

Original Research The effect of different water immersion strategies on delayed onset muscle soreness and inflammation in elite race walker

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Abstract

Background: This study aimed to investigate the effects of cold water immersion (CWI) and contrast water therapy (CWT) on serum interleukin 6 and prostaglandin 2 levels in self-perceived exertion, and muscle soreness of elite race walkers over a 15-day high-intensity training period. **Methods**: Thirty elite male race walkers were randomly divided into three groups: control group (C, n = 10), cold-water immersion (CWI, n = 10) group, contrast water therapy (CWT, n = 10) group. After daily training, elite race walkers were exposed to either CWI (10 minutes at 10 °C) or CWT (4 cycles of 2.5 minutes, alternately at 12 °C and 38 °C). Elite race walkers in the control group only performed simple stretching without any additional treatment. The serum interleukin 6, prostaglandin 2, self-perceived exertion, and muscle soreness were tested at 6 training points at baseline (B), light load-1 (L1), heavy load-1 (H1), medium load (M), heavy load-2 (H2), light load-2 (L2), respectively. **Results**: When compared with the CWT group, the interleukin 6 level, prostaglandin 2 level, self-perceived exertion, and muscle soreness of the C group were not significantly different. When compared with the CWT group, the interleukin 6 level in the CWI group was significantly lower at the time point of L1 and H2. Similarly, CWI significantly different in both groups. **Conclusions**: The results from this study demonstrate that CWI may be more effective than CWT for reducing inflammatory markers at certain points in a training cycle, but it does appear that this effect can be induced in a predictable fashion.

Keywords: water immersion; interleukin 6; prostaglandin 2; elite race walker

1. Introduction

Delayed onset muscle soreness (DOMS) refers to the phenomenon of muscle soreness caused by micro-damage of muscle cell structure following eccentric exercise or unaccustomed high-intensity exercise, which traditionally peaks at 24–72 hours [1–3]. In addition to localized muscle pain, symptoms of DOMS may include swelling, joint stiffness, reduced joint range of motion, and reduced muscle strength, all of which can seriously affect athletic training [4].

Studies have shown that interleukin 6 level is a sensitive indicator of skeletal muscle micro-injury [5,6]. In addition, prostaglandin 2 is the main substance that causes local pain in the muscles [7,8]. How to reduce the chemical substances which, induce DOMS, has long been the focus of exercise physiology research [9]. Currently, foam rolling, massage, and soft tissue oscillation are often used to mitigate the effects of DOMS and accelerate recovery [10-12]. In addition, cryotherapy is a modality used for recovery that acutely lowers intramuscular temperature. This in turn reduces blood flow in an attempt to reduce muscle metabolism and inflammation, and tissue damage in animal models of muscle injury [13]. CWI is one of the most popular cryotherapy modalities following a variety of exercises [14]. It has also been found that CWI has a better alleviation effect on DOMS [15]. Studies have indicated that cold water immersion can relieve delayed muscle soreness and inflammatory responses primarily through two pathways. On one hand, cold water induces vasoconstriction, decrease tissue temperature and nerve conductivity, thereby reducing the production of muscle spasm and pain after exercise [16]. On the other hand, changes in blood flow caused by hydrostatic pressure may promote the clearance of muscle metabolites, thereby improving metabolic recovery after intense exercise [17].

CWT is another kind of post-exercise recovery treatment [18]. CWT requires alternate temperature immersion, such as going from a hot to a cold bath and vice versa. Vascular 'pumping' produced by temperature changes has been hypothesized as a mechanism that might promote recovery and it has been thought to improve sports related recovery by improving blood lactate elimination, decreasing inflammation and edema, stimulating circulation, alleviating stiffness and discomfort, enhancing range of motion, and lowering delayed onset of muscular soreness [19].



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Currently, there is only one study that has investigated the long-term effects of CWI in a highly trained group of athletes. The study concludes that prolonged cold water immersion can have beneficial effects on mean power and sprint power in athletes [20]. However, the effect of CWI and CWT on serum interleukin 6 levels, prostaglandin 2 levels, self-perceived exertion, and muscle soreness in elite race walkers over a periodic high-intensity training period has not been extensively investigated. Therefore, the objectives of this study were to investigate two different water immersion strategies on delayed onset muscle soreness and inflammation in elite race walkers during a 15-day highintensity training period.

2. Methods

2.1 Experimental designs and procedures

This experimental research was an applied study. All race walkers (as shown in Table 1) who participated in this study were given a full familiarization of the protocol, 2 weeks before the study. The informed consent was obtained from each race walker. In addition to that, we received informed consent from the guardians of some underage race walkers who were younger than 18 years old. All the study participants signed an informed consent form. Elite race walkers in the control group only performed simple stretching exercises without any treatment. A schematic representation of the protocol was provided in Fig. 1. The study was stretched over a period of 15 consecutive days which was at the beginning of the competitive season. The training was followed according to the schedule described in Table 2. Immediately after each training day, the race walkers in the CWI and CWT groups were exposed to the recovery intervention. Therefore, recovery intervention of the race walkers in the CWI and CWT were completed 6 times each week. Each athlete would be immersed in cold water in a seated position and the water would reach the levels of the nipples of the chest. CWI was continuously immersed in water temperatures of 10 °C (body function promotion system Turbo-cool XP and six-person pool, Australian iCool) [21]. In the CWT group, the subjects were immersed alternatively at the same water levels at 38 °C and 12 °C with 4 cycles of 2.5 minutes in each immersion (thermostatic heating portable pool M-009L) [22]. After the recovery intervention, 10 mL of blood was collected from the antecubital vein at the 6-time points of baseline (B), heavy load-1 (H1), light load-1 (L1), medium load (M), heavy load-2 (H2), light load-2 (L2). The serum was separated by centrifugation (10 minutes at 1370× g), aliquoted and stored at -20 °C until analysis. All blood samples were collected directly into a serum separator collection tube. Serum was separated by centrifugation at 4000 revolutions per minute for 5 minutes and stored frozen at -80 °C until analysis. Prostaglandin 2 and the cytokines IL-6 levels were measured in serum using a Laboratory Systems Multiskan MS 352 Enzyme Calibrator (Finland) and commercially available assay kits (Jianglai Bio, Shanghai, China). Coefficients for the analyses of Prostaglandin 2, IL-6, were 2.8%, 2.6%, respectively.



Fig. 1. Experimental designs and procedures.

2.2 Subjects

Thirty national 1st-level race walkers from the Liaoning Provincial Race Team were selected for this study as described in Table 1. Participants were randomly divided into C, CWI, and CWT groups, with 10 athletes in each group, according to a random allocation procedure in Excel. Inclusion criteria were: (1) Subjects must be national 1st-level race walkers; (2) Subjects must not have a habit of cold water recovery; (3) Successful completion of the experiment was guaranteed by the subjects. Exclusion criteria were: (1) the athlete was unable to undergo CWI; (2) Completion of the entire experiment was not guaranteed by the subject. The study was ethically approved by the ethical committee of Liao Ning Institute of Sports Science and all study procedures were under relevant guidelines.

2.3 Training program

Table 2 represents the training schedule and test dates. The schedule consisted of four light load sessions (10 km, 5 minutes 20 seconds/km), one moderate load session (20 km, 5 minutes/km) and two high load sessions (30 km, 4 minutes 50 seconds/km). Data was collected in this experiment after an adjustment period (B), after the first high intensity (H1), after the first adjustment (L1), after moderate training (M), after the second high intensity (H2), and after the second adjustment period (L2).

2.4 Subjective measurements

The modified Borg rating of perceived exertion (RPE) and visual analog scale (VAS) was measured at 6-time points during 15 days training schedule.

Table 1. Comparison of participant characteristics.

Group	Age (years)	Training (years)	Fat (%)	BMI
C (n = 10)	18.9 ± 2.5	7.6 ± 3.7	7.1 ± 3.7	19.08 ± 2.2
CWI (n = 10)	19.1 ± 3.1	8.4 ± 4.4	7.3 ± 3.5	19.38 ± 2.9
CWT (n = 10)	20.1 ± 3.3	8.4 ± 3.9	7.8 ± 3.2	19.18 ± 2.2

There was no significant difference (p > 0.05) between the three groups in all indexes above.

Table 2. Training schedule and test dates.

Days	1–3	4	5	6	7–9	10	11	12	13	14–15
Intensity	light	Moderate	High	Rest	Low	Moderate	High	Low	Rest	Low
Abbreviation	В		H1		L1	М	H2			L2
Test	\checkmark		\checkmark		\checkmark	\checkmark	\checkmark			\checkmark

2.5 Statistical analyses

We assessed the normality of each variable from a normal probability plot, and confirmed the distribution using the Kolmogorov-Smirnov statistic. Levene test was used to assess the homogeneity of variance. Each of these analyses was performed using SPSS 25.0 (IBM, Chicago, IL, USA). One-way analysis of variance was followed by the Bonferroni test which was then used to compare subjective and objective indicators in different groups at the same time points. A *p*-value of p < 0.05 was considered statistically significant. The date was shown as mean \pm SD.

3. Results

3.1 Serum interleukin 6 levels six times during the study in the three group

As shown in Fig. 2, there was no significant difference of interleukin 6 in the C, CWT and CWI group at B ($F_{(2,27)} = 2.744$, p = 0.082), H1 ($F_{(2,27)} = 0.076$, p = 0.927), M ($F_{(2,27)} = 1.549$, p = 0.231), L2 ($F_{(2,27)} = 2.671$, p = 0.087). At the time point of L1 ($F_{(2,27)} = 8.862$, $\eta^2 = 0.396$), the interleukin 6 level in the CWI group (7.94 ± 0.39) was significantly lower than the C group (8.34 ± 0.28 , p = 0.036) and CWT group (4.31 ± 0.31) was significantly lower than the C group (4.78 ± 0.28 , p = 0.035).

3.2 Serum prostaglandin-2 levels six times during the study in the three group

As shown in Fig. 3, there was no significant difference of prostaglandin-2 in the C, CWT and CWI group at B ($F_{(2,27)} = 1.246$, p = 0.304), H1 ($F_{(2,27)} = 1.842$, p = 0.178), L1 ($F_{(2,27)} = 0.888$, p = 0.423). At the time point of M ($F_{(2,27)} = 21.982$, $\eta^2 = 0.620$), the prostaglandin-2 level in the CWI group (394.62 ± 17.99) was significantly lower than the C group (448.24 ± 21.61, p = 0.017) and CWT group (423.74 ± 15.76, p = 0.023). At the time point

of H2 ($F_{(2,27)} = 10.999$, $\eta^2 = 0.449$), the prostaglandin-2 level in the CWI group (476.78 ± 26.90) was significantly higher than the C group (439.61 ± 18.54, p = 0.021) and CWT group (442.99 ± 9.25, p = 0.028). At the time point of L2 ($F_{(2,27)} = 68.948$, $\eta^2 = 0.836$), the prostaglandin-2 level in the CWI group (360.49 ± 6.29) was significantly lower than the C group (435.77 ± 8.54, p = 0.037) and CWT group (419.53 ± 23.88, p = 0.012).

3.3 RPE of elite race walkers in the three group

As shown in Fig. 4, there was no significant difference of RPE in the C, CWT and CWI group at B ($F_{(2,27)} = 0.374$, p = 0.691), H1 ($F_{(2,27)} = 3.473$, p = 0.055), L1 ($F_{(2,27)} = 0.861$, p = 0.434), M ($F_{(2,27)} = 0.526$, p = 0.597), H2 ($F_{(2,27)} = 3.312$, p = 0.052), L2 ($F_{(2,27)} = 3.136$, p = 0.06).

3.4 Muscle soreness of race walkers in each group

As shown in Fig. 5, there was no significant difference of muscle soreness in the C, CWT and CWI group at B ($F_{(2,27)} = 1.344$, p = 0.278), H1 ($F_{(2,27)} = 0.256$, p = 0.776), L1 ($F_{(2,27)} = 1.136$, p = 0.336), M ($F_{(2,27)} = 2.887$, p = 0.073), H2 ($F_{(2,27)} = 0.247$, p = 0.783), L2 ($F_{(2,27)} = 0.385$, p = 0.684).

4. Discussion

Water immersion as an effective physical intervention has been widely used in the post-exercise recovery of athletes [23]. CWT and the effect of long-term immersion on the recovery of elite race walkers at the beginning of the training season have been less studied. We coordinated with the training practice of competitive walking teams and studied the effect of different water immersion strategies for 15 consecutive days during the recovery of athletes (Table 2). In addition, we found no significant differences between the two groups by comparative analysis of personal characteristics (Table 1). The study data mentioned above proves that the results are credible.



Fig. 2. Serum interleukin 6 level in each group. *p < 0.05, "p < 0.05 represent the difference between CWI and CWT at the same time point.

4.1 Analysis of the effects of different water immersion schemes on interleukin 6 levels

Interleukin 6 is a pleiotropic cytokine produced by the immune system and has a variety of biological functions [24]. In addition to interleukin 6 is an important cytokine that connects the immune and endocrine systems in sports-induced cell damage. Studies have shown that among all cytokines related to skeletal muscle injury during exercise, the interleukin 6 level in the blood is the earliest and the most significant index [25]. After the highintensity or unsuitable level of exercise stress for about 24 hours, macrophages will secrete various inflammatory mediators such as interleukin 6, and prostaglandin 2, which reduces the pain threshold [26]. In the subsequent training, the athletes felt muscle pain, which seriously affected their athletic ability. The secretion of prostaglandin 2 is also affected by the feedback of interleukin 6. Therefore, the relationship between the production of cytokines and the increase of prostaglandin 2 synthesis is less known and quite complicated. From the results of this study, there was no significant difference in interleukin 6 levels between the C and CWT group at each time point. There was also no significant difference in the interleukin 6 values between the CWI and CWT groups after the first low-load training (B). The interleukin 6 values in the CWI group were significantly lower than that in the CWT group at the first

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adjustment period (L1) and second high-load training period (H2) (p < 0.05) (Fig. 2). This coincided with the time point of DOMS occurrence. It has been shown that delayed muscle soreness occurs 24-72 hours after eccentric exercise [27]. The reason may be that the elite race walkers had been in the recovery period after the vacation and did not have high-intensive training. Therefore, on the third day after the first intensive training (L1), the levels of interleukin 6 in the CWI and CWT groups were significantly different (Fig. 2). On the day of the second intensity training (H2), the increased value of interleukin 6 was probably due to a combination of several factors, such as the adaptation of the athlete to the intensity of the training, while the effect of L1 was not eliminated. The value of interleukin 6 in the CWI group was lower than that in the CWT group and this may reduce the influence and the ability to exercise. This result indicates that CWI may be able to lower the level of interleukin 6 and reduce the degree of inflammation, and the substance metabolism in the later stage.

4.2 Analysis of the effect of different water immersion schemes on prostaglandin 2

Prostaglandin 2 levels increase around 24 hours after muscle injury caused by high-intensity exercise. When the cell membrane is damaged, membrane phospholipids initiate the metabolism of arachidonic acid under the ac-





Fig. 3. Serum prostaglandin-2 level in three group. *p < 0.05 represent the difference among C, CWI and CWT at the same time point.





Fig. 4. RPE value at the 6 times point in three groups.





Fig. 5. Muscle soreness values at the 6 times point during study in three groups.

tion of phospholipase. At this stage, the body secretes prostaglandins 2, which acts on the nervous system through macrophages as part of the immune system [28]. When prostaglandin 2 acts on the human nervous system, it can reduce the local pain threshold and increase the sensitivity of pain receptors to painful stimuli, and make the nervous system more sensitive to substances produced by stressful exercise [29]. In other words, when the prostaglandin 2 is raised, local pain can be generated even if the pain does not usually occur, which ultimately affects the ability to exercise [30]. After the body cannot adapt to the training intensity, prostaglandin 2 secreted by the above reasons can mediate the autocrine feedback mechanism and inhibit endotoxin-induced macrophage production of interleukin 6. Therefore, although both prostaglandin 2 and interleukin 6 can indicate the occurrence and extent of DOMS in the body, they have a complex interaction mechanism. According to the results of this experiment, there was no significant difference in C and CWT group at each time point and there was no significant difference in the values of prostaglandin 2 between the two types of water immersion after the first high-intensity (H1) (Fig. 3). However, after the adjustment, rest for 4 days and then the moderate-intensity training day (M), the value of prostaglandin 2 in the CWI group was significantly lower than that in the CWT group (p < 0.05) (Fig. 3). The second day of the moderate-intensity training day (M) was followed by the second high-intensive training. Subsequently, the prostaglandin 2 level in the CWI group was shown to be significantly higher than that in the CWT group (Fig. 3). After the adjustment for 3 days (L2), the level of prostaglandin 2 in the CWT group was significantly lower than that in the CWT group in the second recovery period (p < 0.05). Based on the results, we can speculate that: (1) prostaglandin 2 change is not uniform, which also shows the complexity of the production and mechanism of prostaglandin 2 and (2) In the case of water immersion, the volatility of prostaglandin 2 is greater, but after continuous high-intensity training, the athletes participated in CWI group had lower serum prostaglandin 2. This means that the choice of water immersion method should be based on the characteristics of the project and the rhythm of the game being selected.

4.3 Analysis of the effect of different water immersion schemes on perceived exertion and muscle soreness

Our findings show that perceived exertion and muscle soreness were not affected by two water immersion methods. The results of this research are consistent with previous studies (Figs. 4,5). One of the previous studies has shown that there was no significant difference in perceived muscle soreness after 19 minutes of water immersion [31]. However, contradictory findings have also been reported that CWT can decrease muscle soreness. Currently, the effect of water immersion techniques on perceived exertion and muscle soreness are conflicting [32]. Our experimental study shows that cold/contrast water therapy cannot induce a significant difference in perceived exertion and muscle soreness. These results may be because of our intervention conditions differed from interventions methods reported in earlier studies.

5. Conclusions

Although our experimental data show that the effects of CWI on interleukin 6 and prostaglandin 2 may be better than that of CWT. Both strategies had no significant differences in self-perceived exertion and muscle soreness in the elite race walkers. We have to emphasize that there were only a small number of significant differences over a lot of measurements and time points, and a lot of the changes in the inflammatory markers look like possible noise, so they may very well not hold meaning.

6. Limitations

The topic of research selected for this experiment was meant for national level top race walkers. The sports subjects were more specific, therefore, a smaller sample was selected.

Author contributions

CG, YZF and CYZ designed the research study. CG and CYZ performed the research. CYZ provided help and advice on the ELISA experiments. XYK analyzed the data. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript.

Ethics approval and consent to participate

The study was ethically approved by the ethical committee of Liao Ning Institute of Sports Science and all study procedures were under relevant guidelines (Ethical approval number: 2018A103). All the study participants signed an informed consent form.

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Conflict of interest

The authors declare no conflict of interest.

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