>
</tr>
<tr>
<td>Between basal and T0</td>
<td>-</td>
<td>0.2 (0; 0.2)</td>
</tr>
<tr>
<td>Between T0 and T15</td>
<td>-</td>
<td>1.3 (-1.2; 0.1)</td>
</tr>
<tr>
<td>Between T15 and T30</td>
<td>-</td>
<td>0.8 (-0.6; 0.2)</td>
</tr>
</tbody>
</table>

*The values indicate the mean (standard deviation) unless another statistical test is indicated.
maining 18 knees experienced changes greater than normality, during some of these intermediate periods. Up to 5 additional trajectories were identified among the 18 knees:

- There were two trajectories in which an initial temperature decrease occurred at time 0: trajectory 2 \((n = 5)\) which, saw a recovery of the basal values at T30; trajectory 3 \((n = 5)\) in which an increase of the temperature followed which was higher than basal values and greater than the normality threshold.
- There were two trajectories in which an initial temperature increase was registered at time 0: trajectory 4 \((n = 4)\) in which, subsequently, a recovery of basal values occurred at T30; trajectory 5 \((n = 1)\) in which this temperature increase was maintained.
- In trajectory 6 \((n = 3)\) no change occurred at T0, as happens in trajectory 1, however, subsequently an increase was registered at T30.

The pain intensity did not vary between subjects presenting one or another pattern, however there was a significant relation between trajectory 2 (initial decrease followed by a recovery of the basal values) and the subjects that manifested apprehension towards the intervention \(n = 5\) (26.3%).

As a consequence of all these trajectories describing temperature variations, at the end of overall period (pre-30 period), 47.4% of subjects experimented a skin temperature increase in the intervened knee that was greater than the normality intervals experimented by the control group (fig. 4). This change pattern was different to that occurring...
Effects of percutaneous needle electrolysis of the patellar tendon on local and contralateral temperature during the intermediate periods. Thus, the intermediate periods did not share a homogenous pattern between them. It is worth highlighting that the only period in which a decrease pattern occurred was during the initial period (pre-0).

**Magnitude and change patterns in the contralateral knee compared to the intervened knee**

Table 2 features the amount of temperature increase, or decrease, occurring in the intervened knees and their contralateral sides for all subjects that had a decreasing or increasing pattern in the intervened knee during each period assessed (pre-30, pre-0, 0-15, 15-30). It can be observed that in both knees there were no statistically significant differences in the pre-0, 0-15, 15-30 intervals (P-value > 0.05), and that the behaviours during these periods were similar between the intervened and contralateral knees. However, in the overall pre-30 period, statistically significant differences exist (P-value = 0.034).

Table 3 compares the percentage of subjects who have suffered an increase, decrease or no change in the temperature of the intervened knee and the contralateral side, for subjects in the experimental group, during each of these periods (pre-30, pre-0, 0-15, 15-30). Statistically significant differences were not observed between the percentages of both groups (P-value > 0.05), and, therefore, we can affirm that the pattern of change for both knees is similar.

**Discussion**

To our knowledge, this is the first study to evaluate the thermal changes in the anterior knee region after the application of PNE in the patellar tendon. Our study shows that, at some point during the first 30 minutes after the application of PNE, changes occur in the skin temperature of the area that are greater than those recorded in the control subjects. Furthermore, the study showed that the initial change response and the ensuing change after 30 minutes is highly variable across subjects. Thus, we identified up to six different trajectories, which represented a non-homogeneous pattern of change between periods. Lastly, our study also provides evidence that the magnitude and the pattern

**Table 2** Magnitude of temperature changes registered in the intervened knees and the contralateral sides among subjects who have experimented changes in the intervened knee during each of the study periods

<table>
<thead>
<tr>
<th>Subjects and periods</th>
<th>Intervened knee*</th>
<th>Contralateral knee*</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subjects with an increasing pattern</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Between pre-intervention and T30</td>
<td>0.53 (0.14)</td>
<td>0.31 (0.25)</td>
<td>0.034</td>
</tr>
<tr>
<td>• Between pre-intervention and T0</td>
<td>0.36 (0.054)</td>
<td>0.26 (0.11)</td>
<td>0.059</td>
</tr>
<tr>
<td>• Between T0 and T15</td>
<td>0.41 (0.24)</td>
<td>0.24 (0.37)</td>
<td>0.065</td>
</tr>
<tr>
<td>• Between T15 and T30</td>
<td>0.4 (0.14)</td>
<td>0.24 (0.28)</td>
<td>0.461</td>
</tr>
<tr>
<td>Subjects with a decreasing pattern</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Between pre-intervention and T0</td>
<td>−0.28 (0.19)</td>
<td>−0.35 (0.25)</td>
<td>0.14</td>
</tr>
</tbody>
</table>

*The values indicate the mean (standard deviation) unless another statistical test is indicated.

**Table 3** Frequency with which change patterns were found for the skin temperatures of subjects from the experimental group, during the overall study period (pre-30) and during each of the intermediate periods

<table>
<thead>
<tr>
<th>Patterns of change by periods</th>
<th>Intervened knee</th>
<th>Contralateral knee</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between pre-intervention and T30</td>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>• No change</td>
<td>10 (52.6%)</td>
<td>10 (52.6%)</td>
<td></td>
</tr>
<tr>
<td>• Increase</td>
<td>9 (47.4%)</td>
<td>9 (47.4%)</td>
<td></td>
</tr>
<tr>
<td>Between pre-intervention and T0</td>
<td></td>
<td></td>
<td>0.754</td>
</tr>
<tr>
<td>• Decrease</td>
<td>10 (52.6%)</td>
<td>9 (47.4%)</td>
<td></td>
</tr>
<tr>
<td>• No change</td>
<td>4 (21.1%)</td>
<td>6 (31.6%)</td>
<td></td>
</tr>
<tr>
<td>• Increase</td>
<td>5 (26.3%)</td>
<td>4 (21.1%)</td>
<td></td>
</tr>
<tr>
<td>Between T0 and T15</td>
<td></td>
<td></td>
<td>0.746</td>
</tr>
<tr>
<td>• No change</td>
<td>9 (47.4%)</td>
<td>11 (57.9%)</td>
<td></td>
</tr>
<tr>
<td>• Increase</td>
<td>10 (52.6%)</td>
<td>8 (42.1%)</td>
<td></td>
</tr>
<tr>
<td>Between T15 and T30</td>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>• No change</td>
<td>14 (73.7%)</td>
<td>13 (68.4%)</td>
<td></td>
</tr>
<tr>
<td>• Increase</td>
<td>5 (26.3%)</td>
<td>6 (31.6%)</td>
<td></td>
</tr>
</tbody>
</table>
of change experienced in the contralateral knee are similar to that displayed by the intervened knee.

This study has methodological strengths that we wish to highlight. In the first place, the maximum and minimum values reached by control subjects of similar characteristics and from the same geographical context were used as a change threshold. In second place, and due to the fact that skin temperature is related with the amount of skin blood flow, and that this is regulated by the autonomic sympathetic nervous system and the hypothalamus, there are different factors that can vary the skin blood flow, such as the thermoregulation processes for increasing internal thermal homeostasis and other non-thermoregulatory processes related with the variation in blood pressure and/or exercise. Thus, in this study we attempted to control these factors, both in the CG as well as in the EG, by performing a process of acclimatisation in a room with controlled environmental temperature which minimized the effect of the same. The only difference between both groups was that the EG received the intervention with musculoskeletal PNE.

In the intervened knee, six behaviour trajectories have been established. It is worth noting that all subjects in trajectory 2, in which initial vasoconstriction occurs at time 0 and basal recovery in time 30, displayed apprehension during the intervention. This may be explained by an activation of the sympathetic autonomic nervous system (fight or flight), which maintains this skin vasoconstriction throughout the thirty minutes that the measurements last, without knowing whether, afterwards, a thermal increase occurred associated with skin vasodilation. It is also worth highlighting that all subjects in trajectory 3, in which a decrease in temperature occurred in time 0 followed by an increase, had pathological structural findings in the intervened tendon. Of the six subjects with a pathological tendon to whom PNE was applied, only one subject, who displayed apprehension towards the intervention did not have the trajectory 3, but rather followed trajectory 2. The trajectories 4 (n = 4), 5 (n = 1) and 6 (n = 3) correspond to subjects who, at some point within the measurement periods, had a vasodilator effect, and did not display an increase in sympathetic tone (vasoconstriction) at any time during the measurement process. These subjects did not display any type of apprehension towards the technique, unlike those belonging to trajectory 2, and all of them had a healthy tendon, unlike the subjects following to trajectory 3.

As previously commented, the pattern of temperature change in the knee of application was not homogenous over the different periods, this may be due to the competiveness between the local effects and the central effects caused by the intervention. The pattern of change in the contralateral knee was similar to that of the intervened knee. In general, we observed that certain subjects have a bilateral response of skin temperature decrease, which suggests a sympathetic autonomic activation at a central level. On the other hand, in some subjects, a temperature increase occurs in both knees. In the assessed knee, this vasodilation may have its origin in the local response to the tissue damage produced by the needle as well as the inflammatory effect of percutaneous needle electrolysis in the tissue, as demonstrated in previous studies. The inflammatory cascade produced by the intervention produces a liberation of vasoactive substances that may be the origin of skin vasodilation.

The presence of similar response patterns among knees can only be explained as a consensual autonomic sympathetic response at the central level, which has already been demonstrated by authors, such as Ying et al., in a study in which Doppler laser was used to register the contralateral vascular effect in response to mechanical stimuli. Also, Marshall et al. demonstrated the central vasodilator effect in their study in the year 1991 where a thermal stimulus in a hand achieved vascular effects on the contralateral side. Lastly, Guangjun et al., in a study using acupuncture, produced contralateral vascular skin effects on the contralateral side, which the authors explained as being some type of activation of autonomic mechanisms or even being related to the somatosensory cortex.

Concerning the VAS, a striking aspect is that no relation was observed between the assessment of pain expressed by the subject and the type of response, although, as commented previously, there was a relation between apprehension and the pattern of vasoconstriction maintained over time after the application. It is also important to highlight that a correlation did not exist, either, between the VAS and the degree of apprehension, although this sometimes does not reflect the reality that the patient experiments or that the physiotherapist observes during the application of invasive techniques, as in this case.

**Study limitations**

An important limitation of the present work is that the samples included in the EG and the CG were heterogeneous regarding the presence of tendon pathology. This may have biased the response and, therefore, in the future it is necessary to study a group of subjects who all have tendon pathology. Another limitation is that it is possible that a bias may have existed during the measurements due to the fact that neither of the physiotherapists participating in the study were blinded and because both indistinctly performed both measurements and interventions, which may influence the results obtained. Lastly, the fear-apprehension attitude was measured qualitatively with an instrument that is not sensitive to detecting different levels of apprehension. Future research should seek to include the PPAS (Personal Psychological Apprehension Scale).

**Conclusion**

The PNE technique applied to the patellar tendon produces thermal changes mediated by central mechanisms, both upon the skin area of the knee receiving treatment, as well as on the contralateral knee. The response patterns obtained, despite their heterogeneity, may be conditioned by the pathological state of the tendon and the apprehension displayed during the intervention. Future studies should research this aspect and validate the preliminary results obtained in this study.

**Acknowledgements**

We wish to thank all those who have collaborated towards making this study possible.
Conflicts of interest
The authors declare that they have no conflicts of interest.

References