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# TOPICAL COOLING (ICING) DELAYS RECOVERY FROM ECCENTRIC EXERCISE-INDUCED MUSCLE DAMAGE

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TE-CHIH LIU,<sup>2</sup> CHENG-HSIU LAI,<sup>2</sup> M. BRENNAN HARRIS,<sup>5</sup> AND CHIA-HUA KUO<sup>1,3</sup>

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## ABSTRACT

Tseng, C-Y, Lee, J-P, Tsai, Y-S, Lee, S-D, Kao, C-L, Liu, T-C, Lai, C-S, Harris, MB, and Kuo, C-H. Topical Cooling (Icing) Delays Recovery From Eccentric Exercise-Induced Muscle Damage. *J Strength Cond Res* 27(5): 1354–1361, 2013—It is generally thought that topical cooling can interfere with blood perfusion and may have positive effects on recovery from a traumatic challenge. This study examined the influence of topical cooling on muscle damage markers and hemodynamic changes during recovery from eccentric exercise. Eleven male subjects (age 20.2 ± 0.3 years) performed 6 sets of elbow extension at 85% maximum voluntary load and randomly assigned to topical cooling or sham groups during recovery in a randomized crossover fashion. Cold packs were applied to exercised muscle for 15 minutes at 0, 3, 24, 48, and 72 hours after exercise. The exercise significantly elevated circulating creatine kinase-MB isoform (CK-MB) and myoglobin levels. Unexpectedly, greater elevations in circulating CK-MB and myoglobin above the control level were noted in the cooling trial during 48–72 hours of the post-exercise recovery period. Subjective fatigue feeling was greater at 72 hours after topical cooling compared with controls. Removal of the cold pack also led to a protracted rebound in muscle hemoglobin concentration compared with controls. Measures of interleukin (IL)-8, IL-10, IL-1β, and muscle strength during recovery were not influenced by cooling. A peak shift in IL-12p70 was noted during recovery with topical cooling. These data suggest that topical cooling, a commonly used clinical intervention, seems to not improve but rather delay recovery from eccentric exercise-induced muscle damage.

**KEY WORDS** ice pack, inflammatory cytokines, creatine kinase, myoglobin, baseball players, muscle injury

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## INTRODUCTION

In clinical practice, topical cooling with a cold pack is a frequently used therapeutic modality aimed to reduce acute pain and inflammation of injured tissues (30). However, it is currently unknown whether this treatment is suitable for improving recovery after eccentric exercise-induced muscle damage as a result of strength training.

Lowering tissue temperature during the cooling period can reduce peripheral blood perfusion (5,7). However, blood perfusion may return after removal of the treatment. This hemodynamic fluctuation may influence the magnitude of muscle damage, healing rate, and the associated fatigue feelings during recovery (32). Tissue damage caused by such fluctuations in blood perfusion has been well demonstrated in the ischemic reperfusion model that can elicit acute inflammation (22). In addition, this change profoundly alters circulating cytokine levels (21,35). Although temperature shifts can influence tissue blood perfusion (16,17), no investigation has focused on the moderate change in tissue perfusion after topical cooling and the influence of this condition on recovery rates after eccentric exercise-induced muscle damage.

Inflammation is an acute physiological response required for tissue healing after tissue damage (28,37) and is orchestrated by a series of cytokines released by the surrounding tissues and immune cells (8,36,37). Previous studies on exercise-induced changes in inflammatory cytokines have produced contrasting results (26,27). For example, eccentric treadmill exercise has been shown to increase production of proinflammatory cytokines including interleukin (IL)-8 and tumor necrosis factor-alpha (TNF-α) (4). However, another study reported no change or reduced concentrations of these cytokines after eccentric-induced muscle damage, in addition to no change in the anti-inflammatory cytokine IL-10 (15). In contrast, Nieman et al. (24) found elevated IL-10 after ultramarathon exercise, a type of exercise known to induce significant muscle damage. Some cytokines are responsible for the regulation of the proliferation,

self-renewal, and differentiation capacity of uncommitted progenitor cells during tissue healing process. Both IL-12p70 and IL-6 act synergistically in supporting proliferation of human primitive hemopoietic progenitors (12). Together with satellite cells, marrow-derived hemopoietic stem cells migrate into areas of degenerative muscle fibers and take part in the regeneration of the damaged fibers (9). However, it has been reported that IL-12p70 decreases after a 30-minute cycling at 80%  $\dot{V}O_{2,max}$  (29), whereas the response of IL-12p70 against eccentric contraction-induced muscle damage has not yet been reported. Very few studies have examined the effect of topical cooling on inflammatory cytokines and muscle damage during the recovery from eccentric exercise-induced muscle damage. Topical cooling has been shown to result in significantly greater decreases in the pro-inflammatory mediator IL-1 $\beta$  during the first hour of recovery after sprints, but no significant changes in IL-6 and IL-10 levels were detected (23). The time period measured during recovery and the mode of exercise can lead to different results in inflammatory cytokine response. A time course study design would provide better insight into the role of cytokines in the healing process after eccentric exercise-induced muscle damage.

The current study was undertaken to determine whether topical cooling can improve recovery in eccentric contraction-induced muscle damage. We hypothesized that topical cooling can change the rate of recovery after eccentric contraction-induced muscle damage, and this change is associated with the fluctuation in blood perfusion in exercised skeletal muscle and the inflammatory cytokine response. The biomarkers of muscle damage and inflammatory cytokine profile were monitored during a 72-hour recovery period after an acute bout of eccentric exercise.

## METHODS

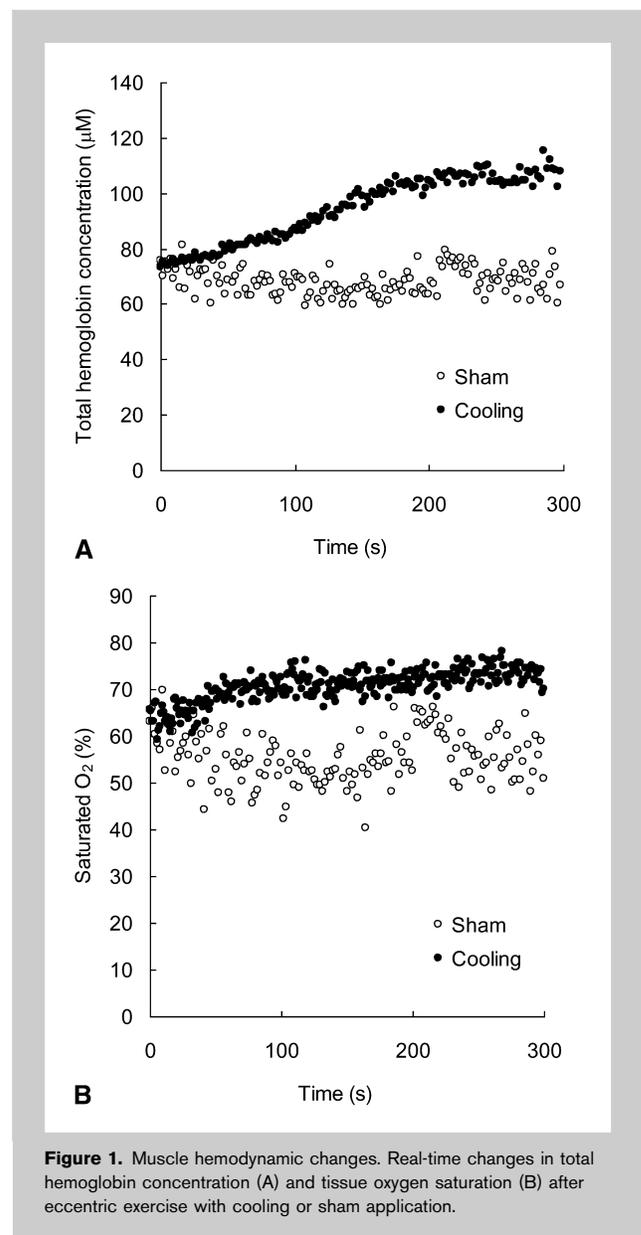
### Experimental Approach to the Problem

This study was designed to ask the question (a) whether topical cooling can affect the rate of recovery after eccentric exercise-induced muscle damage and (b) whether this change is associated with the fluctuation in muscle blood perfusion and changes in inflammatory cytokine response. To achieve these aims, subjects were balanced in number and randomly assigned into 2 trials: sham application (Sham) and topical cooling with cold pack (Cooling) in a 2-period crossover design with 4-week separation between trials. Five sessions of 15-minute cold pack application to the exercised muscle were conducted at 0, 3, 24, 48, and 72 hours after eccentric exercise-induced muscle damage. The subjects used in this study were collegiate baseball players and the long-term activity of the elbow in these athletes rules out any protective effect of training and demonstrates the usefulness of the eccentric exercise as being different enough to cause soreness and damage. In addition, because of the high rate of injury in these athletes, it is important to investigate methods of increasing the strength of the elbow.

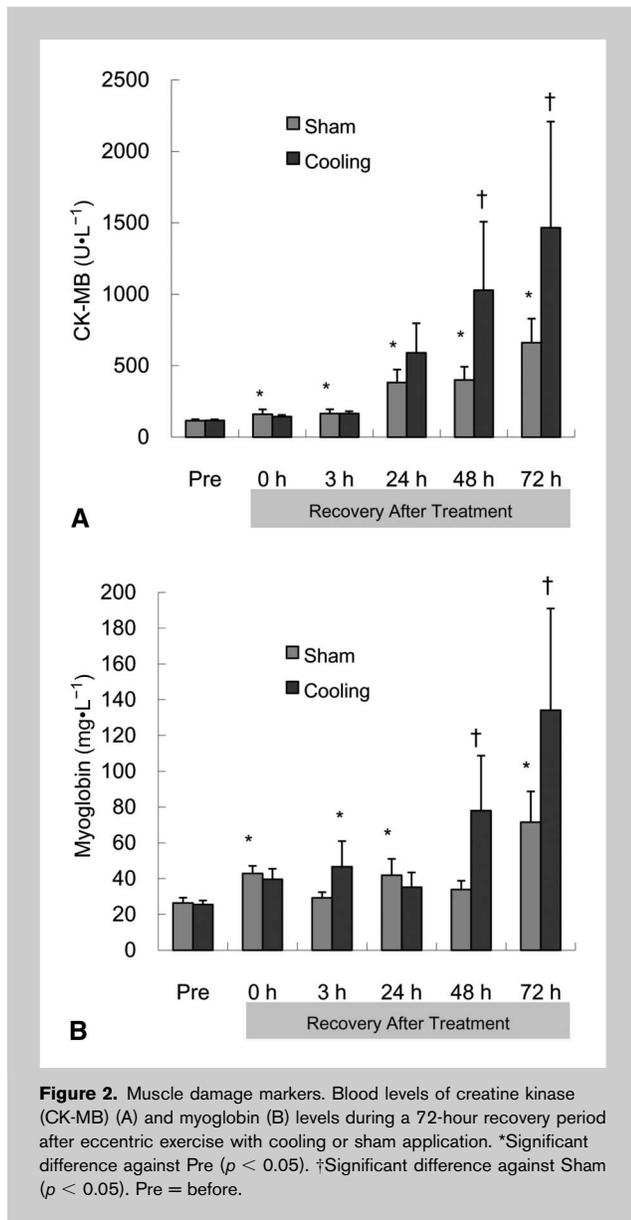
Although this study focuses on the acute markers of muscle damage and soreness, a broader future study examining the use of eccentric exercise to prevent future injury while demonstrating that injury prevention is diminished by topical cooling would provide the most direct clinical application of our findings.

### Subjects

Eleven male college baseball players were recruited ( $20.2 \pm 0.3$  years old,  $175.6 \pm 1.6$  cm tall, and  $69.5 \pm 3.1$  kg body weight) to voluntarily participate in this study. All subjects played in the National University Baseball League. The study was performed during off-season in 2010. None of the subjects performed weight training or any other form of exercise training for at least 1 week before the



**Figure 1.** Muscle hemodynamic changes. Real-time changes in total hemoglobin concentration (A) and tissue oxygen saturation (B) after eccentric exercise with cooling or sham application.



experimental trials. Participants with musculoskeletal disorders were excluded. This study was approved by the Taipei Physical Education College Institutional Review Board. Subjects signed an informed consent after an explanation of the experimental procedures and possible risks involved in this study.

#### Procedure

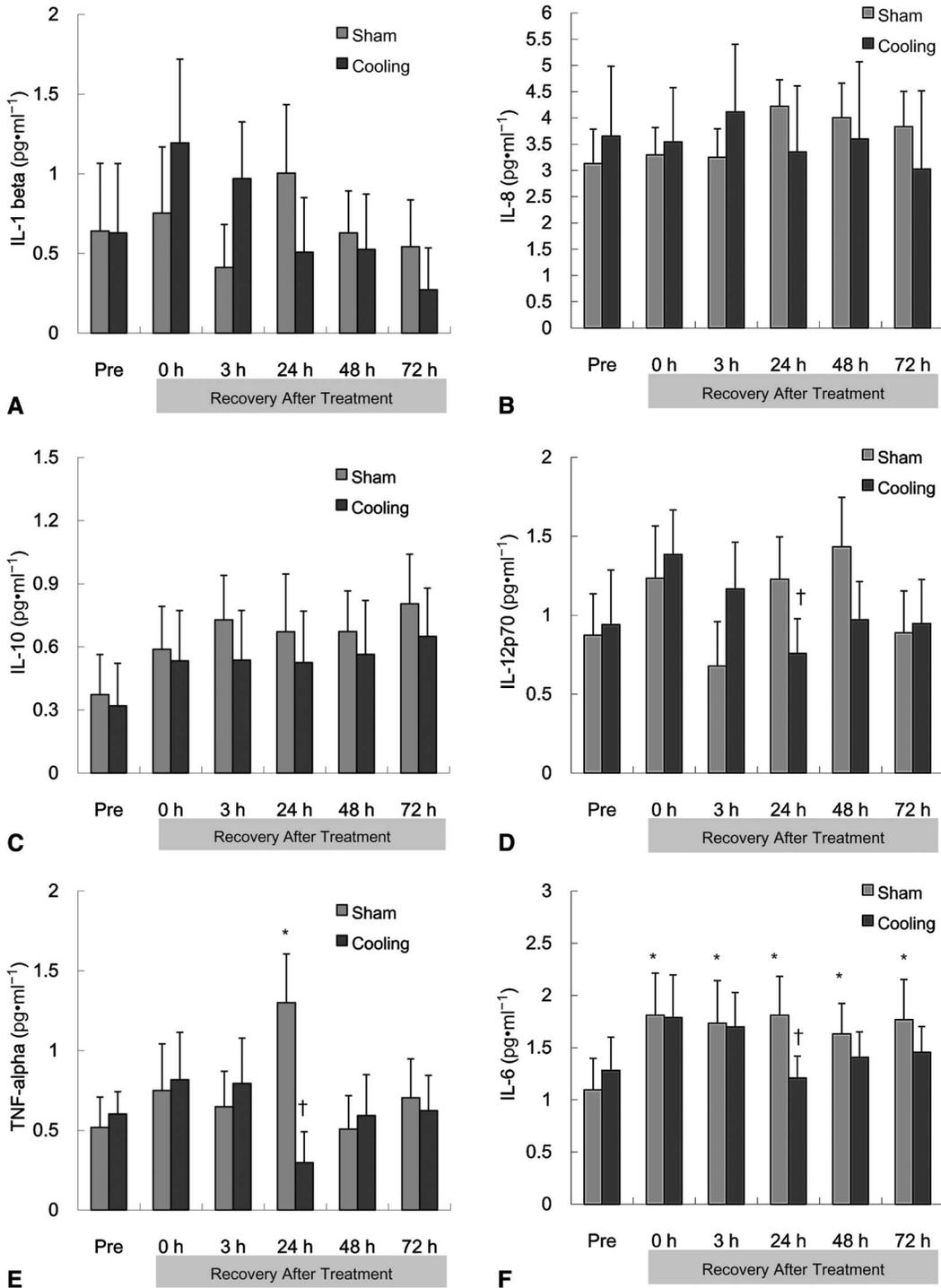
**Eccentric Exercise Protocol.** Eccentric exercise protocol was performed at 0900 in the morning under an overnight-fasted condition with ad libitum water intake. One-repetition maximum (1-RM) concentric elbow flexion strength for all subjects was determined before eccentric exercise challenge. They performed stretching and a warm-up on a cycloergometer for ~5 minutes before the determination of maximal strength. Five maximal trials separated by 3 minutes of rest

were used to estimate 1-RM for each individual. The exercise protocol used to elicit skeletal muscle damage consisted of 6 sets of 5 eccentric contractions with 2-minute rest between sets (15). The weight of barbell was set at 85% of their maximal strength according to Spiering et al. (31). All subjects performed 2 bouts of eccentric exercise of the elbow flexors using both arms. During the exercise, subjects were seated on a bench with both arms positioned in front of a supported pad, and the forearms were supinated throughout the range of motion while holding a barbell. Participants were requested to lower the barbell from full elbow flexion to a completely extended position (180°) in 7 seconds under the supervision to maintain a constant movement velocity. After each eccentric movement, the arm was assisted back to the flexed position by research assistants to eliminate any concentric actions.

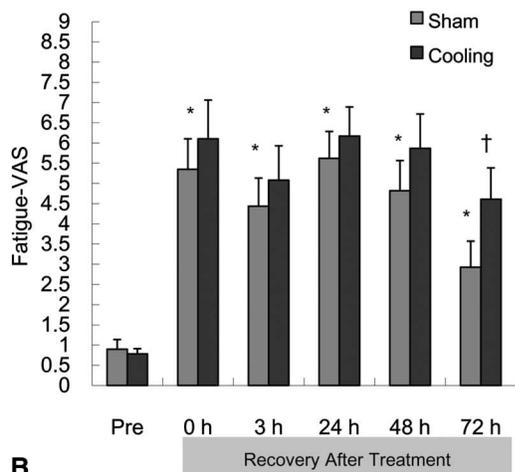
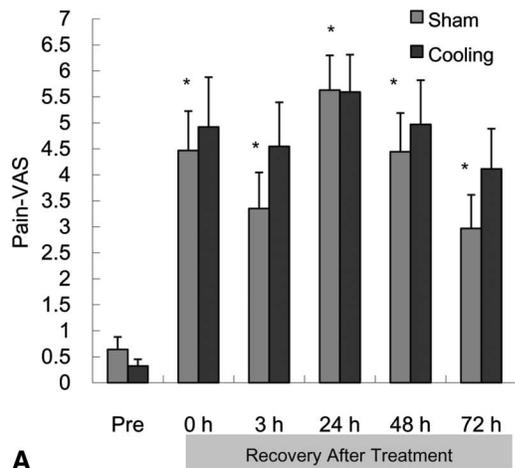
Muscle hemodynamic changes, muscle damage markers, inflammatory cytokines, subjective pain level, and isometric muscle strength were assessed before (pre) and after (post) the above eccentric exercise protocol. Blood samples were collected from the antecubital vein right after each single topical cooling during the recovery period.

**Muscle Hemodynamics.** Local blood perfusion is tracked by tissue hemoglobin concentration in exercised muscle which was detected by near-infrared spectroscopy (NIRS) (Oxiplex TS; ISS, Champaign, IL, USA) (1). Near-infrared spectroscopy is a well-accepted method to estimate regional blood flow and oxygenation in tissues (13,20). While in the supine position, the sensor for NIRS was applied over the biceps area of the arm to determine the total hemoglobin concentration (THC). In the study, NIRS measurements were performed using a frequency-domain tissue spectrometer, which have 2 laser diodes for measurement of light absorbance with wavelengths of 690 and 830 nm. The optodes were placed over the targeted muscles with the sampling rate of 1·per second. Near-infrared spectroscopy enables noninvasive continuous measurement of changes in the concentration of oxygenated hemoglobin (HbO<sub>2</sub>) and deoxygenated hemoglobin (deoxyHb). The tissue THC was calculated by the sum of HbO<sub>2</sub> and deoxyHb concentrations, which is used to indicate blood distribution.

**Inflammatory Cytokines.** Cytokines including IL-1 $\beta$ , IL-6, IL-8, IL-10, IL12p70, and TNF- $\alpha$  were analyzed in duplicate using a multiplex bead kits (Human Inflammation Kit, BD Cytometric Bead Array; Becton Dickinson and Company, Franklin Lakes, NJ, USA). Each multiplex assay was performed according to the manufacturers' instruction. Raw data (fluorescent intensity) were detected by FACSCalibur (Becton Dickinson and Company) to obtain concentration values based on a standard curve showing linear relationship between known cytokine concentration and fluorescent values. Standard curves for each cytokine were generated by using the reference cytokine concentrations supplied by the manufacturer.



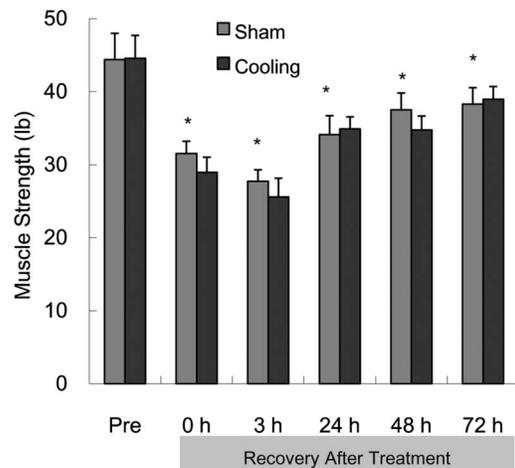
**Figure 3.** Inflammatory cytokines. Blood levels of interleukin (IL)-1 beta (A), IL-8 (B), IL-10 (C), IL-12p70 (D), tumor necrosis factor (TNF)-alpha (E), and IL-6 (F) during a 72-hour recovery period after eccentric exercise with cooling or sham application. \*Significant difference against Pre ( $p < 0.05$ ). †Significant difference against Sham ( $p < 0.05$ ). Pre = before.



**Figure 4.** Subjective pain and fatigue feelings. Self-report of visual analog scale (VAS) of pain (A) and fatigue (B) feelings during a 72-hour recovery period after eccentric exercise with cooling or sham application. \*Significant difference against Pre ( $p < 0.05$ ). †Significant difference against Sham ( $p < 0.05$ ). Pre = before.

**Muscle Damage Markers.** Two muscle damage markers CK-MB and myoglobin were measured to confirm the results of muscle damage. Blood samples for the biochemical analyses were kept on ice for 30 minutes before they were centrifuged at 3,000 rpm for 10 minutes to obtain serum. All blood samples were stored at  $-80^{\circ}\text{C}$  before analysis. For the quantitative measurements of serum myoglobin and CK-MB levels, a commercially available chemiluminescent enzyme labeled immunometric assay was used on an autoanalyser (IMMULITE; Diagnostic Products Corporation, Los Angeles, CA, USA).

**Visual Analog Scale for Pain and Fatigue.** A subjective rating of the perceived muscle pain and fatigue was determined using a visual analog scale (VAS). While sitting, the participants were asked to rate the amount of pain or fatigue experienced within



**Figure 5.** Isometric muscle strength. A 72-hour recovery period after eccentric exercise with cooling or sham application. \*Significant difference against Pre ( $p < 0.05$ ). †Significant difference against Sham ( $p < 0.05$ ). Pre = before.

their biceps during active flexion and extension of the elbow joint on a scale from 0 to 10, where 0 corresponded to “no pain/fatigue” and 10 was equated to “very painful/fatigue” (3).

**Muscle Isometric Strength.** Maximal isometric strength of the arm was assessed using a hand-held dynamometer (micro-FET; Hoggan, West Jordan, Draper, UT, USA). Each participant was seated in an upright position with their shoulder held in a neutral position, elbow at  $90^{\circ}$  flexion, and wrist supinated against fixed resistance. Participants were asked to perform 3 maximal isometric contractions for 3 seconds each within 30 seconds between contractions. The maximal value achieved during these 3 repetitions was selected as the isometric muscle strength for all individuals.

**Statistical Analyses**

To compare the mean values of all variables between cooling and sham treatments during a 72-hour recovery, a two-way analysis of variance with repeated measure was used. Bonferroni post hoc test was used to distinguish the significant difference between pair of groups. For the probability of Type I error, significance was set at  $p < 0.05$ . All values were expressed as mean  $\pm$  SE. The intraclass correlation coefficient of test-retest reliability were as follows: CK-BM, 0.97; myoglobin, 0.97; maximal isometric strength, 0.86; VAS for fatigue, 0.85; VAS for pain, 0.80; cytokine assay, 0.93–0.97; and NIRS measurements, 0.82.

**RESULTS**

Immediately after topical cooling, real-time hemodynamic changes were monitored for 300 seconds by tracing hemoglobin in exercised muscle with NIRS measurements.

Topical cooling elicited rapid and sustained elevations in THC (Figure 1A) and muscle tissue oxygen saturation (Figure 1B) above the control level.

Circulating levels of CK-MB (Figure 2A) and myoglobin (Figure 2B), biomarkers of muscle damage, increased immediately, 3, 24, 48, and 72 hours after eccentric exercise in both the control and cooling trials ( $p < 0.05$ ). The greatest CK-MB and myoglobin levels were noted during 48 and 72 hours post-exercise. Topical cooling resulted in significantly ( $p < 0.05$ ) higher circulating CK-MB and myoglobin than control at the 48 and 72 hours post-exercise time points.

No significant change in the levels of IL-1 $\beta$  (Figure 3A), IL-8 (Figure 3B), and IL-10 (Figure 3C) were observed after the muscle-damaging eccentric exercise in either the control or topical cooling conditions and no differences in these cytokines were found between the control and cooling trials throughout the 72-hour observation period. Interleukin-12p70 (Figure 3D), TNF- $\alpha$  (Figure 3E), and IL-6 (Figure 3F) were significantly ( $p < 0.05$ ) lower with topical cooling applied post-exercise compared with the control trial at 24 hours post-exercise ( $p < 0.05$ ). No significant differences in these cytokines were observed between the 2 conditions at the other time points. Interleukin-6 was significantly increased at 0, 3, 24, 48, and 72 hours post-exercise in the control trial but not as much after topical cooling.

Increased VAS pain scores were noted at 0, 3, 24, 48, and 72 hours after eccentric exercise for both the control and cooling trials (Figure 4A) ( $p < 0.05$ ). No significant differences in the subjective pain or muscle strength were noted between the control and cooling trials during the 72-hour recovery period. However, topical cooling significantly ( $p < 0.05$ ) increased subjective ratings of fatigue above the control level at 72-hour post-exercise (Figure 4B). Eccentric exercise significantly ( $p < 0.05$ ) decreased the muscle strength of subjects, which was not completely recovered 72 hours after exercise in both the control and cooling trials (Figure 5). No significant difference in muscle strength during recovery was found between the control and cooling trials.

## DISCUSSIONS

Tissue damage can occur when blood supply returns to the tissue after a period of ischemia (21). In this study, an immediate elevation of blood perfusion in the exercised muscle was observed after a period of cold pack treatment. In line with this hemodynamic fluctuation, an increased level of muscle damage markers (CK and myoglobin) after an acute bout of eccentric exercise was manifested. Furthermore, the subjective measures of pain and fatigue were somewhat elevated with topical cooling treatment. Topical cooling caused either no change (IL-1 $\beta$ , IL-8, and IL-10) or a decrease (TNF- $\alpha$  and IL-6) in the release of inflammatory cytokines after eccentric exercise-induced muscle damage. The peak of IL-12p70 response was shifted to the right during recovery. Collectively, our results provide evidence that this commonly used clinical intervention for traumatic injury seems

to delay the recovery from eccentric exercise-induced muscle damage.

Inflammation plays an important role in the adaptation of skeletal muscle after exercise challenge (38). It seems that during the early period of inflammation, damaged muscle cells are eliminated by phagocytosis of macrophages (39). Subsequently, muscle cell regeneration takes place by recruiting stem cells from surrounding tissues (9,11,36). The early response can last for a week (10,25), in parallel with elevated circulating levels of CK-MB and myoglobin during the regeneration phase. Alteration in the phagocytosis rate would be expected to affect the timing of muscle regeneration. The result of greater increases in exercise-induced CK-MB and myoglobin levels between 48- and 72-hour post-exercise and peak shift in inflammatory cytokine IL-12p70 may reflect a change in the time course of cell turnover and muscle regeneration by topical cooling. Tumor necrosis factor- $\alpha$  is known as an inducer for apoptosis of damaged or unhealthy cells (38). Interleukin-6 and IL-12p70 are essential for stem cell recruitment for tissue regeneration after apoptosis and phagocytosis (11,12,38). In the cooling trial, the attenuated IL-6 and TNF- $\alpha$  responses and peak shift in IL-12p70 after eccentric exercise may be partly involved with the slower rate of cell turnover and subsequent greater increases in muscle damage marker observed during 48–72 hours of recovery.

Interleukin-8 is a cytokine that recruits neutrophils to the site of inflammation for phagocytosis, which plays an important role for the early step of damaged cell removal. Similar to the previous reports (14,33), no significant change in IL-8 levels after exercise was observed. However, not all studies have generated similar results. Hirose et al. (15) reported a decreased IL-8 after elbow flexor eccentric exercise, and IL-8 response against eccentric exercise was decreasing with exercise experience. In this study, both dominant and nondominant arms were recruited, whereas in Hirose's study, only dominant arm was recruited during exercise. Thus, different results among studies may be associated with the use of dominant and nondominant arms during exercise.

In addition to the ratings of pain, a greater subjective rating of fatigue in the topical cooling trial compared with the control trial at 72 hours was found during the recovery period without affecting muscle strength. The topical cooling used in this study results in differential findings compared with cryotherapy (ice water immersion) that shows no significant improvement in power but an increase in isokinetic muscular endurance for patients with the history of lower extremity injury (19). Topical cooling has been found to enhance the recovery of muscle strength after electrical-stimulated muscle damage (19). However, we did not find similar results in the current muscle damage model. One possible explanation for the divergent results could be the differences in subject characteristics and model use. Elite baseball players may have less fluctuation in muscle strength than others.

Although the IL-6 signaling cascade has been suggested to be related to perceived pain (34), the results from our study do not provide sufficient support for the association between IL-6 and eccentric exercise-induced pain. Immediately after exercise, subjective ratings of pain were dramatically elevated but the surge in IL-6 did not occur until later in recovery. Furthermore, increased IL-6 concentrations were attenuated 24 hours after topical cooling during post-exercise recovery, whereas subjective pain was persistent. Cytokine-induced pain is associated with neuropathic pain, suggested by the fact that inhibition of IL-6 action prevented neuropathic pain (18). In this study, topical cooling was unable to relieve but seems to increase pain after the eccentric exercise-induced muscle damage. However, topical cooling has been shown to effectively relieve pain after surgery (2) and acute traumatic tissue injury (6). Therefore, we speculate that pain induced by muscle damaging exercise is mechanistically different from other types of pain.

We must note that this study does not provide evidence on whether recovery from pitching-induced muscle damage would be slowed down by topical cooling. In baseball players, repeated pitching is the main cause for the accumulation of microtrauma in the muscles that decelerate the shoulder. The result of this study encourages future investigations looking specifically at whether topical cooling deteriorates recovery from functionally relevant activities such as pitching. In addition, longer-term studies examining injury prevention and performance would add to the clinical relevance of our observations.

### PRACTICAL APPLICATIONS

As it stands, our results provide evidence that topical cooling does not enhance and seems to delay the return to normal of muscle damage markers and subjective fatigue feeling after eccentric exercise. The surge in tissue oxygenation after removal of the cooling application may be part of the mechanisms involved in the delayed recovery. Collectively, these results indicate that intervention with topical cooling disrupts the normal adaptive responses to exercise.

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