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The Effect of Cold Therapy on Delayed Onset Muscle Soreness and Quadriceps Femoris Strength After High-Intensity Eccentric Training

Senouci Abdelkarim ^{1*} Asli Houcine ² Belkadi Adel ³ Bouhella Hafid ⁴ Koutchouk Sidi Mohamed ⁵ ^{1, 3,5} Institute of Physical Education and Sports, University of Mostaganem,27000 Algeria. ² University of Oran, 31000 Algeria. ⁴Institut of physical education and sports, University Alger 3,16000 Algeria.

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Abstract

This study investigated the immediate post-exercise effects of cold therapy on muscle strength, joint mobility, and pain perception in the quadriceps femoris muscle. Methods: Nineteen subjects were recruited in a strength training protocol, followed by a 60-minute cold bandage application to one leg. Muscle strength, joint mobility, and pain perception were assessed at baseline and 48 hours post-intervention. Results: The cold-treated leg experienced significantly less perceived pain during active knee extension. There was a significant increase in mean torque for the untreated leg at 180°/s. No significant difference was found in knee joint mobility, perceived exercise pain at Ely's test. Conclusion: While cold therapy may alleviate post-exercise pain, it does not appear to mitigate functional impairments. Further research is needed to clarify the mechanisms and optimal application of cold therapy in exercise recovery.

1. Introduction

Exercise soreness is a common phenomenon after eccentric, high-intensity or above-ground exercise (Fitzgerald, Rothstein, Mayhew, & Lamb, 1991; Peñailillo, Blazevich, & Nosaka, 2015). Reported symptoms of exercise-induced pain include inflammation, edema, aching, and reduced muscle function and joint mobility (Hody, Croisier, Bury, Rogister, & Leprince, 2019). There have been several approaches in

^{*} *E-mail*: abdelkrim.senouci@univ-mosta.dz, tel.00213662701880

the literature to try to treat the symptoms associated with exercise-induced pain (Hody et al., 2019; Serinken, Gençoğlu, & Kayatekin, 2013).

Cryotherapy is widely used in the treatment of muscle injuries and is also thought to be effective in the treatment of exercise pain (Hohenauer, Taeymans, Baeyens, Clarys, & Clijsen, 2015; Knight, Brucker, Stoneman, & Rubley, 2000). There is evidence in the literature to suggest that cold water immersion (CWI), a form of cryotherapy in which the affected limb is immersed in cold water, reduces the symptoms experienced with exercise-induced pain (Bleakley et al., 2012; Wilson et al., 2018). However, there is currently a lack of consensus on the effectiveness of the treatment and there are no guidelines on how to carry out the treatment (Kuenze & Hart, 2010). The authors sought to investigate the efficacy of cryotherapy and to study a treatment method using cold packs, which is more accessible than CWI for individuals who exercise and train at a moderate level of fitness (Bleakley et al., 2012; Wilson et al., 2018).

Muscle fibres are made up of small cylindrical myofibrils, which are in turn divided into segments made up of myosin and actin filaments(Plotnikov, Millard, Campagnola, & Mohler, 2006). When a nerve signal from the brain reaches the muscle, voltage-sensitive channels open into the myofibril. This results in a muscle contraction due to the sudden influx of Ca^{2+} into the muscle. As the muscle contracts, the myosin and actin filaments slide along each other.

The development of exercise soreness tends to occur within the first 24 hours after exercise, peaking after two to three days and symptoms have been reported to persist for several days (Barnett, 2006; Braun & Dutto, 2003; Cheung, Hume, & Maxwell, 2003). Symptoms associated with exercise- induced pain include inflammation, edema, decreased muscle function (Braun & Dutto, 2003; Eston & Peters, 1999; Mokhtar, Adel, & Wahib, 2019; Schoenfeld, 2012), decreased maximal muscle strength, pain in the area of injury and muscle tendon attachments (Guilhem et al., 2016; Slater, Arendt-Nielsen, Wright, & Graven-Nielsen, 2005; Yacine et al., 2020), and decreased mobility in affected joints (Cherara, Belkadi, Asli, & Benbernou, 2019; Jakeman, Macrae, & Eston, 2009).

There are several causes of exercise pain described in the literature. These can be divided into a primary and a secondary phase, where the primary phase involves the initial metabolic and mechanical effects, which damage the muscles during exercise(Berria, Bachir, Eddine, & Adel, 2018; Leeder, Gissane, Someren, Gregson, & Howatson, 2012; Proske & Morgan, 2001). The secondary phase, in turn, involves the subsequent effects of inflammation in the muscle (Howatson & Van Someren, 2008).

It has been suggested that the initial effects of eccentric training can be divided into metabolic and mechanical causes (Hedayatpour & Falla, 2015; Howatson & Van Someren, 2008; Tee, Bosch, & Lambert, 2007). The metabolic component has been suggested to be due to ischemia or hypoxia, as a result of prolonged exercise. Ischemia is thought to cause changes in local ion concentrations, an increase in metabolic waste and a deficiency of adenosine triphosphate (ATP) in the muscle. This ultimately results in tissue damage (Hedayatpour & Falla, 2015). In case of hypoxia in the tissue, the Na⁺/K⁺ pump is stopped, which increases the extracellular concentration of K⁺ and depolarizes the nociceptors and, through an increased impulse traffic, gives rise to pain (Mohamed, Mohamed, Mohammed, Mokrani, & Belkadi, 2019; Wright & Sluka, 2001). However, the metabolic effects have recently been reported as unlikely causes of exercise pain induced by eccentric exercise, and are instead thought to be limited to exercise pain induced by prolonged exercise (Belkadi, Alia, & Mohammed, 2020; Howatson & Van Someren, 2008; Jakeman et al., 2009; Proske & Morgan, 2001)

The mechanical hypothesis suggests that the damage is a direct consequence of the mechanical stress on the myofibrils. Eccentric contractions can generate more force than isometric and concentric contractions. In eccentric work, the sarcomeres work under uneven conditions, resulting in some of the myofilaments in the muscle being stretched and no longer overlapping inside the sarcomere. This causes a greater mechanical load on the fibers that are still overlapping (Guilhem et al., 2016; Plotnikov et al., 2006). The eccentric component and the associated mechanical load may be the main cause of the injury and ischemia then only tends to further aggravate the injury from the eccentric contractions. However, the exact underlying mechanisms of muscle injury occurring during exercise are not fully understood (Eston & Peters, 1999; Slater et al., 2005; Tee et al., 2007; Youcef, Mokhtar, & Adel, 2022).

In the literature, an increase in inflammatory markers in the blood after eccentric exercise has been observed (Beboucha, Belkadi, Benchehida, & Bengoua, 2021; Hedayatpour & Falla, 2015; Proske & Morgan, 2001; Tee et al., 2007). The secondary phase exacerbates muscle damage through the processes associated with inflammation (Proske & Morgan, 2001). The phase is initiated by a disturbance in intracellular Ca^{2+} -homeostasis, leading to further damage to the myofibrils of skeletal muscle (Belkadi, Benchehida, Benbernou, & Sebbane, 2019; Benhammou, Mourot, Mokkedes, Bengoua, & Belkadi, 2021; Tee et al., 2007). Eccentric exercise in rats has been shown to lead to an impairment of membrane integrity and thus an increase in the flow of Ca^{2+} into the intracellular environment (Gushchina et al., 2017) and a leakage of intramuscular proteins, which contributes to further aggravate the musculoskeletal damage ^{28,36}. The increased leakage of proteins also leads to edema at the local level (Mokhtar et al., 2019).

MacIntyre et al. studied the anterodistal part of the quadriceps muscles after 300 eccentric contractions of the right quadriceps muscles. It was observed that neutrophil activity in that part of the muscle was increased up to 6 hours after the exercise. It was concluded that a significantly increased infiltration of inflammatory markers is strongly related to exercise pain after eccentric exercise (MacIntyre, Sorichter, Mair, Berg, & McKenzie, 2001). Fielding et al. observed a direct correlation between intramuscular inflammatory markers and signs of muscle fibre damage. However, there is some literature suggesting that exercise-induced pain is not related to inflammation, but is instead an effect of the muscle's adaptation to exercise and not a muscle injury (Hody et al., 2019; Serinken et al., 2013; Vaegter & Jones, 2020)

A number of different interventions have been applied both preventively and

therapeutically to reduce the negative effects associated with exercise-induced pain (Rice et al., 2019). Some of the most commonly used methods in the literature are described below.

Muscle stretching has long been a frequently used method to relieve exercise pain, both preventively and therapeutically. The primary purpose of muscle stretching is to increase range of motion (ROM) and this can be achieved through a number of variations of stretches. However, there is not the same level of evidence for the intervention in relieving exercise pain (Hody et al., 2019)).

In elite athletes, there is extensive use of massage therapy, which is thought to reduce edema and pain and relieve exercise soreness, but also to improve the transport of lactate from the muscle's blood vessels through increased blood flow. However, studies show that only skin blood flow is increased by massage, and it has been found that lactate transport is thus not increased (Hody et al., 2019; Tee et al., 2007; Wilson et al., 2018).

Active recovery, i.e. lighter activity initially after exercise, is mainly based on the idea of increased lactate transport. However, most studies have shown that there is no correlation between recovery and lactate levels. Light physical activity increases muscle blood flow. Thus, active recovery seems to be a better option than massage, if increased blood flow has any effect on recovery (Barnett, 2006; Wilson et al., 2018). However, the evidence for active recovery is currently scarce (Jakeman et al., 2009; Leeder et al., 2012).

Transcutaneous electrical nerve stimulation (TENS) has been used for a wide range of musculoskeletal problems. There is some evidence in the literature that TENS has a good effect in the treatment of exercise pain. However, Howatson et al. argue that the equipment needed for the treatment is too expensive and difficult for the average person to handle. There are also currently no clinical guidelines on how to apply the treatment method in the treatment of exercise-induced pain (Howatson & Van Someren, 2008).

Cryotherapy involves the use of cold as a therapeutic intervention (Kuenze & Hart, 2010). The hypothesis is that the reduced tissue temperature results in a vasoconstriction of local blood vessels, thus reducing the inflammatory response and edema associated with muscle injury (Knight et al., 2000). The treatment method is used in the acute stage after, for example, a trauma and is suggested to reduce the negative effects of an acute muscle injury and is also believed to be suitable as a treatment for recovery after exercise (Belkadi et al., 2015; Serinken et al., 2013). In theory, the effect of cryotherapy is enhanced by cold water immersion (CWI), i.e. immersion of the limb in question in cold water. Cold water immersion as a treatment for exercise-induced pain is commonly used by elite athletes (Bleakley et al., 2012; Eston & Peters, 1999; Mokhtar et al., 2019). Sellwood et al. concluded from their study that CWI is ineffective in treating multiple parameters such as pain, swelling, function, maximal isometric strength and creatine kinase levels (Sellwood, Brukner, Williams, Nicol, & Hinman, 2007). There is some discrepancy in the literature regarding the effectiveness of the treatment and guidelines on how to perform the treatment (Bleakley et al., 2012).

The authors further wanted to investigate whether cryotherapy is effective as a treatment method for relieving exercise pain. One problem with CWI, according to the authors, is that the treatment method is cumbersome and the resources needed for treatment are massive.

Cryotherapy in the form of a cold bandage should be a more convenient and accessible option. The authors have not seen any research on the use of cold packs for exercise pain.

2. Material and methods

Purpose

The aim of the study was to investigate how CWI treatment with a cold bandage applied over the quadriceps muscle flexors immediately after performing concentric and eccentric training of the quadriceps affects muscle strength, thigh circumference, muscle flexibility and pain perception 48 hours after training.

Participants

The subjects in the study were all students at Institute of physical education and sports and were recruited via mass mail through Mostaganem University's mail system or orally through personal contact. Ten men and nine women between the ages of 21 and 31 were recruited for the study.

Inclusion and exclusion criteria

Inclusion criteria:

• Previous experience with exercise pain.

Exclusionary script:

• Exercise pain in the lower extremity at the first test.

• Injury or disease that impairs the ability to perform maximum muscular activity with the lower limbs.

• Lower limb training within three days before the first measurement.

Implementation

The study continued over a period of three days. On the first occasion, day one, the subjects received oral and written information about the study, and provided written consent to the study. It was noted which of the legs was dominant by asking the subjects to tell which leg they were kicking the ball with. Subjects were each given a personal code number. Before the strength measurements were performed, the subjects went through the other measurements, which were later followed by a five-minute warm-up on an ergometer bike set at the lowest load.

Immediately after the warm-up, the subjects performed strength measurements in the isokinetic training machine (BiodexTM System) and training of the quadriceps femoris in the form of concentric and eccentric contraction through a leg park exercise. After the leg park exercise, the subjects performed jumps.

Anthropometric data

All measurements, review of set-up and information, and training were conducted in the Laboratory for Optimizing Research Programmes on Physical and Sports Activities. The laboratory was equipped with an isokinetic training and measuring machine (Biodex system 4) as well as an ergometer bike, brits and the evaluation instruments described below. Each evaluation instrument is described below in more detail and individually in the chronological order in which they were carried out.

When the subject was lying supine on a cot, a line was drawn between the superior iliac spine (SIAS) and the base of the patella. One measurement point was in the middle of this line, where the rectus femoris muscle bulge is thought to be at its largest, and the other measurement point was 10 cm cranially about the base of the patella, about 5 cm above the junction between the muscle and the patellar tendon. The measurement points were marked with a marker pen at the first measurement session and were used as measurement points also at the second measurement session. The circumference was measured with a tape measure and was given in centimetres with an accuracy of 0.5 cm. The authors have not seen any reliability testing in the literature regarding the error margin of the tape measure.

Muscle flexibility when measuring range of motion with Ely's test, a standardized goniometer was used. The subjects lay on a bench in the prone position and test leader 1 performed a passive maximum flexion of the knee joint, after which the range of motion was recorded using the goniometer. The measurement was performed on both legs separately. The reference points for the measurement were the longitudinal direction of the femur and fibula, respectively (Gogia, Braatz, Rose, & Norton, 1987). Measuring the mobility of the knee joint with a goniometer has proven to be a valid method for this particular purpose. Similarly, both inter- and the intra-rater reliability of the assessment instrument has been shown to be high. There is only moderate support for the reliability of Ely's test (Peeler & Anderson, 2008).

Experienced exercise pain

During the second measurement session, subjects were asked to rate their experience of exercise pain by verbally giving a number between 0 and 10, where 0 represents "no exercise pain" and 10 "worst possible exercise pain". Exercise pain was measured by VAS and subjects were asked to rate it individually for each thigh, during two different activities.

Strength measurements

After the warm-up, the subjects were given a verbal briefing on the isokinetic machine and the procedure of the exercise to be performed. The machine was then adjusted to the subject according to the Biodex System 4 manual. The range of motion was set for the knee joint in question at 0°-90°. The strength measurement protocol was then performed and consisted of concentric contractions of the hamstring and quadriceps femoris muscles only. The measurements were performed at 60°/s, and 180°/s. Subjects were instructed to perform 3 submaximal contractions and then with maximal force to extend the knee joint and with maximal force to flex the knee joint. Subjects performed 5 maximal contractions at 60°/s and 10 maximal contractions at 180°/s. Between the tests at the different speeds, the subjects were allowed to rest for 30 seconds. The subjects were allowed to rest for about two minutes after the measurements, before the strength training was performed. The BiodexTM System 3 has been shown to have a high reliability for isokinetic measurement data (Lund et al., 2005).

Implementation of strength training Intervention

Immediately after training, each subject received unilateral cold of the left m. quadriceps. A cold bandage (IceBand® Knee, IB Medical AB), was applied over the left thigh. No specific reference points were used for the application of the cold bandage. Test leader 1 checked that the cold bandage was in place with the lower edge cranial to the patella. Subjects kept the ice bandage on for 60 minutes and were instructed to walk or cycle home, but otherwise to perform as little physical activity as possible while the ice bandage was on. All subjects received their own cold bandage.

Statistical analyses

All data from the protocols were manually transferred to the Statistical Package for the Social Sciences (SPSS) version 22 for Windows. All analytical calculations were performed in SPSS. All variables were normally distributed, and therefore for descriptive data the mean and standard deviation (SD) were obtained. To demonstrate statistical significance, paired t- tests were used. The significance level was set at 5% ($p \le 0.05$). The percentage difference from time 1 to time 2 between the right and left leg was determined by dividing the mean of time 2 by the mean of time 1. This difference was then treated in the same way as the other normally distributed variables by analysis with a paired t-test.

3. Results and Discussions

A total of 19 subjects participated in the study. Of these, 10 were men and 9 were women. All subjects completed all study components, so there was no dropout. The mean age of the subjects was 24 years, with a standard deviation (SD) of 3 years.

	T1							T2				
Right			Left leg*		n volvo	Right leg		Left leg*				
Variable	mean	SD	mean	SD	p- value	mean	SD	mean	SD	p- value		
Thigh circumferenc 1	47,8	2,6	48.02	2,6	0.853	48,7	2,7	48,6	2,6	0.474		
Thigh circumferenc 2	57,1	2,7	56,6	2,9	0.033	57,6	2,8	57,5	3,1	0.574		

Table 1. the difference between the left and right leg (p-value) at time 1 and 2, for the measured thigh circumference measured in centimetres (cm)

* Left leg received cold treatment on occasion 1

When measuring thigh circumference at a distance of 10 cm cranially about the base of the patella (thigh circumference 1), there was no significant difference between the legs at time 1 (Table 1), nor was there a significant difference between the legs at time 2 (Table 1).

In the measurements where the thigh was assumed to be the largest (thigh circumference 2), there was a significant difference between the legs at time 1 (Table 1). There was no significant for thigh circumference 2 between legs at time 2.

Table 2. Mean (median) and standard deviation (SD) for right and left leg
at time 1 and 2, and the significance value for the difference between left
and right leg (p-value) at time 1 and 2, for Ely's test measured in degrees (°)

	Right leg		Left	Left leg*		Righ	t leg	Left l	eg*	
Variable	mean	SD	mean	SD	p- value	mean	SD	mean	SD	p- value
Ely's test	140	6,9	140	6,3	0.578	142	5,5	141	4,9	0.749

* Left leg received cold treatment on occasion 1

When comparing the legs individually between time 1 and 2, a significant increase was found for both the right and left leg at both measurement sites. Between session 1 and 2, the difference measured for measurement site 1 became significant for the right and left leg (p =0.001 and p = 0.006 respectively). The same analysis for measurement point 2 yielded a significant difference for the right and left leg (p = 0.001, respectively).

When comparing the difference (measured in percent) from time 1 to time 2, between the two legs at measurement site 1, no significant difference was found (p = 0.335). In the same analysis for measurement point 2, no significant difference was found (p = 0.100).

Table 3. Difference between left and right leg (p-value) at time 2,for self-rated exercise pain during Ely's test and duringactive maximal knee extension

	Right le	g	Left leg	p- value			
Variable	mean	SD	mean	SD			
Ely's test	4,6	2,5	3,7	2,9	0.142		
Knee extension	5,0	1,8	3,3	2,0	< 0.001		

* Left leg received cold treatment on occasion 1

When comparing the difference in range of motion (measured in percent) between session 1 and 2, between the two legs, no significant difference was found (p = 0.942).

Self-rated exercise pain during Ely's test and knee joint extension No significant difference between legs in self-rated exercise pain during Ely's test at time 2 was observed (Table 3). Significant differences between legs were found when comparing self-rated exercise pain during active maximal knee extension at time 2 (Table 3), with the advantage of the left (treated) leg.

Table 4. The difference between left and right leg (p-value) at time 1 and 2, for maximum
(PT) and average torque (APT) measured in newton metres (Nm) and total work (TW)
measured in joules (J), at speed $60^{\circ/s}$

	Right leg		Left leg		p- value	Right leg		Left leg*		p- value
	mean	SD	mean	SD	-	mean	SD	mean	SD	- -
РТ	192,5	59,7	195	49,2	0.656	173,2	57,8	182	54,1	0.062
AT	176	57	180,2	42,8	0.458	161	52,8	163,3	51,4	0.712
TW	879,7	274,9	911	224,8	0.3	810,2	267,7	859,3	245,5	0.161

* Left leg received cold treatment on occasion 1

Maximum torque, average maximum torque and total work Measurements at speed 60°/s: No significant difference was found when comparing maximum torque (PT) between the right and left leg, neither at session 1 nor 2 (Table 4).No significant difference was found when comparing the average maximum torque (APT) between the right and left leg, neither at time 1 nor 2 (Table 4).No significant difference was found when comparing total work done (TW) between the right and left leg, neither at time 1 nor 2 (Table 4).

Table 5. The difference for each leg between session 1 and session 2,in terms of maximum (PT) and average torque (APT) and totalwork (TW at speed 60°/s

Variable (opportunity 2 /opportunity 1) (%)	Right leg		Left leg*		p-value
	mean	SD	mean	SD	
РТ	92,4	25,1	94,2	20	0.567
APT	95,8	31,8	91,2	21,6	0.358
TW	96,1	32,3	95,3	17,3	0.883

* Left leg received cold treatment on occasion 1

When analysing the difference in PT, APT and TW (measured in percentage) between session 1 and 2, between the two legs, no significance was found (Table 5).

Table 6 The difference between left and right leg (p-value) at time 1 and 2, formaximum (PK) and average torque (APK) measured in newton metres (Nm) and total work(TW) measured in joules (J), at speed 180°/s

Variable	Right leg		eg Left leg		p- value	Right leg		Left leg		p- value
	mean	SD	mean	SD	-	mean	SD	mean	SD	
РТ	131,9	40,5	136,2	39,3	0.301	132,3	42	133,5	37,6	0.601
APT	114	37,3	121,4	37,1	0.006	120,4	38	120	34,1	0.886
TW	1250,9	395	1364	400,2	0.002	1320,2	422,2	1369,1	357,6	0.079

* Left leg received cold treatment on occasion 1

Measurements at speed 180°/sNo significant difference was found when comparing maximum torque (PT) between the right and left leg, neither at session 1 nor 2 (Table 6).No significant difference was found when comparing APT between right and left legs at time point 2 (Table 6). When analysing the difference in APT between the right and left leg at time 1, a significant difference was found (Table 6).No significant difference was found when comparing TW between right and left legs at time point 2 (Table 6). When analysing the difference in APT between right and left legs at time point 2 (Table 6). When analysing the difference was found (Table 6).No significant difference was found when comparing TW between right and left legs at time point 2 (Table 6). When analysing the difference in TW between right and left legs at time 1, a significant difference was found.

Discussions

The aim of the study was to investigate the effects of cryotherapy in the form of an ice bandage, and its suitability as a preventive treatment against adverse effects associated with exercise pain. The effects of cryotherapy have been studied previously, including the use of CWI, but the authors have not found any literature in which the effects of cryotherapy in the form of an ice bandage have been investigated. The literature calls for more research in this area, particularly with regard to the type of cold and duration to be applied during treatment (Howatson & Van Someren, 2008).The authors therefore considered it relevant to perform an experimental study focusing on cryotherapy, and specifically on the use of an ice bandage. All measurements, calibrations and procedures were carried out according to the pre- established protocol, which means that the authors, as test supervisors, always performed the same repetitive tasks during the meetings with the subjects. This was done to increase inter- rater reliability. To further improve the methodology, external, blinded colleagues could have carried out the measurements, in an attempt to counteract possible bias.

In the current study, the left leg was treated with cold packs on all participants. A randomization of legs for treatment would also have increased methodological quality. A weakness of the study is that the authors did not control for adherence to treatment or activities performed by the subjects during the study period. Participants in the study did not know in advance that they would keep the ice bandage. This was to avoid that participants recruited for the study would volunteer only because they received a gift, which could potentially affect the results.

As exercise pain is strongly related to inflammation (Miles & Clarkson, 1994; Sluka, Law, & Bement, 2018), the circumference of both thighs was measured. In order to obtain a good interobserver reliability, the measurement points on the thigh were marked with a marker pen, so that the same measurement points could be used at both measurement occasions. The authors have not seen any reliability testing in the literature regarding the margin of error of the tape measure. Thus, no statement can be made regarding the tape measure as a measurement method.

Measuring the mobility of the knee joint with a goniometer has proven to be a valid method for this purpose (Gogia et al., 1987). Similarly, both inter- and Intrarater reliability of the assessment instrument has been shown to be high (Lund et al., 2005). However, intra-rater reliability appears to be slightly higher than inter-rater reliability (Hedayatpour & Falla, 2015). Ely's test is a commonly used orthopaedic test for measuring rectus femoris flexibility and mobility of the knee joint, yet there is only moderate scientific support for its reliability. In one of the few studies that have measured the reliability of the instrument, the minimum difference between two separate measurements allegedly needs to exceed at least 11 degrees to be considered real (Peeler & Anderson, 2008) Ely's test was chosen as an evaluation instrument because the length of the entire knee joint is m. quadriceps could be evaluated. In the case of knee flexion alone without simultaneous hip extension, the length of the rectus femoris would be neglected.

In previous studies, cycling (ergometer bike) has been used as a warm-up for measurements in an isokinetic machine (Aguilar et al., 2012; Portes, Portes, Botelho, & Souza Pinto, 2007). Different loads and durations have been used. The authors considered 5 minutes to be sufficient time for warm-up, in accordance with Krishnan et al. and Portes et al. The authors did not seek to fatigue the subjects before the exercise- induced exercise and thus chose to set the bicycle to the lowest possible load during the warm-up (Aguilar et al., 2012; Krishnan & Williams, 2009; Manar, Adel, Lalia, & Saddak, 2023). This type of warm-up and setting has been implemented in previous studies (Portes et al., 2007). Lund et al have tested the reliability of the Biodex system 3 as an isokinetic measuring instrument. It was concluded that the isokinetic measurement data obtained from the Biodex[™] System 3 has a high reliability (Lund et al., 2005). The speeds of 60 and 180°/s used during the measurements and the induction of exercise pain are common in the literature where isokinetic testing is concerned. Usually, subjects are tested with the lowest speed first (Aguilar et al., 2012; Diracoglu, Baskent, Yagci, Ozçakar, & Aydin, 2009; Krishnan & Williams, 2009; Portes et al., 2007; Stupka et al., 2000). Before the actual measurements begin, test repetitions are often performed. Several papers used a protocol of 3 submaximal test repetitions, which subjects performed once for each speed, before the measurements started (Gogia et al., 1987; Krishnan & Williams, 2009).

Repeatedly in the literature, researchers have used leg kicks to induce exerciseinduced pain (Sluka et al., 2018). Furthermore, the number of repetitions has been 10 per set in almost all studies found (MacIntyre et al., 2001; Sellwood et al., 2007). However, the number of sets varied, from 3 to 6 (Stupka et al., 2000).

The authors chose to perform 6 sets of 10 repetitions (Braun & Dutto, 2003), Warren et al. suggest that the induction of exercise soreness should be similar to the strength tests (Weerapong, Hume, & Kolt, 2005). Henceforth, the authors considered it appropriate to use 3 sets of 10 repetitions at 60°/s and 3 sets of 10 repetitions at 180°/s. In order to ensure that the subjects actually experienced exercise pain, 100 upward jumps were also added to the exercise protocol. This has been shown to be an effective way of inducing exercise- induced pain (Serinken et al., 2013; Sluka et al., 2018; Vaegter & Jones, 2020)During both the measurements and the training session, the test supervisors used verbal encouragement, which has been shown in previous literature to increase performance when measuring with isokinetic measuring equipment (Moussa, Zouita, Salah, Behm, & Chaouachi, 2020). The use of the VAS and a Likert scale to assess exercise pain was studied. It has been reported that the VAS is more sensitive to changes in the perception of pain. Vickers et al. have investigated the exercise pain experience of untrained/inactive people (Harbach, Sifi, Zabchi, & Mokrani, 2022; Vickers, Fisher, Smith, Wyllie, & Lewith, 1997). It can be assumed that these individuals do not have much experience with exercise pain. Thus, a disadvantage of the VAS may be a difficulty in grading the soreness in relation to the expressions "no pain" and "worst possible pain". In the current study, the authors investigated individuals with previous experience of exercise pain, so the authors chose to use the VAS to evaluate soreness. The authors have not seen any validity testing of the VAS in exercise pain. However, the VAS is the instrument that appears to be most prevalent in the literature addressing perceived exercise pain(Adel Belkadi et al., 2020).

Gender differences in exercise-induced pain have been reported in humans and animals. Stupka et al. had eight women and eight men perform heavy eccentric exercise (120% of 1 RM). The results showed that the gender differences in the effect of exercise pain are not due to the damage sustained by the sarcomeres, but are a consequence of the inflammatory response. Women were found to have a less pronounced inflammatory response despite the presence of the same mechanical injury(Boudehri, Belkadi, Dahoune, & Atallah, 2023). The treatment in this study was based on inflammation suppression by cold. In case of a low inflammatory response, treatment may therefore have failed. Thus, no significance level would be reached if, due to these gender differences, the treatment works on men but not women. However, most studies have shown that there are no gender differences in relative maximal isometric strength after repetitive maximal eccentric contractions. In one study, women were shown to have a greater relative loss of strength immediately after exercise(Abdelkader et al., 2021). Studies in the literature have also shown that the recovery of strength after eccentric exercise does not differ between men and women. In studies assessing exercise pain, no gender differences or significant associations with oestrogen levels have been observed. However, in one study, differences between men and women have been reported for exerciseinduced pain. However, this study only looked at the first 24 h after, taking into account that exercise- induced pain peaks after 48-72 h (Hody et al., 2019; Sluka et al., 2018). It would have been interesting to study possible gender differences in the current study, but due to the small size of the study material, the authors chose not to perform such an analysis.

4. Conclusions

Cold treatment in the form of a cold bandage applied over the quadriceps femoris immediately after high-intensity concentric and eccentric training may possibly alleviate perceived training pain, but does not counteract reduced average maximum torque, maximum torque, total work done and range of motion, and does not reduce swelling. Against the previous literature and the discrepancy therein, further research is necessary to determine the effects of cold treatment on exercise pain.

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