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# Comparison of dry needling and ischaemic compression techniques on pain and function in patients with patellofemoral pain syndrome: a randomised clinical trial

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## Abstract

**Background:** To compare the effectiveness of ischaemic compression (IC) applied directly to the knee versus dry needling (DN) with respect to pain, functional status and sensitivity to mechanical stimulation of vastus medialis obliquus (VMO) myofascial trigger points (MTrPs) in patients with patellofemoral pain syndrome (PFPS).

**Methods:** A total of 54 patients with unilateral PFPS aged 20–30 years were selected randomly from patients referred to physical therapy clinics of Babol University of Medical Sciences in Iran. Twenty-seven patients were allocated to either IC or DN groups. Three sessions of treatment were applied over 1 week with follow-up at 1 week, 1 month and 3 months. Primary outcome measures comprised the Kujala questionnaire score for functional status, numerical pain rating scale (NPRS) for pain intensity and pressure pain threshold (PPT) for sensitivity to mechanical stimulation; these were measured and recorded before treatment and 1 week, 1 month and 3 months after the last treatment session.

**Results:** There were no statistically significant differences in the between-group comparisons of any variables at the various follow-up points. Both groups (n = 27 participants each) had significant improvements ($p < 0.05$) in pain, functional status and PPT values at follow-up.

**Conclusions:** There were no differences in markers of pain, function or pressure sensitivity over a 3-month follow-up period between patients with PFPS treated with DN and IC. Temporal improvements in both groups suggested that the two techniques may be similarly effective for the treatment of PFPS.

## Keywords

dry needling, ischaemic compression, myofascial trigger point, patellofemoral pain syndrome, rehabilitation

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## Introduction

Patellofemoral pain syndrome (PFPS) is claimed to be one of the most significant clinical knee problems in young active individuals[1] and may affect up to 40% of the population.[1,2] The disorder presents with pain in the anterior or medial aspect of the knee,[1–3] aggravated by patellofemoral compressive forces and active overloading activities, such as prolonged sitting, squatting and stair climbing,[3,4] which can limit several aspects of daily life.[5,6] Although the aetiology of PFPS is not yet clear,[2] there is consensus that multiple factors, including repeated trauma, overuse and extensor mechanism dysfunction, are the most probable causes.[7–9] The vastus medialis obliquus (VMO) is

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regarded as an important medial dynamic stabiliser of the patellofemoral joint. An insufficient VMO may be unable to counterbalance the lateral pull of the vastus lateralis (VL) in an attempt to establish patellar stability.[10] Also, delayed onset of VMO activity[5,11,12] predisposes individuals to patellar maltracking and increased patellofemoral joint contact pressure. Hence, the person becomes prone to PFPS.

Myofascial trigger points (MTrPs), localised hyperirritable areas of taut band-like hardness in muscles, are known to be a common source of pain in patients referred to rehabilitation clinics.[12–14] Based on the literature, MTrPs may develop anywhere in the body, following repetitive unbalanced eccentric and concentric loading activities[12–16] or repetitive overuse or overload.[9,17,18] There is also evidence that MTrPs in the quadriceps femoris muscle group, particularly in the VMO,[9,19,20] can provoke a combination of PFPS signs and symptoms. Considering VMO insufficiency to be an important predisposing factor in the development of PFPS, recent studies have evaluated the prevalence of trigger points in this muscle group. Simons showed that extensor dysfunction in the quadriceps femoris muscle group might be related to the development of MTrPs.[21] Dippenaar[18] evaluated the prevalence of MTrPs in the quadriceps muscle of patients with PFPS and found that the most common locations of active MTrPs were in the mid-belly of the VL, distal muscular portion of the VL and distal muscular portion of the vastus medialis (VM). The repetitive cycle of pain dysfunction due to overload may be perpetuated, leading to VMO insufficiency and weakness. This theory could explain the development of MTrPs in the VMO of the quadriceps muscle group.[3,8,21]

Management of PFPS commonly comprises non-operative interventions. Considering the multifactorial aetiology of PFPS, different treatment strategies, including exercises for the hip and knee joints,[2,22,23] neuromuscular electrical stimulation,[24] soft tissue manipulation and acupuncture,[8] have been proposed. So far, conventional treatments for PFPS have aimed to improve the balance and strength of the quadriceps muscle components[5,24,25] and may not fully address muscle inhibition successfully.[9,25] Therefore, treatments that directly focus on reducing the patient's pain, improving motor function and decreasing pain sensitivity, are desirable. The use of techniques, such as dry needling (DN)[26,27] and ischaemic compression (IC),[8,26] is claimed to be effective in the treatment of patients with MTrPs. The IC technique works by increasing local blood flow, facilitating tissue recovery and normalising impaired motor activation patterns in the muscle.[8,27] In patients with PFPS, applying IC to MTrPs in the VMO improved VMO function, which is one of the main factors contributing to appropriate VMO/VL coordination. Therefore, IC can improve patellar tracking.[28–30]

DN is a common technique in which an acupuncture-like needle is inserted into the muscle at the location of a MTrP. It has been suggested that DN causes stimulation of A-delta nerve fibres, thus activating the enkephalinergic inhibitory dorsal horn interneurons and causing opioid-mediated pain suppression.[12–14] Adding DN of the VL and VM to the intervention programme of patients with PFPS improved the outcomes of disability and pain at 3-month follow-up.[30] The efficacy of VM DN in conjunction with a rehabilitation programme has been confirmed in patients with anterior cruciate ligament (ACL) reconstruction; it was shown that DN improved the range of motion and functionality in these patients.[13,31]

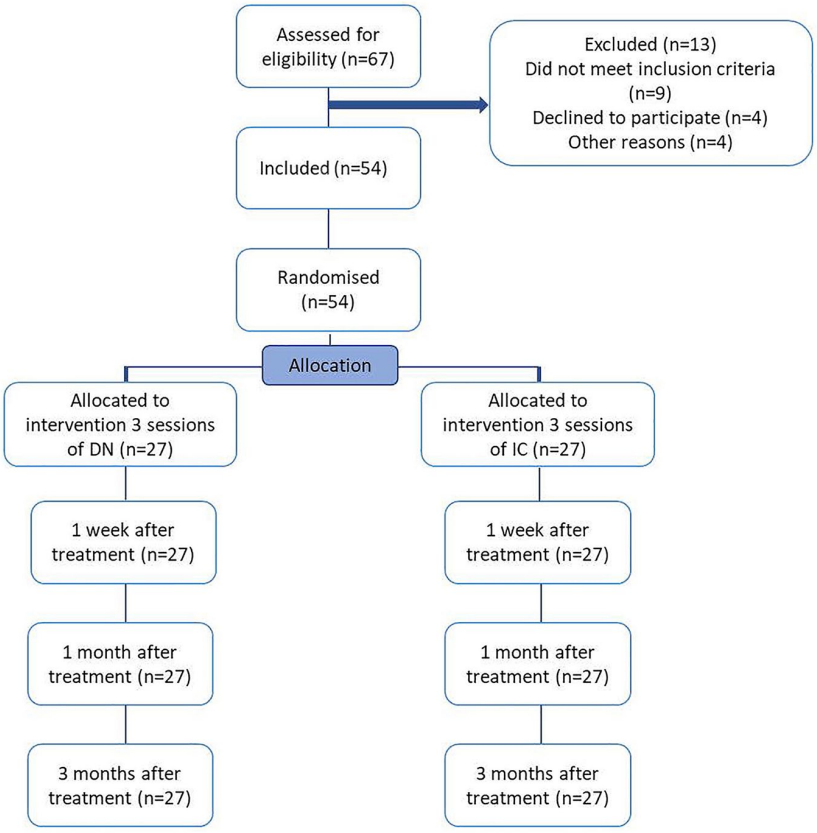
There is evidence for both IC and DN in the treatment of neck pain patients with MTrPs in the upper trapezius muscle.[32] However, comparison of these methods has yet to be performed in patients with PFPS. To the best of our knowledge, this study is the first randomised clinical trial designed to compare the short- and long-term effects of IC and DN on symptoms in patients with PFPS.

## Methods

### Design

Fifty-four young adults aged between 20 and 30 years with a diagnosis of unilateral PFPS were recruited in this single-blind randomised clinical trial. The participants were recruited from patients referred to the physical therapy clinics of Babol University of Medical Sciences in Babol, Iran, between March 2017 and June 2017, using simple random sampling. The sample size was calculated with Minitab software version 17 on the basis of data acquired from a pilot study with 10 patients (5 patients per group) and based on functionality, as one of the primary outcome measures, at 1-week, 1-month and 3-month follow-up ($Z_{1-\alpha/2}=1.96$, $Z_{1-\beta}=0.85$, $S^2_{IC}=41.94, S^2_{DN}=37.44$ and $d=5$). Assuming a dropout rate of 30% with the assumption of a two-tailed $\alpha$ level of 0.017 ($p<0.05$ corrected for multiple testing with three primary outcome measures) and 80% power, it was estimated that 27 patients would need to be assigned to each group. During an initial interview, an independent research assistant (who was neither involved in the interventions nor involved in data collection) performed screening for inclusion criteria. Fifty-eight patients met the inclusion criteria and four patients declined to participate. All participants read and signed an informed consent form. Another examiner recorded baseline values of weight, height, pain intensity, pressure pain threshold (PPT) and Kujala questionnaire score. An independent assistant managed group delivery with a block randomisation method based on a randomisation table provided by computer-generated random number sequence. Group allocation was carried out using sealed envelopes containing slips of paper labelled with either 'I' or 'D' letters representing assignment to the IC or DN groups, respectively. Two expert physical therapists monitored treatment sessions during the trial; the first therapist considered the outcome measures

**Figure 1.** Flowchart of procedures. DN: dry needling; IC: ischaemic compression.




and the second one performed the interventions. The second therapist was not aware of the recorded outcome measures. The primary outcome measures were anterior knee pain (Kujala) questionnaire, numerical pain rating scale (NPRS) and PPT. The protocol of this study was approved by the Vice-Chancellery for Research of Babol University of Medical Sciences, and the trial was prospectively registered in the Iranian Registry of Clinical Trials on 8 March 2017 (reference no. IRCT2016062028542N1). A flowchart of the study procedures is shown in Figure 1.

## Participants

Participants were eligible if they had: (1) pain during eccentric step test, patellar apprehension test or VM coordination test; (2) pain for more than 6 weeks during at least two of the following activities of walking, squatting, prolonged sitting, stair climbing, isometric quadriceps contraction and patellar compression;[5,8] (3) the presence of at least one active MTrP in the VMO of the symptomatic knee; and (4) an NPRS score $>4$ and Kujala questionnaire score between 40 and 70. Patients with: a history of ligamentous insufficiency of the knee, meniscal damage, patellar subluxation or dislocation; spinal or lower extremity surgery; any systemic, orthopaedic or neurological disorder; or recent physiotherapy programme; were excluded.[8] Demographic characteristics of the participants are shown in Table 1.

## Outcome measures

All three outcome measures (Kujala questionaire, NPRS and PPT) were recorded at baseline and 1 week, 1 month and 3 months after treatment.

## Anterior knee pain (Kujala) questionnaire

The Kujala questionnaire is a self-administered scale designed to evaluate functionality in patients with PFPS. The reliability and validity of the Persian version of the questionnaire have been previously confirmed.[33,34] The score ranges from a minimum of 0 to a maximum of 100, with higher scores indicating less pain and disability.

## NPRS

The NPRS is known to be a sensitive and reliable scale for clinical and experimental evaluation of pain intensity.[34] The score ranges from 0, indicating no pain, to 10, expressing

**Table 1.** Mean scores for numerical pain rating scale (NPRS), Kujala questionnaire and pressure pain threshold (PPT) comparing between dry needling (DN) and ischaemic compression (IC) groups.

| Group | Baseline | | | 1-week follow-up | | | 1-month follow-up | | | 3-month follow-up | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | IC | DN | p value | IC | DN | p value | IC | DN | p value | IC | DN | p value |
| Kujala | 62.43 ± 4.62 | 62.32 ± 4.83 | 0.93 | 81.82 ± 4.74 | 81.71 ± 7.00 | 0.95 | 81.46 ± 4.87 | 81.21 ± 7.03 | 0.88 | 78.21 ± 6.02 | 77.82 ± 7.04 | 0.82 |
| NPRS | 6.71 ± 0.66 | 6.69 ± 0.73 | 0.92 | 1.55 ± 0.97 | 1.68 ± 1.02 | 0.64 | 1.60 ± 0.95 | 1.68 ± 1.12 | 0.80 | 1.96 ± 1.13 | 1.89 ± 1.16 | 0.82 |
| PPT | 3.28 ± 0.16 | 3.25 ± 0.16 | 0.51 | 4.05 ± 0.18 | 4.02 ± 0.25 | 0.64 | 3.97 ± 0.21 | 3.95 ± 0.25 | 0.73 | 3.84 ± 0.17 | 3.80 ± 0.2 | 0.52 |

worst imaginable pain. Participants were asked to score their perceived pain on the NPRS before treatment and at the predetermined follow-up periods.[34]

### PPT

PPT has been defined as the lowest stimulus provoking the earliest perception of pain.[8] The reliability, reproducibility and validity of PPT have been demonstrated in several studies.[8] Two examiners collected the data. The first examiner, who performed the treatment, was blinded to the PPT values recorded by the other examiner. The first examiner, using a pressure algometer consisting of a circular 0.86-cm pliance capacitance pressure sensor (pliance®, novel; Munich, Germany) worn on the palpating thumb, applied firm and gradually increasing pressure over the marked MTrP. The examiner asked the patient to say 'now' as soon as they felt discomfort or pain. Then, the test was stopped and the other examiner recorded the value from the algometer monitor.[35] The PPT value recording was repeated three times and averaged.

### Trigger point recognition

Trigger points were identified by an expert examiner according to the following criteria: (1) presence of a palpable taut band, (2) focal tender spot within the taut band and (3) referred pain recognised by the patient as a 'familiar' pattern.[36] Inter-rater and intra-rater reliability of locating MTrPs has been reported as moderate to high.[37,38] The recognised MTrP had to elicit pain directly over the affected area and to produce a local twitch response on palpation.

### Interventions

Participants were asked to lie in a supine position with extended knees. The skin was cleaned with alcohol. The identified MTrP(s) was immobilised between thumb and index fingers. Then, the needle was inserted perpendicularly through the skin over the MTrP region, using the fast-in and fast-out technique described by Llamas-Ramos et al.[14] and moved forward until a local twitch response was elicited. Once the first local twitch response was obtained, the needle was moved up and down (using 2- to 3-mm vertical motions with no rotations) at a rate of approximately 1 Hz for 25–30 s.[14]

A stainless-steel single-use sterile acupuncture needle (Dongbang; Needle Pro Inc., Brisbane, Australia) of 0.25 mm diameter and 50 mm length, with insertion tube, was used to provide a noxious stimulus. In order to minimise the pain of insertion and thus improve the patients' tolerance to the needle, a certain pressure was applied to the skin using the insertion tube, and then each needle was inserted swiftly through the skin overlying the trigger points.

### IC

IC is a manual technique defined as the application of a gradually increasing pressure against the MTrP to induce the most tolerable pain.[39] The patients were asked to lay relaxed in a supine position with extended knees. The first examiner applied slow and steady increasing pressure to the marked MTrP until the pain perception was reported as seven on the NPRS, and the second examiner controlled the PPT value. This pressure was maintained for 90 s. If the patient reported a reduction in pain, the pressure was gradually increased to restore perceived pain to the previous value. Compression was applied three times with a 30-s rest interval in each session.[8,39] The exact point of the MTrP was marked to apply IC and post-treatment PPT measurement accurately.

### Statistical analysis

All descriptive and inferential statistical analyses were carried out using the Statistical Package for the Social Sciences (SPSS) version 21 (SPSS Inc., Chicago, IL, USA). The Kolmogorov–Smirnov test was used to verify normal distribution of the variables in each group. The independent samples' t-test was used to compare demographic data between the treatment groups. A mixed analysis of variance (ANOVA) with repeated measures, with 'time' (baseline, 1-week follow-up, 1-month follow-up and 3-month follow-up) as the 'within-subjects' factor and 'group' (DN, IC) as the 'between-subjects' factor, was used to determine the effects of the intervention on pain, PPT value and Kujala scores. Bonferroni correction was used for pairwise comparison of variables. The effect size of the interventions was evaluated by partial eta squared, values of which were categorised as small (0.01), medium (0.09) or large (0.25). The minimal detectable change (MDC) was calculated based on standard error of measurement (SEM) according to the following formulae

$$\text{MDC} = 1.96 \times \sqrt{2} \times \text{SEM}$$
$$\text{SEM} = \text{SD}\left(\text{standard deviation}\right) \times \sqrt{1-\text{r}}$$

The value of 1.96 is the z-score associated with 95% confidence interval (CI). The $\sqrt{2}$ is used for underlying extra uncertainty during measurement at two points of time. All data are presented as mean ± standard deviation (SD). The significance level for all statistical tests was set at $p<0.017$ ($p=0.05$ divided by 3 for multiple primary outcomes). The clinical significance of the effect size was determined with 95% CIs.

## Results

Fifty-four patients satisfied the eligibility criteria, agreed to participate and were randomised into either the DN group (n=27) or the IC group (n=27). No statistically significant differences were observed in demographic data between the two groups; age (26.4 ± 2.9 vs 26.3 ± 2.7), height (1.72 ± 8.0 vs 1.73 ± 8.0), weight (64.6 ± 15.1 vs 64.6 ± 14.9) and body mass index (BMI; 21.5 ± 3.5 vs 21.4 ± 3.5) were equivalent in DN versus IC groups (all $p>0.05$). The sex (female/male) ratio in both groups was 16/12. Regarding pain variables, the main effect of time was significant ($F(1,54)=1467.69$ and $p<0.001$), but the interaction between group and time was not significant ($F(1,54)=0.06$ and $p=0.88$). The main effect of group was not statistically significant when comparing pain between the groups ($F(1,54)=0.05$ and $p=0.83$). The effect size of interaction between group and time was small ($\eta_p^2 = 0.001$). The effect size was large for both DN ($\eta_p^2 = 0.97$) and IC ($\eta_p^2 = 0.96$). The main effect of time was also significant for function ($F(1,54)=347.91$ and $p<0.001$) and PPT ($F(1,54)=62.68$ and $p<0.001$), respectively. The main effect of group was not statistically significant for either function ($F(1,54)=0.02$ and $p=0.87$) or PPT ($F(1,54)=0.59$ and $p=0.45$). The interaction between group and time was not significant for function ($F(1,54)=0.02$, $p=0.93$ and $\eta_p^2 = 0.005$) or PPT ($F(1,54)=0.74$, $p=0.42$ and $\eta_p^2 = 0.01$). The effect size for function was large for DN ($\eta_p^2 = 0.88$) and IC ($\eta_p^2 = 0.85$). Regarding PPT, the effect size was large for DN ($\eta_p^2 = 0.87$) and IC ($\eta_p^2 = 0.37$). The pairwise comparison of variables at different time intervals in both groups is summarised in Table 2. Both groups showed similar patterns of change in function and pain (Table 1; Figure 2(a) and (b), respectively). Although the pattern of PPT differed between groups in such a way that PPT decreased from 1-week follow-up (4.05) to 1-month follow-up (3.97) in the DN group and increased from 1-week follow-up (3.88) to 1-month follow-up (3.95) in the IC group, the interaction of time and group was not significant. The pattern of change for PPT is depicted in Figure 2(c) and Table 1. The obtained results regarding the SEM and MDC of the interventions at different time points are summarised in Table 3.

## Discussion

The objective of this study was to compare the effects of the DN and IC techniques on pain, function and PPT at 1-week, 1-month and 3-month follow-up time points in a group of patients with PFPS. No significant difference was detected between the interventions; therefore, we cannot say which of the interventions (if any) was better than the other. However, all the evaluated variables improved during the follow-up period in both groups.

Our findings are consistent with those from other studies providing direct and indirect manual treatments for PFPS.[6,26,28,40] Clinical improvement was considered as a 10-point score reduction on the Kujala questionnaire and 2-cm reduction on the NPRS. However, comparison with the earlier studies was difficult because few studies have examined the presence of MTrPs in the quadriceps and also investigated MTrP sensitivity.

**Table 2.** Pairwise comparisons of numerical pain rating scale (NPRS), function (Kujala questionnaire) and pressure pain threshold (PPT) variables for time intervals in dry needling (DN) and ischaemic compression (IC) groups.

| Group | Variable | Baseline Mean ± SD (95% CI) | 1-week follow-up Mean ± SD (95% CI) | 1-month follow-up Mean ± SD (95% CI) | 3-month follow-up Mean ± SD (95% CI) |
|---|---|---|---|---|---|
| DN | Kujala | 62.43 ± 4.62 (60.64–64.22) | 81.82 ± 4.74[a] (79.98–83.66) | 81.46 ± 4.87[a] (79.57–83.35) | 78.21 ± 6.02[a, b, c] (75.87–80.55) |
| | NPRS | 6.41 ± 0.76 (6.42–7.01) | 1.68 ± 1.02[a] (1.28–2.07) | 1.68 ± 1.12[a] (1.24–2.11) | 1.89 ± 1.16[a, b, c] (1.44–2.34) |
| | PPT | 3.28 ± 0.16 (3.22–3.34) | 4.05 ± 0.18[a] (3.98–4.12) | 3.97 ± 0.21[a,d] (3.89–4.05) | 3.84 ± 0.17[a, b, c] (3.77–3.90) |
| IC | Kujala | 62.32 ± 4.83 (60.45–64.19) | 81.71 ± 7.00[a] (78.99–84.43) | 81.21 ± 7.03[a] (78.49–83.94) | 77.82 ± 7.04[a, b, c] (75.09–80.55) |
| | NPRS | 6.68 ± 0.61 (6.44–6.92) | 1.57 ± 0.96[a] (1.19–1.94) | 1.57 ± 0.92[a] (1.21–1.93) | 1.95 ± 1.15[a, b, c] (1.50–2.40) |
| | PPT | 3.25 ± 0.16 (3.18–3.31) | 3.88 ± 0.80[a] (3.57–4.19) | 3.95 ± 0.26[a] (3.85–4.05) | 3.81 ± 0.20[a,c] (3.73–3.88) |

CI: confidence interval; SD: standard deviation.
[a]Significant difference compared with baseline.
[b]Significant difference between 1-week and 1-month follow-up.
[c]Significant difference between 1-week and 1-month follow-up.
[d]Significant difference between 1-week and 1-month follow-up.

On examination, all the participants had at least one MTrP in the VMO of the symptomatic knee, which produced similar pain patterns. Many studies have reported short- or long-term improvements in pain and function after manual treatment.[6,25,27,40] The purported aim of these treatments, despite the difference in techniques, is to increase motor neuron pool activity, to normalise VMO contraction timing and to realign the patellar tracking system.[6,25,27,39]

IC applied directly to the peripatellar region has been recommended as a successful intervention that can lead to reduction of a patient's symptoms.[8,39] Hains et al. evaluated the efficacy of myofascial manual therapy (IC) directly on knee pain in patients with PFPS; the study showed that pain reduction was significantly greater in the treatment group compared to the control group at 1-month and 6-month follow-up.[39] Also, a study comparing the effectiveness of lumbopelvic manipulation and IC showed significant changes after treatment in both groups, with greater significant long-term effects on pain, function and PPT in the IC group.[8]

Based on the prevailing research, DN is an effective treatment for deactivating MTrPs in patients with PFPS. De-la-Llave-Rincon et al.[27] compared the effects of deep DN versus superficial DN of MTrPs combined with manual therapy on pain, disability and pressure pain sensitivity in patients with PFPS and showed significant improvements in knee pain intensity and PPT over the VM in comparison to the control group and the other portions of the quadriceps muscle. Another study investigated the efficacy of DN at MTrPs of the VL on the total work in long-distance runners with PFPS and showed no significant difference between DN and sham DN groups, although both interventions appeared beneficial.[40]

In this study, both groups experienced pain reductions, functional improvements and reduced pressure pain sensitivity at 1-week, 1-month and 3-month follow-up. No significant differences were observed in the variables at the different follow-up time points. Regarding the calculated minimum detectable concentrations (MDCs), both groups showed clinical improvements in the evaluated variables over the different follow-up periods.

## Study limitations

Our study had some limitations. Since the effect of some biomechanical and structural factors, such as Q angle and the degree of internal rotation at the hip, were not measured, the validity of the findings of this trial should be viewed with appropriate caution. We compared the isolated effects of these two interventions; hence, we are not sure if the same results would be obtained when these techniques are used as an adjunct to other routine interventions. Accordingly, future studies with sham treatment or exercise protocols are warranted to verify our findings. Also, future studies should examine VMO to VL activity ratio, which may provide additional information that could be used to develop a more valid

**Figure 2.** (a) Kujala questionnaire scores, (b) numerical pain rating scale (NPRS) scores and (c) pressure pain threshold (PPT) scores at 1-week, 1-month and 3-month follow-up time points in patients undergoing dry needling (DN) or ischaemic compression (IC).

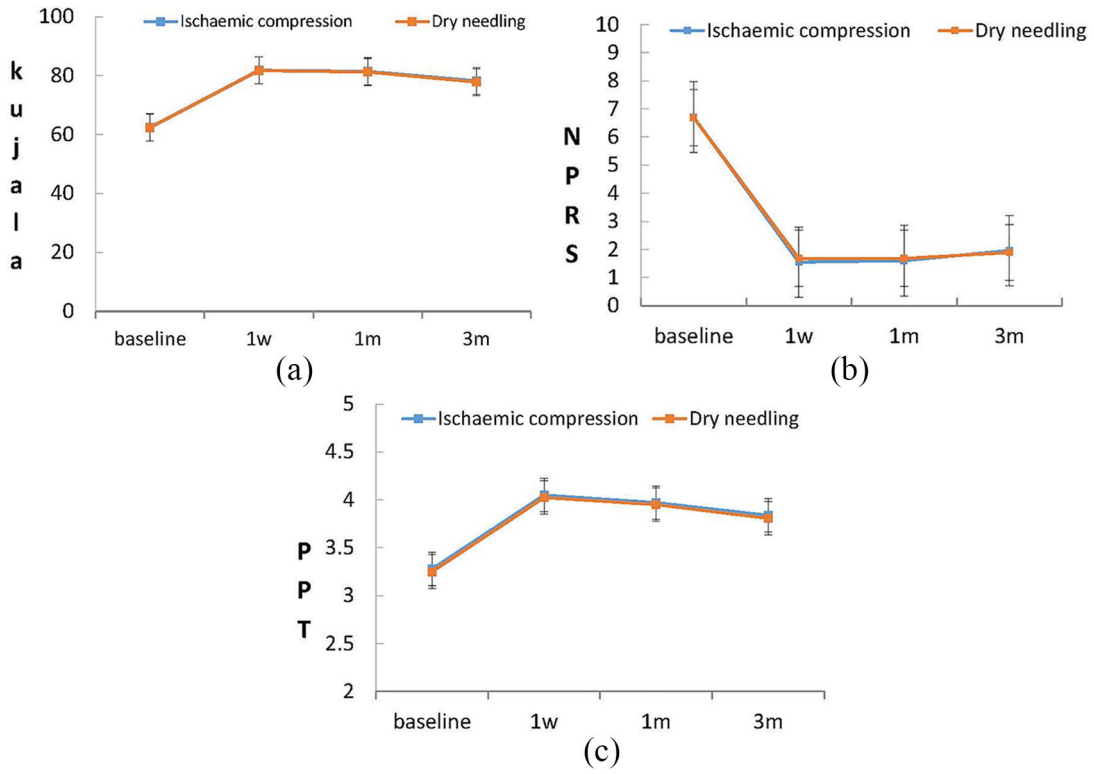



**Table 3.** Standard error of measurement (SEM) and minimal detectable change (MDC) values in the dry needling (DN) and ischaemic compression (IC) groups.

| Variable | Paired time points | DN group (n = 28) | | IC group (n = 28) | |
|---|---|---|---|---|---|
| | | SEM | MDC | SEM | MDC |
| Kujala | Baseline – 1-week follow-up | 5.67 | 15.5 | 5.88 | 16.13 |
| | Baseline – 1-month follow-up | 5.67 | 15.5 | 6.0 | 16.46 |
| | Baseline – 3-month follow-up | 6.5 | 17.8 | 5.7 | 15.6 |
| NPRS | Baseline – 1-week follow-up | 0.49 | 1.34 | 0.61 | 1.67 |
| | Baseline – 1-month follow-up | 0.62 | 1.7 | 0.33 | 0.91 |
| | Baseline – 3-month follow-up | 0.74 | 2.03 | 0.92 | 2.52 |
| PPT | Baseline – 1-week follow-up | 0.16 | 0.43 | 0.7 | 1.92 |
| | Baseline – 1-month follow-up | 0.18 | 0.49 | 0.12 | 0.32 |
| | Baseline – 3-month follow-up | 0.16 | 0.43 | 0.10 | 0.27 |

NPRS: numerical pain rating scale; PPT: pressure pain threshold.

hypothesis and more efficacious intervention programmes for patients with PFPS. Additional studies with more accurate diagnostic methods, such as X-ray, are also required.

## Conclusion

This study found that there were no significant differences between the DN and IC techniques in the treatment of

patients with PFPS. Moreover, the effect of time suggested that both techniques were similarly effective. The apparently beneficial effects of the interventions were evident, even at the 3-month follow-up.

## Contributors

S. B. is responsible for the conception and design of the study. She collected the data and wrote an initial draft. M.A.-A. read and revised the article critically for important intellectual content. M.A.-A. and A.K.Y. analysed the data. All authors contributed to the interpretation of data and reviewed and approved the final version of the article accepted for publication.

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## Ethical approval

This study was approved by the ethics committee of Babol University of Medical Sciences (reference no. MUBABOL. REC.1395.190).

## Trial registration

Trial registration no.: IRCT2016062028542N1.

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