

# Effect of cold water immersion on muscle damage indexes after simulated soccer training in young soccer players

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## Summary

*Study aim:* To investigate the effect of cold water immersion (CWI) on muscle damage indexes after simulated soccer activity in young soccer players.

*Material and methods:* Eighteen professional male soccer players were randomly divided into two groups: CWI (n = 10, age 19.3 ± 0.5, body mass index 22.2 ± 1.3) and control (n = 8, age 19.4 ± 0.8, body mass index 21.7 ± 1.5). Both groups performed a simulated 90-minute soccer-specific aerobic field test (SAFT<sup>90</sup>). Then, the CWI group subjects immersed themselves for 10 minutes in 8°C water, while the control group subjects sat passively for the same time period. Blood samples were taken before, immediately after, 10 minutes, 24 hours and 48 hours after the training session in a fasted state. Blood lactate, creatine kinase (CK) and lactate dehydrogenase (LDH) enzyme levels were measured.

*Results:* Lactate, CK and LDH levels increased significantly after training (p < 0.001). There were significant interactions between groups and subsequent measurements for CK (p = 0.0012) and LDH (p = 0.0471). There was no significant difference in lactate level between the two groups at any aforementioned time.

*Conclusion:* It seems that CWI after simulated 90-minute soccer training can reduce the values of muscle damage indexes in soccer players.

**Keywords:** Cold water immersion – Recovery – Lactate – CK – Soccer

## Introduction

Athletes often must participate in competitions or training sessions several times over a relatively short period of time. A common concern for athletes is full physiological recovery, which can affect their performance and training efficiency. Residual fatigue from consecutive matches has an adverse effect on the performance [8]. Soccer players' activities during the competition season include one-week cycles, each consisting of training, tapering, competition and recovery. Furthermore, at elite levels, some players frequently encounter additional commitments, such as national tournaments, knockout matches or even national team camps. All of these competitions can put pressure on the different physiological systems in a particular muscular system [30].

Exercise-induced muscle damage (EIMD) is an ephemeral circumstance caused by unaccustomed strenuous exercise. Delayed onset muscle soreness (DOMS), topical

swelling, and increased levels of intramuscular enzyme such as creatine kinase (CK) and lactate dehydrogenase (LDH) are indirect indicators of exercise-induced muscle damage (EIMD). Morphological changes including disturbances of sarcomeres, cytoskeletal components, and sarcolemma are some intracellular events that often occur simultaneously with muscle damage. Furthermore, reduction in muscle operation such as lower capability of force production, which is another indirect marker of muscle damage, usually occurs after DOMS and leakage of muscle proteins such as CK into the bloodstream [9, 25]. Therefore, performing some post-workout procedures in order to relieve these restrictive conditions can be crucial in order to prepare athletes for the next match as soon as possible.

There are some recovery methods such as stretching, massage, muscle compression by garments, anti-inflammatory drugs, antioxidants, and CWI [6, 12]. The beneficial effects of CWI on DOMS, EIMD and inflammatory markers have been demonstrated by some previous

studies. For example, Ascensão et al. compared the effects of immersion in cold (10°C) and warm (35°C) water after a soccer match in male soccer players. They reported a decrease in CK, myoglobin, and C-reactive protein with CWI [4]. In another study, Missau et al. concluded that applying CWI (15°C) reduces the inflammatory response and DOMS in untrained individuals undergoing resistance exercise [24]. Moreover, Alshoweir et al. reported that CWI (12–13°C) has positive effects on alleviation of DOMS, reducing immediate muscle tenderness, improving performance and reducing pain in rugby players after intense eccentric exercise [2]. However, some studies did not provide supporting evidence. For example, Rowsell et al. found that immediate post-match CWI (10°C) does not alter physical performance or indices of muscle damage and inflammation but does diminish the perception of general fatigue and leg soreness between matches in junior male soccer players [27]. Sellwood et al. stated that CWI (5°C) had no positive impact on most pain parameters, tenderness, isometric strength, swelling, or serum CK after an eccentric loading protocol in untrained volunteers [29]. Also Nardi et al. found that CWI (15°C) did not induce modifications of inflammatory and hematological markers after a training session in young soccer players [11]. Dantas et al. reported that CWI (10 min in 10°C) did not cause any improvement in CK decrement and pain after running 10 km [10]. Interestingly, Vieira et al. concluded that water immersion at warmer temperature (15°C) may be more effective than colder temperatures (5°C) in promoting recovery from strenuous exercise [35].

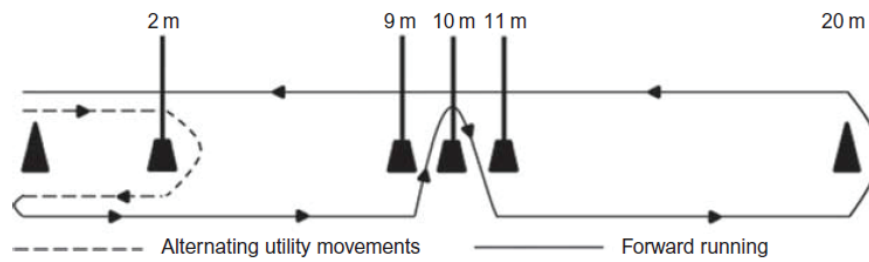
Findings from diverse studies that have investigated the effect of CWI on the muscle damage and fatigue indices are conflicting and contradictory. Moreover, although a large number of investigations have been carried out in this field, studies designed specifically for soccer are rare. Some of these studies used the Loughborough Intermittent Shuttle Test (LIST) for the purpose of imitating a soccer match and CWI in order to mitigate muscle damage. But it should be noted that CWI application in these studies was various in terms of time and temperature. Leeder et al. performed two studies and used 14 min exposure in 14°C water in both [19, 20]. Bailey et al. and Bouzid et al. applied 10 min immersion in 10°C water, but it should be mentioned that in the research by Bouzid et al. they compared thermoneutral versus cold water immersion and there was no control group, so interpreting the results of that study to compare the CWI effects exclusively might not be sensible [5, 7]. Since no positive effect corresponding with muscle damage has been reported in the examinations (except one case in one stage of blood sampling [19]), variables related to the time and temperature have been modified to determine the probable effects in a novel condition. Hence water temperature and immersion time have been adjusted to 8°C and 10 min. Consequently, the

present study aimed to evaluate the effect of CWI (10 min in 8°C) on muscle damage indices after simulated soccer training in young soccer players.

## Material and methods

Eighteen male professional soccer players of a soccer club who were participating in the second division of a professional soccer league in Iran volunteered for this study. All the subjects had been attending team training sessions 6 times per week and all the soccer positions except goalkeepers were included. Prior to testing and after explanations of the study design and potential risks, all the subjects signed informed consent and completed a health-scanning questionnaire. Also the ethics committee of the university for testing of human subjects confirmed the study's compatibility with the Helsinki Declaration. The participants were controlled from 48 hours in advance of the start of experimental period and they did not do any considerable physical activity as training session throughout the period, other than SAFT<sup>90</sup>. In addition, they were asked to abstain from consuming and applying any ameliorative interventions, such as anti-inflammatory drugs, compression garments, or alcoholic and caffeinated liquids. Furthermore, they were required to maintain their ordinary dietary habits until the end of study. Subjects were randomly divided into CWI (n = 10, age 19.3 ± 0.5, body mass index 22.2 ± 1.3) and control (n = 8, age 19.4 ± 0.8, body mass index 21.7 ± 1.5) groups. After 10 hours of fasting, the first blood specimens were taken at 9:00 a.m. Then, after two days all subjects performed SAFT<sup>90</sup>. Before starting the exercise protocol players did 15 minutes of general and 10 minutes of specialized warm up. The second blood sampling was performed after SAFT<sup>90</sup>. Then, subjects of the CWI group immersed their lower body (from the hip to the toes) in cold water at 8°C for 10 minutes in a sitting position. A thermometer was monitoring the temperature of the water continuously and the temperature was maintained by adding or removing ice. On the other hand, the control group sat passively during the same period; they performed no stretching, walking or other physical activity. Immediately after CWI, the third stage of measurement was performed. When all the subjects were in a fasted state as well as first time sampling, the fourth and fifth phases of sampling were also done at 10:00 a.m. 24 h and 48 h after the exercise protocol, respectively

The soccer-specific aerobic field test (SAFT<sup>90</sup>) [22, 31] is a simulated soccer match protocol in which players perform different running patterns and figures that frequently take place in a regular soccer game, namely acceleration, deceleration, reverse and side running, and the side-step cutting maneuver. The test (SAFT<sup>90</sup>) was performed on



**Fig. 1.** SAFT<sup>90</sup> Protocol scheme. The path taken by the subjects. [22]

a 20-m track (Fig. 1). It was 90 minutes in total and was divided into two sections (half) by a 15-minute passive break. During the rest subjects were allowed to drink water ad libitum. The audio file in each half repeated a 15-minute voice three times consecutively. The pre-recorded sound gave instructions to the participants and they had to adjust their movements according to the signals. Overall, the test involved different speeds including standing ( $0 \text{ km} \cdot \text{h}^{-1}$ ), walking ( $4.00 \text{ km} \cdot \text{h}^{-1}$ ), jogging ( $10.3 \text{ km} \cdot \text{h}^{-1}$ ), gentle running ( $15.00 \text{ km} \cdot \text{h}^{-1}$ ), and sprinting ( $20.4 \leq \text{km} \cdot \text{h}^{-1}$ ). The players covered a distance of 10.7 kilometers totally with 1296 changes in speed and 1350 changes of direction.

### Biochemical assays

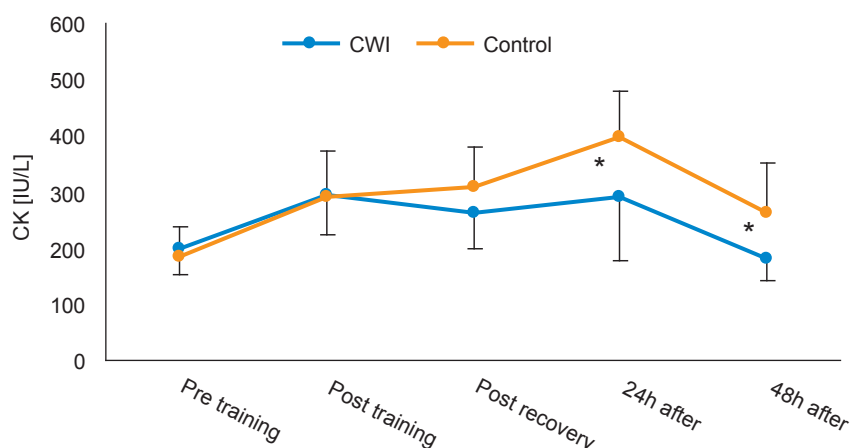
Blood was centrifuged at  $3000 \text{ rev} \cdot \text{min}^{-1}$  for 10 min for serum separation. Serum CK was assessed by a spectrophotometric method using a commercial kit (Pars Azmun Co., Iran, coefficient of variation 1.6%, accuracy 1.0 IU/L). Also serum LDH was measured by the enzymatic colorimetric method and using a commercial kit (Pars Azmun Co., Iran, coefficient of variation 2.1%, accuracy 1.0 IU/L). Furthermore, plasma lactate was evaluated by the photometric method using a commercial kit (Pars Azmun Co., Iran, variation coefficient 1.16%, accuracy 1.0 IU/L).

### Statistical analysis

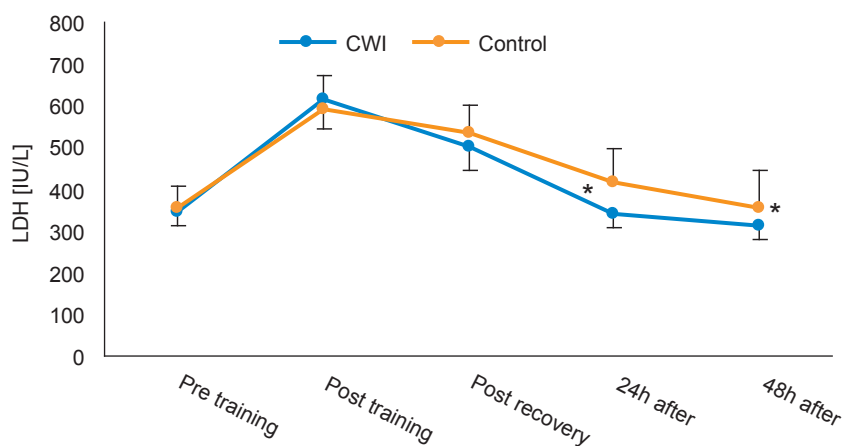
The data are expressed as mean ( $X$ )  $\pm$  standard deviation (SD). The Shapiro-Wilk test was used to calculate data distribution. CK was not normally distributed in the post-recovery period and 48 h after. We used log transformation for all CK measurements. Repeated measure analysis of variance (mixed design) was used for assessment of the interaction effect between the two groups over time. Due to lack of sphericity we used G-G correction. For detailed comparisons we used the least significant difference test (LSD) with Bonferroni correction ( $p$  values multiplied by 5 – as we have 5 comparisons). The significance level was considered at  $\alpha = 0.05$ .

### Results

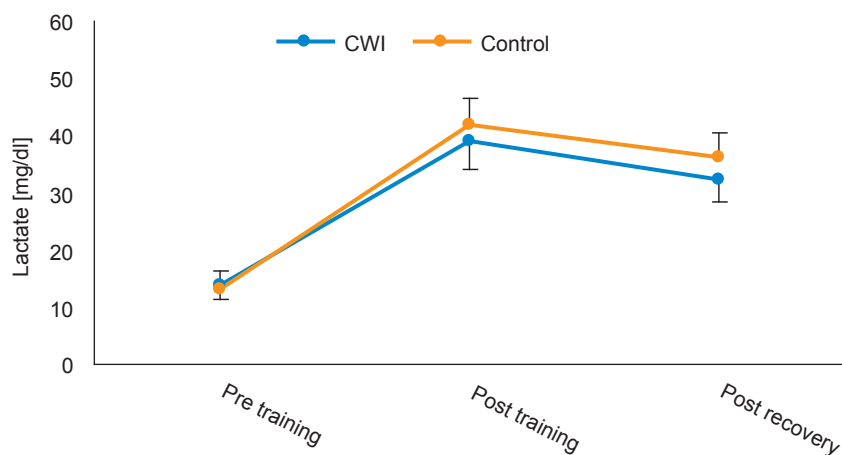
Lactate, CK and LDH levels increased significantly after training ( $p < 0.001$ ). There were significant interactions between groups and subsequent measurements for CK ( $F_{4,64} = 6.64$ ,  $p = 0.0012$  and  $\eta^2 = 0.293$ ) and for LDH ( $F_{4,64} = 2.86$ ,  $p = 0.04712$ ,  $\eta^2 = 0.152$ ), which means that groups responded in a different way. The LSD post-hoc test (with Bonferroni correction) indicated significant



**Fig. 2.** Cratine Kinase (CK) levels over time in cool water immersion (CWI) and control groups  
\* – significant differences between two groups ( $p < 0.05$ ).



**Fig. 3.** Lactate Dehydrogenase (LDH) levels over time in cool water immersion (CWI) and control groups  
\* – significant differences between two groups ( $p < 0.05$ ).



**Fig. 4.** Lactate levels over time in cool water immersion (CWI) and control groups  
\* – significant differences between two groups ( $p < 0.05$ ).

difference between the groups at 24 ( $p = 0.031$ ) and 48 h ( $p = 0.045$ ) after exercise in CK and at 24 h ( $p = 0.015$ ) in LDH (Figs 2 and 3). There was no significant interaction between groups and subsequent measurements for lactate ( $F_{4,64} = 2.74$ ,  $p = 0.0640$ ,  $\eta^2 = 0.146$ ) (Fig. 4).

## Discussion

The results of this study showed that CK, LDH and lactate increased after SAFT<sup>90</sup> in accordance with other analogous studies [1, 20, 33]. Therefore, it can be proved that the exercise protocol has an adequate load and entailed muscle damage. Although the between-groups differences in lactate did not reach a significant level, lower CK and LDH levels at 24 and 48 hours after SAFT<sup>90</sup> in the CWI group represented a reduction effect of cold water on muscle damage indices.

In contrast with the current study, some studies indicated that CWI has no reduction effects on muscle damage markers in blood [3, 15, 20, 33]. In addition, there is research reporting incremental effects [36]. However, many similar papers have results consistent with this study and have supported the benefits of CWI for reducing markers associated with EIMD [13, 26, 28, 34]. Since there is a wide range of variables (various training protocols, CWI temperature, duration and repetition) which could influence study outcomes in this field, providing a definite conclusion about exact causes might not be possible. In spite of this, perhaps a potential solution for this controversy can be found in differences in water temperature used in relevant studies. The reasoning for this idea derives from comparing water temperature in most similar studies to the present [5, 7, 19, 20]. In those studies, water with 14°C and 10°C temperature was applied, similar to the majority of studies, and also because of a suggestion in a review

article [21], and the preponderant results did not report any merit in CWI. Nevertheless, 8°C water in this study was effective in reducing muscle damage markers (CK, LDH) in blood. Therefore, the finding in this study is incompatible with those which suggested that water temperature in the range of 11°C to 15°C entails the best results for recovery [21]. The suggested mechanisms contributing to the reduction effect of CWI on muscle damage include hydrostatic pressure causes by CWI that results in an increase in the osmotic slope and clearing [14, 18, 32]. Also, hydrostatic pressure and vasoconstriction can increase central venous pressure and volume, thereby eliminating debris [16, 17, 23]. The results related to lactate in this research were accordant with others, and the majority of those have revealed that CWI is not an efficient treatment for lactate evacuation. Nonetheless, in a study carried out by Adamczyk et al. CWI (3 min at 8°C) was effective [1]. This contrast may be due to several reasons, but there is a possibility that using untrained subjects was influential in this case, because in other studies athlete participants were used.

In conclusion, the results of this study supported other studies in which CWI had a reducing effect on markers of muscle damage in the blood. However, this finding is contrary to those that have suggested a higher temperature (10–15°C) as the optimal condition. Because of the short off-season period (1 month) subjects were unavailable for the next hours (72 and 96 hours) after recovery for sampling purposes. This and lack of full-scale control of participants' daily routine habits such as sleeping pattern and food intake were some limitations of the current study. Hence, further comprehensive and restricted studies are required to reveal the precise effects and mechanisms, and also to identify the best practical circumstances for CWI.

**Conflict of interest:** Authors state no conflict of interest.

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Received 20.05.2020

Accepted 28.10.2020

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