Acute Effects of Cold Pack for Different Periods on the Biomechanical Properties of the Rectus Femoris Muscle

Nuray Alaca, PhD; Nilüfer Kablan, PhD

ABSTRACT
Context • Cold packs are silica gel packs that are commonly used in clinics. However, the packs are applied for various amounts of time, and the relationship between these times and temperature changes isn’t fully understood.
Objectives • The study intended to investigate the acute effects of cold-pack application for different periods of time on the biomechanical properties of the rectus femoris muscle.
Design • The study was randomized, controlled trial.
Setting • The study took place at Acibadem Mehmet Ali Aydinlar University in Istanbul, Turkey.
Participants • Participants were 60 healthy volunteers from the community, aged 18 to 23 years.
Interventions • Participants were divided into four groups with n = 15 in each group. The cold packs were applied on the dominant rectus femoris muscle: (1) for 10 minutes in Group 1, (2) for 12 minutes in Group 2, (3) for 15 minutes in Group 3, and (4) for 20 minutes in Group 4.
Outcome Measures • The outcome measures were the skin temperature, determined using a thermal camera, and biomechanical properties—tone and stiffness and muscle decrement—using a device that delivers a short mechanical impulse to the tissue. Outcomes were measured at baseline before the cold application, immediately post intervention after the cold application, and at 5, 10, 15, 20, and 30 minutes post intervention.
Results • The mean skin temperatures were significantly lower in all groups compared to those before cold application (P < .05), and no significant differences existed between any of the groups (P > .05). Post intervention, while Groups 1 and 2 showed an increase in muscle tone and stiffness and a decrease in elasticity (P < .05), they began to approach their baseline state by the fifth and fifteenth minutes, respectively (P > .05). In Groups 3 and 4, the muscle stiffness increased at all time points (P < .05).
Conclusions • The study showed that the rectus femoris muscle of healthy people becomes stiffer and less elastic as a result of cooling with cold packs that were applied for different time periods. The amount of cold-pack time that minimized the biomechanical corruption of the muscle and provides cooling was 10 minutes. Careful warming up is recommended before and after intense athletic performance, and caution in cooling the skeletal muscle should be exercised. (Altern Ther Health Med. 2021;27(5):92-99).

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Cryotherapy is used in various clinical and sports environments to reduce edema, nerve-conduction velocity, and tissue metabolism as well as to facilitate recovery after exercise-related muscle damage.1,2 Tissue thermodynamics have shown that local cooling produces significant temperature drops in superficial and deep tissues. Although both superficial and deep intramuscular tissues reach a minimum temperature in the post-cooling period, deeper tissues will reach a nadir in temperature later in the post-cooling period as heat from deep tissues is lost to colder superficial tissues.3 After a cold application, a rapid increase occurs initially in the superficial tissue temperature, while a slow recovery is observed in the deep tissue, with a gradual decrease in temperature.4,7 The first physiological response to
temperature change is to alter local cellular activities, including local control of blood flow and signal transduction in the efferent nerves. The second change may generate specific temperature signals in the nerves or alter nonthermal, afferent-nerve information, and thereby, elicit reflex responses of different degrees of complexity. These responses feed from the central nervous system back to the body by innervating the somatomotor or autonomic system or even by altering hormone levels. Third, and physiologically most important, thermoregulation changes various activities in the body if its heat balance becomes disturbed, with the aim of reestablishing the balance between heat production and heat loss.5

Local physiological responses that occur due to cooling include decreased circulation, metabolism, pain, edema, and responsiveness of the contractile system.6,7 Some studies8,9 have shown that local cold application can cause an increase in resistance against movement. This can make it difficult to perform physical activities after cold application because the cold can cause the agonist muscle to expend more energy to reach the desired performance and endurance and may cause an increase in the stiffness from injury due to the deterioration of muscle flexibility.

### Cold Application Duration

Despite the widespread use of cryotherapy, limited and inconsistent information is available about the most effective application mode, cold-application duration, physiological changes after cold, and effects in the other areas.4,12,13

According to in-vitro animal studies, muscle tissue shows high resistance to stretching after it has cooled.14-16 Akgün et al8 stated that they applied cold packs on dogs’ legs for different periods of time, from 10 to 30 min, and skin temperature (13.4–16.2°C) and intramuscular temperature (3.8–5.0°C) decreased more as the application time increased.

In a review on cryotherapy treatments published in 2001, Auley9 indicated that cold applications don’t have a specified frequency and duration and showed that cold had been applied for 5 to 85 min in many studies. Auley reported that application of a cold pack on the skin for 15 minutes decreased the temperature an average of 7.3°C, as measured by telemetriography, and stated that some evidence existed of a temperature drop at between 10 minutes and 20 minutes of cold application that was smaller than the drop in the first 10 minutes. However, it should be remembered that the temperature drop is also related to the contact area between the surfaces, the temperature difference, and the tissue conductivity.

When Ring et al10 in 2004 applied 15 minutes of cold to the L4 level of the spine, the researchers observed an average temperature decrease of 7°C. However, Kennet et al19 showed that applying a cold pack for 20 minutes to the right ankle provided 12°C of cooling.

Costello et al.20 in a review published in 2012, summarized the studies evaluating the effects of various cryotherapy methods on skin temperatures, using thermal cameras. They observed that few studies have occurred on this characteristic, even though the most commonly used cryotherapy method in clinics is the cold-pack application; the studies have generally been comparisons of other cryotherapy methods.

In human studies, only one study has evaluated the direct mechanical responses of the muscles after cooling. In that study by Mustalampi et al,4 parameters [frequency (tension), decrement (elasticity) and stiffness] were measured indicating the biomechanical properties of the muscle immediately after application of a cold pack to the quadriceps muscle, for 20 minutes or 15 minutes post intervention. The study found that the muscle became stiffer and less elastic in terms of its biomechanical properties.

Although Jutte et al21 have shown that the intramuscular temperature after cold application is related to the skinfold measurement (subcutaneous fat thickness), the application’s duration, the individual’s core temperature, and the room temperature, some researchers were concerned about the cold-application duration and the intervals between treatments.4,12,13 The researchers also noted that more studies were needed on the appropriate application duration and intervals.20

Acute trauma management and rehabilitation requires a determination of the duration of the cold application that will achieve effective cooling and produce the least change in the muscles’ biomechanical properties.

### Cold Packs

A cold pack can be applied for a relatively long time without losing its coldness due to its low conductivity.1-4

Cold-pack treatment can be applied for quite different periods of time, and the relationship between those times and temperature changes aren’t fully understood. Determining the most effective and shortest application time is crucial for clinical use.4 In addition, it’s important that the biomechanical changes that occur in healthy subjects after cooling—in the muscles’ stiffness and elasticity—return to the state before cold application as soon as possible. A careful warm-up is suggested after cooling before performing maximally.9

### Muscle Stiffness and Elasticity

The oscillation stiffness parameter characterizes the muscle’s ability to resist a force that shapes it. Increased stiffness means more energy is needed to change the shape of the muscle.10,22 Increased stiffness can cause damage to the stretched muscle group (antagonist).

Elasticity refers not to the ability of a muscle to stretch (elongation), but to its ability to return to its original shape after a force that creates deformation in the muscle.22 The higher the decrement parameter, the less the elasticity.22-24

Noonan et al22 found that increased biomechanical stiffness had occurred in chilled rabbit skeletal muscles. Mutungi and Ranatunga25 showed that the stretching of rat muscle-fiber bundles increased at low experimental temperatures.

Some studies looking at indirect muscle parameters had similar findings. Price and Lehmann25 found that 30 minutes
of cold increased ankle-joint stiffness. Similarly, Muraoka et al\textsuperscript{19} determined the same results for legs after 60 minutes of a cold-water bath. Lakie et al\textsuperscript{18} showed that cooling the forearm in cold water caused a stiffer wrist for joint movements. Unlike other studies, Kubo et al\textsuperscript{23} found that cooling the leg for 30 minutes with cold water didn’t change the ankle-joint stiffness or the extension of the joint motion for the joint fascicle, tendon, and aponeurosis.

**Current Objective**

The intended purpose of the current study was to investigate the acute effects of cold-pack therapy for healthy individuals on the skin temperature and the biomechanical properties of the rectus femoris muscle during its use for different periods of time—10, 12, 15, and 20 minutes.

**METHODS**

**Participants**

The study was a randomized controlled trial that was completed between February and March 2020. It took place at Acibadem Mehmet Ali Aydinlar University in Istanbul, Turkey. Participants were volunteers from the community who responded to the study’s advertisement, posted via social media. After 80 applications, the advertisement was removed via social media. 62 people came to the evaluation. First author managed the intake this study. Prospective participants were included in the study if they: (1) had a subdermal fold thickness of the quadriceps muscle between 5 mm and 15 mm, (2) didn’t smoke or use any drugs, and (3) had no history of cardiovascular or peripheral vascular disease, diabetes mellitus, neuromuscular pathology, peripheral neuropathy, lower extremity pain, or previous lower extremity surgery.\textsuperscript{19,26} Individuals who had a history of insensitivity to local heat or cold were excluded from the study. Sociodemographic data were evaluated with a survey.

The research team obtained approval from the local ethics committee (Atadek-2019/17/19) and retrospectively registered the study under clinical trial number (NCT04277481). All procedures were performed in accordance with the 1964 Helsinki Declaration, and the participants signed an informed consent form.

**Procedures**

**Sample size.** The GPower V.3.1.7 program (Kiel University, Kiel, Germany) was used to determine the appropriate sample size. To obtain the power ratio of the study as 80% at a 95% confidence limit, with 0.05 error margin, the effect size was determined to be 0.44. The research team calculated the oscillation frequency post intervention for the 0-minute mean (SD) to be 14.3 (1.8), the post intervention 15-minute mean (SD) to be 13.5 (1.4), and the sample size.\textsuperscript{9} Using this method, the sample size was determined to be 60.

**Skinfold measurement.** A Saehan skinfold caliper (Gyeonggi-do, Namyangju-city, South Korea) was used to measure the subdermal fold thickness. The measurement was made while the individual was in a standing position, with his or her body weight being transferred to the unmeasured foot. The knee on the limb measured was slightly flexed at 15 to 30 degrees, and the foot was in loose contact with the ground.

For the measurement, the skin and subcutaneous adipose were separated from the muscle by the physiotherapist’s thumb and forefinger. The arms of the skinfold instrument were placed on the skinfold, and the caliper was read in millimeters (mm). The measurement was repeated 3 times; the average was taken and recorded in mm.

**Randomization.** The participants were randomly assigned to groups using the block randomization method and divided into four groups with the aid of a Random Allocation Software computer program created by one member of the research team.

**Cold pack application.** A 35*29 cm cold pack that used a stitch-free technology and a chemical gel in a plastic bag (Merck, Kenilworth, New Jersey, USA). The pack was kept at -17, -20°C and was applied without compression. The application was achieved by wrapping a thin towel on the rectus femoris part of the quadriceps, with the towel cloth being used to prevent the individual from developing a cold burn.\textsuperscript{19}

**Outcome measures.** To help balance systemic blood flow, participants stopped exercising 12 hours prior to and caffeine and alcohol consumption one hour prior to baseline. All participants were evaluated between 12:00 and 1:00 PM. Individuals were asked to be in the room 20 minutes before the evaluations. This time interval was chosen to adapt participants’ skin surface temperature to the temperature of the room and to control circadian changes in body temperature.\textsuperscript{26}

All measurements were carried out by the same physiotherapist, one member of the research team, at a normal room temperature of 22-24°C and relative humidity of <50%, with the room condition being monitored by a thermohygrometer (PCE-HVAC 3, PCE Deutschland, Meschede, Germany).

**Intervention**

Participants were divided into four groups with n = 15 in each group. The cold packs were applied on the dominant rectus femoris muscle: (1) for 10 minutes in Group 1, (2) for 12 minutes in Group 2, (3) for 15 minutes in Group 3, and (4) for 20 minutes in Group 4.

**Outcome Measures**

The primary outcomes of skin temperature and biomechanical properties were measured at baseline before cold application, post intervention immediately after cold application, and at 5, 10, 15, 20, and 30 minutes post intervention. The times for application duration and measurement minutes were chosen because they have commonly been used for cold-pack application in the literature.\textsuperscript{17,20} Since the rectus muscle is widely used with
studies of biomechanical properties and with thermal cameras, the research team decided to use that muscle in the study.17,20,22,24,27,28

**Skin temperature.** The skin temperature was measured with the help of a noncontact thermal camera, a P45 thermographic camera with high thermal sensitivity (Flir System, ThermaCAM, Täby, Sweden). This technique is a safe and noninvasive method. Noncontact thermal cameras use a technology that is a complex way of receiving electromagnetic radiation and converting it into electrical signals. These signals are finally displayed in grayscale or in colors that represent temperature values. Noncontact thermal cameras have recently become popular for evaluating skin temperature following cryotherapy in individuals, such as cold-air cryotherapy, cold-water immersion, or ice-cube or cold-pack applications.20

The skin temperature of the area to be treated, the rectus femoris muscle, was measured while the participant was standing upright. Using the FLIR Quick-Report 1.2 software, the skin temperature was determined from the heat data obtained from the region. In the calculation of skin temperature, the emissivity value of human skin was accepted as 0.98.27

**Biomechanical properties.** Abnormally high muscle tone and high intramuscular pressure causes muscle to become tired more quickly and to delay muscle recovery by limiting blood flow.29 This increase in intramuscular pressure can put pressure on the blood vessels in the muscle, leading to poor circulation.30 These deficiencies may include muscle pain, fatigue, and weakness.31

Muscle tone, stiffness, and elasticity were measured with the MyotonPro (Myoton, Tallin, Estonia), which is a recently developed and reliable device that measures the passive mechanical properties of superficial muscles. It gives a short mechanical impulse to the tissue and records the mechanical response of the muscle through an acceleration probe. The frequency (muscle tone), decrement (elasticity), and stiffness of the oscillation are calculated from the acceleration wave obtained. The device can measure subcutaneous tissue up to a 2-cm depth by applying low-intensity (0.58 N) and a short (15 ms) mechanical impulse on the skin overlaying the muscle.22

The acceleration value of the first period after the oscillation caused by the stroke of the device's probe shows the deformation in the muscle; the frequency of the oscillation (Hz) is obtained by simple manufacturer calculation from the next oscillation period. The frequency of the damped oscillation gives information about muscle tone.

Dynamic stiffness (N/m) is a muscle's biomechanical property that characterizes the resistance to a contraction or an external force that distorts its initial shape. Increased stiffness in the muscle means that greater strength is required for the antagonist muscle to create movement and that it’s not economical for movement. The calculation of the logarithmic decrease of the damped oscillation shows the muscle's elasticity properties.

Elasticity is defined as the ability of a structure that changes shape due to a force applied to it, to return to its former shape. The higher the decrement parameter, the lower the elasticity.22,24,28 The MyotonPro has good test-repeat reliability (ICC: 0.84-0.85) in the measurement of the passive mechanical properties of the fragment of the quadriceps rectus femoris.24

Measurements were carried out by the physiotherapist while the participant was in a resting position, without active muscle contraction. While the participant was in a long sitting position (sitting with legs extended), with the knee being slightly flexed (15-30 degrees), a measurement was made from the middle region of the rectus femoris muscle—from the middle of the line extending between the anterior superior iliac spine and the upper edge of the patella.28

**Statistical Analysis**

The statistical analyses were performed using SPSS 21.0 program (SPSS, Chicago, IL, USA). Normality was evaluated using the Shapiro-Wilk test. Descriptive statistics are given as the mean ± the standard deviation or the frequency in the evaluation of the data. A repeated measures analysis of variance (ANOVA) was used to evaluate variables, and the paired sample t-test and Bonferroni test was used to evaluate binary comparisons. A one-way ANOVA and Tukey's posthoc test were used to compare the groups. A Pearson correlation analysis was used to evaluate the correlation between the skin temperature and the muscle's mechanical properties. Significance criteria was accepted as $P<.05$.

**RESULTS**

**Participants**

Figure 1 shows the participant flow diagram. Sixty volunteers, 30 females and 30 males aged 18 to 23 years were included in the study. Their analyses were all included in the overall results. The demographic data are shown in Table 1. No statistical significance existed in the comparison of the demographic information of the groups at baseline ($P>.05$).

**Skin Temperature**

The results of the mean repeated measurements of ANOVA in the skin-temperature evaluations were significant ($P<.001$). The mean skin temperatures (Table 2) were significantly lower for all groups at all times post intervention compared to before application ($P<.05$). No significant differences existed between any of the groups post intervention ($P=.91$ to $P=.830$, data not shown).

**Biomechanical Properties**

The results of the repeated measure ANOVAs related to the muscle's passive mechanical properties (Table 3) were significant ($P<.01$).

**Frequency.** The frequency measurements evaluating muscle tone increased significantly in Group 1 immediately post intervention ($P=.021$) and then decreased at 5 minutes post intervention ($P=.467$) and increased again significantly.
In Group 3, a significant increase in the frequency was observed only immediately post intervention ($P < .001$); at other times, no significant changes occurred ($P > .05$).

No significant differences in frequency occurred in Group 4 between baseline and any time post intervention ($P > .05$).

Also, no significant differences were found in frequency between any of the groups ($P = .215$ to $P = .548$, data not shown).  

**Stiffness.** Compared to baseline, muscle stiffness significantly increased in Group 1 immediately post intervention ($P = .018$). The value approached the initial value at the other times ($P > .05$).

In Group 2, the frequency measurements significantly increased immediately post intervention ($P < .001$) and significantly increased over the original value but decreased from the value immediately post intervention at 5 minutes ($P = .030$) and 10 minutes ($P = .029$) post intervention. It approached its original value at 15 minutes ($P = .777$). Then, it increased again at 20 minutes post intervention ($P = .046$) and thereafter showed no significant change ($P = .400$).

Table 1. Participant's Demographics at Baseline. Cold-pack applications (n = 15 each group): 10 minutes for Group 1, 12 minutes for Group 2, 15 minutes for Group 3, and 20 minutes for Group 4.

<table>
<thead>
<tr>
<th>Demographics</th>
<th>Group 1 Mean ± SD or Frequency n (%)</th>
<th>Group 2 Mean ± SD or Frequency n (%)</th>
<th>Group 3 Mean ± SD or Frequency n (%)</th>
<th>Group 4 Mean ± SD or Frequency n (%)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>21.60 ± 1.09</td>
<td>21.50 ± 1.06</td>
<td>21.50 ± 0.82</td>
<td>22.60 ± 0.51</td>
<td>.829^</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>8 (53.33)</td>
<td>7 (46.77)</td>
<td>7 (46.77)</td>
<td>8 (53.33)</td>
<td>.064</td>
</tr>
<tr>
<td>Female</td>
<td>7 (46.77)</td>
<td>8 (53.33)</td>
<td>8 (53.33)</td>
<td>7 (46.77)</td>
<td></td>
</tr>
<tr>
<td>Body mass index</td>
<td>23.00 ± 2.18</td>
<td>23.50 ± 3.41</td>
<td>23.40 ± 1.42</td>
<td>24.40 ± 1.22</td>
<td>.197</td>
</tr>
<tr>
<td>Skin thickness</td>
<td>12.60 ± 2.90</td>
<td>12.40 ± 2.80</td>
<td>13.50 ± 1.73</td>
<td>13.20 ± 1.32</td>
<td>.586</td>
</tr>
<tr>
<td>Dominant side</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>.936</td>
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<tr>
<td>Right</td>
<td>14 (93.3)</td>
<td>14 (93.3)</td>
<td>13 (86.7)</td>
<td>13 (86.7)</td>
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</tr>
<tr>
<td>Left</td>
<td>1 (6.7)</td>
<td>1 (6.7)</td>
<td>2 (23.3)</td>
<td>2 (23.3)</td>
<td></td>
</tr>
</tbody>
</table>

^One-way analysis of variance (ANOVA), chi-square test

**Abbreviations:** SD, standard deviation

at 10 minutes post intervention ($P = .018$). The value approached the initial value at the other times ($P > .05$).
Table 2. Skin temperature at Baseline and Postintervention. Cold-pack applications: 10 minutes for Group 1, 12 minutes for Group 2, 15 minutes for Group 3, and 20 minutes for Group 4.

| Thermal Camera Parameters (°C) | Group 1 Mean ± SD | Group 2 Mean ± SD | Group 3 Mean ± SD | Group 4 Mean ± SD | P Value<sup>a</sup>  
|-----------------------------|------------------|------------------|------------------|------------------|---------------  
| Baseline                   | 29.10 ± 1.69     | 28.90 ± 1.27     | 28.80 ± 1.81     | 29.30 ± 1.54     |                
| Postintervention           |                  |                  |                  |                  | <.001          
| Immediately                | 20.80 ± 2.30     | 22.30 ± 2.31     | 22.10 ± 2.52     | 22.00 ± 3.23     | <.001          
| 5 minutes                  | 24.60 ± 1.45     | 25.20 ± 1.25     | 25.20 ± 1.79     | 24.80 ± 2.12     | <.001         0.02  
| 10 minutes                 | 25.90 ± 1.30     | 26.10 ± 1.45     | 26.00 ± 1.37     | 25.20 ± 1.60     | <.001         0.001  
| 15 minutes                 | 26.70 ± 1.26     | 26.50 ± 1.40     | 26.60 ± 1.35     | 25.90 ± 1.20     | <.001         0.002  
| 20 minutes                 | 27.10 ± 1.19     | 26.90 ± 1.45     | 26.80 ± 1.28     | 26.20 ± 1.06     | <.001         0.002  
| 30 minutes                 | 27.90 ± 1.27     | 27.40 ± 1.21     | 27.20 ± 1.33     | 26.90 ± 0.89     | <.001         0.002  

<sup>a</sup>Paired sample t test, within-group comparison, compared with the before application

Abbreviations: SD, standard deviation; °C, degrees centigrade.

Table 3. Muscle Tone (Frequency) and Stiffness at Baseline and Postintervention. Cold-pack applications: 10 minutes for Group 1, 12 minutes for Group 2, 15 minutes for Group 3, and 20 minutes for Group 4.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>Group 4</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Frequency Mean ± SD</td>
<td>Stiffness Mean ± SD</td>
<td>Frequency Mean ± SD</td>
<td>Stiffness Mean ± SD</td>
</tr>
<tr>
<td>Baseline</td>
<td>12.80 ± 1.22</td>
<td>206 ± 36.20</td>
<td>12.70 ± 1.58</td>
<td>212 ± 36.50</td>
</tr>
<tr>
<td>Postintervention</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediately</td>
<td>13.10 ± 1.42</td>
<td>221 ± 35.00</td>
<td>13.40 ± 1.96</td>
<td>229 ± 39.80</td>
</tr>
<tr>
<td>5 minutes</td>
<td>12.90 ± 1.30</td>
<td>221 ± 43.70</td>
<td>12.90 ± 1.61</td>
<td>221 ± 37.70</td>
</tr>
<tr>
<td>10 minutes</td>
<td>12.30 ± 1.6</td>
<td>222 ± 40.90</td>
<td>12.90 ± 1.48</td>
<td>219 ± 36.50</td>
</tr>
<tr>
<td>15 minutes</td>
<td>12.90 ± 1.20</td>
<td>213 ± 35.90</td>
<td>13.00 ± 1.76</td>
<td>216 ± 40.09</td>
</tr>
<tr>
<td>20 minutes</td>
<td>12.80 ± 1.12</td>
<td>211 ± 41.71</td>
<td>13.10 ± 1.71</td>
<td>219 ± 41.80</td>
</tr>
<tr>
<td>30 minutes</td>
<td>12.90 ± 1.23</td>
<td>218 ± 16.0</td>
<td>12.70 ± 1.30</td>
<td>214 ± 38.10</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group 3</th>
<th>Group 4</th>
<th>Group 1</th>
<th>Group 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Frequency Mean ± SD</td>
<td>Stiffness Mean ± SD</td>
<td>Frequency Mean ± SD</td>
<td>Stiffness Mean ± SD</td>
</tr>
<tr>
<td>Baseline</td>
<td>13.40 ± 0.81</td>
<td>217 ± 20.60</td>
<td>13.50 ± 0.90</td>
<td>233 ± 23.50</td>
</tr>
<tr>
<td>Postintervention</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediately</td>
<td>13.70 ± 0.84</td>
<td>235 ± 22.10</td>
<td>13.80 ± 1.06</td>
<td>251 ± 27.20</td>
</tr>
<tr>
<td>5 minutes</td>
<td>13.40 ± 0.90</td>
<td>226 ± 22.90</td>
<td>13.50 ± 0.77</td>
<td>240 ± 24.50</td>
</tr>
<tr>
<td>10 minutes</td>
<td>13.70 ± 1.05</td>
<td>229 ± 25.60</td>
<td>13.60 ± 0.81</td>
<td>244 ± 20.10</td>
</tr>
<tr>
<td>15 minutes</td>
<td>13.50 ± 0.84</td>
<td>229 ± 19.20</td>
<td>13.70 ± 0.71</td>
<td>245 ± 23.60</td>
</tr>
<tr>
<td>20 minutes</td>
<td>13.60 ± 0.91</td>
<td>228 ± 23.40</td>
<td>13.70 ± 0.63</td>
<td>230 ± 18.60</td>
</tr>
<tr>
<td>30 minutes</td>
<td>13.50 ± 0.85</td>
<td>227 ± 38.20</td>
<td>13.20 ± 0.66</td>
<td>240 ± 21.20</td>
</tr>
</tbody>
</table>

Abbreviations: SD, standard deviation.

intervention ($P < .001$) and remained the same the prior value at 5 minutes post intervention ($P = .127$). Then it significantly increased again at 10 minutes post intervention ($P = .04$) and continued to be low at the other times ($P > .05$).

In Group 2, while the stiffness increased from baseline immediately post intervention, and at 5 and 10 minutes post intervention ($P < .001$ for all), it approached its original value in 15 minutes post intervention ($P = .96$). Then, while it increased again at 20 minutes post intervention ($P = .026$), it continued to decrease until 30 minutes post intervention ($P = .96$).

Stiffness was significantly higher at all time points in Groups 3 and 4 ($P < .05$).

No significant differences existed between any of the groups ($P = .093$ to $P = .253$, data not shown).

Elasticity. Decrement measurements evaluating muscle elasticity (Table 4) increased significantly from baseline in Groups 1 and 2 immediately post intervention—$P = .04$ and $P = .003$, respectively—and decreased at 5 minutes post intervention—$P = .181$ and $P = .577$, respectively. Then it increased again at 10 minutes post intervention—$P = .04$ and $P = .03$, respectively—and continued to decrease at the other times ($P > .05$).

In Group 3, it increased immediately post intervention ($P = .004$) and at 5 minutes post intervention ($P = .03$); then it approached its previous value at 10 minutes post intervention ($P = .190$). Then, while it increased again at 15 minutes post intervention ($P = .04$), it continued to be lower at 20 and 30 minutes post intervention ($P > .05$).
In Group 4, except for at 30 minutes post intervention (P = .29), the decrement was significantly higher at all time points (P < .05).

No significance differences existed between any of the groups (P = .06 to P = .265, data not shown).

No statistically significant relationship was found between skin temperature and the muscle's biomechanical properties (P > .05, data not shown).

**DISCUSSION**

In the current study, a decrease in skin temperature post intervention was seen for all application periods, and this decrease continued even at 30 minutes post intervention. The rectus femoris muscle became harder and less elastic as a result of cooling with the cold. Muscle tissue began to approach its baseline state at 5 minutes post intervention for a 10-minute cold-pack application and at 15 minutes post intervention for a 12-minute cold-pack application. For 15-minute and 20-minute cold-pack applications, muscle stiffness increased at all-time points.

The present study also found a decrease in skin temperature at the same level—6.6 to 7.7°C—for all cold-pack times, and this decrease continued even at 30 minutes post intervention. This finding was unlike Akgün et al's study, which found that skin temperature (13.4-16.2°C) and intramuscular temperature (3.8-5.0°C) decreased more as the application time increased. The current study's findings, however, were similar to Ring et al's, which found an average temperature decrease of 7°C, and Auley's, who reported that application of a cold pack on the skin for 15 minutes resulted in temperatures that decreased an average of 7.3°C.

In the current study, after 10, 12, and 15 minutes of cold-pack application, the muscle tone increased at 5, 15, and 5 minutes post intervention, respectively, and then returned to its baseline state. 20 minutes of cold-pack application weren't effect on muscle tone, in contrast to Mustalamphi et al.

In the stiffness parameter, an increase was observed post intervention in each group in the current study, starting at 5 minutes post intervention for Group 1 and at 15 minutes post intervention for the Group 2, and in the Group 3 and 4, the stiffness parameter was high in all post intervention time periods.

This result was similar to that of Mustalamphi et al, who investigated the mechanical properties of the muscle after a 20-minute cold-pack application. The elasticity decreased at the 10, 12, 15, and 20 minutes of cold-pack application and returned to its normal levels at 5, 15, 10, and 30 minutes, respectively.

The results of the current study in healthy individuals are parallel to most of the results for animal studies of muscle tissue, the mechanical properties of which vary due to local cooling. Some studies looking at indirect muscle parameters had similar findings, including Price and Lehmann, who found that 30 minutes of cold increased ankle-joint stiffness; Muraoka et al, who determined the same results for legs after 60 minutes of a cold-water bath; and Lakie et al, who showed that cooling the forearm in cold water caused a stiffer wrist for joint movements. Kubo et al, however, found that cooling the leg for 30 minutes with cold water didn't change the ankle-joint stiffness or the extension of the joint motion for the joint fascicle, tendon, and aponeurosis.

The current study showed that increasing the duration of a cold-pack application can prevent the biomechanical properties of the muscle from returning to the baseline level after cold application. Interestingly, this situation shows that cold-pack application time doesn't only affect the change in skin temperature but also affects intramuscular temperatures.

Mustalamphi et al, similarly to the current study, found that the skin temperature doesn't correlate with the biomechanical properties of the muscle. Although previous studies have shown that skin temperature during cold application is a weak determinant of the changes in intramuscular properties, local cooling also indicates that the skin can cause a significant temperature drop in superficial tissues, without significantly changing the temperature of tissues deeper than 2.0 cm. These different results show us that more research is needed on this subject. In addition, it is compatible with publications stating that after cold application, the biomechanical properties of the muscle deteriorate further as the application time increases, skin temperatures after cold application return to pre-application.
more easily and quickly, but the return of intramuscular temperatures is slower.\textsuperscript{5,7}

Similar to the current study, Akgün et al\textsuperscript{4} showed that the intramuscular temperature—3.8 to 5.0\textdegree{}C—decreased more as the duration of application increased from 10 to 30 min. This situation shows that it’s difficult for the intramuscular temperature, which recovers more slowly, to return the muscle’s biomechanical properties to the pre-cold-application level.

Limitations

In the current study, participants remained in a resting position following cold-pack application, and evaluations were made in that way. This experimental setup doesn’t give information about the outcome following performance of an activity. When a cold pack is applied during high-level activity in competition or training, with high activity being maintained afterward, the biomechanical effects that occur may be smaller. This is a serious limitation of the current study. This issue needs to be further explored in future studies. In addition, since the current study evaluated muscles without injury, the research team couldn’t examine the effects of cold-pack application after injury. This constitutes another limitation of the study.

CONCLUSIONS

The results of the current study showed that the rectus femoris muscle becomes stiffer and less elastic in healthy people as a result of cooling with a cold pack for various time periods. The amount of cold-pack time that minimized the biomechanical corruption of the muscle and provided cooling was 10 minutes. Careful warming up is recommended before and after intense athletic performance, and caution in cooling the skeletal muscle should be exercised.

AUTHORS’ DISCLOSURE STATEMENT

The authors have no conflict of interest to declare.

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