

**Post-exercise hot water immersion induces heat acclimation and improves endurance  
exercise performance in the heat**

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**Running Head:** Heat acclimation by post-exercise hot bath

## Abstract

We examined whether daily hot-water-immersion (HWI) after exercise in temperate conditions induces heat-acclimation and improves endurance performance in temperate and hot conditions. Seventeen non-heat-acclimatized males performed a 6-day intervention involving a daily treadmill-run for 40min at 65%  $\dot{V}O_{2max}$  in temperate conditions (18°C) followed immediately by either HWI (N=10; 40°C) or thermoneutral (CON, N=7; 34°C) immersion for 40min. Before and after the 6-day intervention, participants performed a treadmill run for 40min at 65%  $\dot{V}O_{2max}$  followed by a 5 km treadmill-time-trial (TT) in temperate (18°C, 40% humidity) and hot (33°C, 40% humidity) conditions. HWI induced heat-acclimation demonstrated by lower resting rectal temperature ( $T_{re}$ , mean, -0.27°C,  $P<0.01$ ), and final  $T_{re}$  during submaximal exercise in 18°C (-0.28°C,  $P<0.01$ ) and 33°C (-0.36°C,  $P<0.01$ ). Skin temperature,  $T_{re}$  at sweating onset and RPE were lower during submaximal exercise in 18°C and 33°C after 6-days in HWI ( $P<0.05$ ). Physiological strain and thermal sensation were also lower during submaximal exercise in 33°C after 6-days in HWI ( $P<0.05$ ). HWI improved TT performance in 33°C (4.9%,  $P<0.01$ ) but not in 18°C. Thermoregulatory measures and performance did not change in CON. Hot-water-immersion after exercise on 6-days presents a simple, practical and effective heat-acclimation strategy to improve endurance performance in the heat.

**Keywords:** Thermoregulation; Hyperthermia; Performance; Running; Heat illness; Hot bath

## **Introduction**

Athletes, military personnel and firefighters are often required to perform in the heat which increases physiological demands and places substantial strain on heat loss mechanisms (Cheung et al., 2000). To reduce the risk of exertional-heat-illness (EHI) and improve exercise capability in the heat these individuals often prepare by completing an exercise heat acclimation protocol. The adaptive responses to exercise heat acclimation have been widely documented (Taylor, 2014) and include, but are not limited to, earlier onset of cutaneous vasodilatation and sweating, increases in sweating rate, reduced cardiovascular strain and, in turn, reduced core temperature and physiological strain, and improved endurance capacity during exercise in the heat. Some evidence supports the notion that the ergogenic benefit of exercise heat acclimation extends to endurance performance in cool conditions (Lorenzo et al., 2010) but recent evidence presents a mixed picture (Neal et al., 2015).

Conventional exercise heat acclimation protocols typically consist of a daily bout of exercise heat stress over a five to sixteen day period where core temperature is increased for 1-2 hours in an artificial hot environment (Nadel et al., 1974; Garrett et al., 2009). Exercise intensity during heat acclimation programs has either been fixed, self-regulated or manipulated to control a precise level of hyperthermia (Fox et al., 1963; Taylor, 2014). Owing to the ensuing adaptations, the training stimulus typically decreases during conventional exercise heat acclimation programs (Taylor, 2014). The controlled hyperthermia technique has the advantage that the adaptive stimulus is kept constant (core temperature clamped at 38.5°C) (Fox et al., 1963), thus optimizing adaptation and, via thermal clamping, affording greater insight into underlying mechanisms (Taylor, 2014). However, there are practical disadvantages using these exercise heat acclimation protocols. These protocols can be costly and impractical for non-acclimated individuals residing in cooler climates as their completion

requires access to an environmental chamber or temporary relocation to a hotter climate to complete the protocol. The controlled hyperthermia technique also requires measurement of core temperature and control of exercise intensity to maintain core temperature at 38.5°C.

One novel, as yet unexplored, approach to these practical limitations may be to have non-heat-acclimated individuals undertake hot-water immersion (HWI) immediately after daily exercise performed in temperate conditions. Related support comes from studies showing thermoregulatory adaptations (Fox et al., 1963; Beaudin et al., 2009) and performance benefits (Scoon et al., 2007) of passive heat acclimation using controlled hyperthermia in a vapor barrier suit or sauna, but these methods are somewhat limited in terms of their practicality and no measures of thermoregulation were reported during exercise-heat-stress (Scoon et al., 2007; Beaudin et al., 2009). Support also comes from studies in non-heat-acclimated individuals showing that repeated HWI over a 10-14 day period decreased core temperature at rest before and during HWI (Brebner et al., 1961; Brazaitis and Skurvydas, 2010) and during light exercise in the heat (Brebner et al., 1961; Bonner et al., 1976). More practical alternatives for heat acclimation would be welcome as these studies used a thermal clamp (Bonner et al., 1976) and an unpleasant 44°C HWI protocol (Brazaitis and Skurvydas, 2010). Extending beyond the obvious practical advantages, as combined elevations in core temperature and skin temperature are important to achieve full heat acclimation (Fox et al., 1964; Regan et al., 1996) there is a reasoned physiological argument for why HWI (elevated skin temperature) immediately after daily exercise in temperate conditions (elevated core temperature) might elicit favorable heat acclimation responses.

To date, no study has investigated whether a daily HWI following exercise in temperate conditions induces heat acclimation and improvements in endurance exercise performance in hot and temperate conditions. To this end, we hypothesized that a short-term (6-day) protocol involving a 40 min HWI each day after sub-maximal treadmill running in a temperate laboratory would induce heat acclimation and performance improvements during a 5 km treadmill time-trial in both temperate (18°C) and hot (33°C) conditions.

## Methods

### Participants

Seventeen physically active, non-heat acclimatized males who had not been exposed to hot environmental conditions in the past 3 months and completed two or more hours of endurance exercise per week were recruited to participate in the study. Participants were randomly assigned to either a 6-day hot water immersion (HWI: N = 10; mean  $\pm$  SD, age:  $23 \pm 3$  years; body mass:  $69.5 \pm 6.9$  kg;  $\dot{V}O_{2\max}$   $60.5 \pm 6.8$  mL/kg/min) or control (CON: N = 7; age:  $23 \pm 3$  years; body mass:  $72.1 \pm 5.8$  kg;  $\dot{V}O_{2\max}$   $60.1 \pm 8.9$  mL/kg/min) intervention. A 6-day intervention was completed to align with other short-term heat acclimation protocols (Aoyagi et al., 1995; Cotter et al., 1997). There were no differences in the characteristics of participants in HWI and CON. The study received local ethical approval and was conducted in accordance with the Declaration of Helsinki (2013). All participants provided full written informed consent, were healthy, non-smokers, free from any known cardiovascular or metabolic diseases and were not taking any medication.

### Study design

Prior to (days -3 to -1), and following (days +1 to +4) the 6-day HWI or CON intervention, experimental trials were completed in temperate ( $18.0 \pm 0.1$  °C,  $42.5 \pm 3.6$  % RH) and hot conditions ( $33.0 \pm 0.3$  °C,  $40.2 \pm 0.7$  % RH; Fig. 1). Experimental trials included a 40 min submaximal run followed by 60 min rest, then a 5 km treadmill time trial (TT). On the day of, and in the 24 hours prior to experimental trials (Fig. 1) no alcohol, any form of diuretics, caffeine or tobacco were consumed and no exercise, other than that prescribed, was undertaken. During the intervention (days 1 to 6) participants were required to consume their normal diet and fluid intake, including caffeine and alcohol ( $\leq 3$  units per day) and to reduce

their regular training by the volume of endurance exercise completed during the intervention in the laboratory.

\*\*\* Fig. 1 near here \*\*\*

### **Preliminary measurements and familiarization**

A continuous incremental exercise test on a motorized treadmill (HP Cosmos Mercury 4.0, Nussdorf-Traunstein, Germany) was used to assess  $\dot{V}O_{2\max}$  in temperate conditions (19 °C, 42 % RH), as described (Fortes et al., 2013). Using the interpolation of the running speed –  $\dot{V}O_2$  relationship, the running speed that elicited 65 %  $\dot{V}O_{2\max}$  was then determined and verified 30 min later. This individualized running speed was used for both the submaximal exercise during experimental trials and in the daily exercise throughout the 6-day intervention. Following the speed verification, participants rested in the laboratory for 60 min. During this time they were familiarized with the speed controls of the treadmill within the environmental chamber (Delta Environmental Systems, Chester, UK) and with all instrumentation and procedures used in the experimental trials. Participants then entered the environmental chamber (18 °C, 40 % RH) and completed a maximal effort 5 km treadmill TT at self-selected intensities. One familiarization was deemed sufficient to mitigate against learning effects (Laursen et al., 2007). The chamber was silent and the only information the participant received was the distance covered displayed on a screen in front of them.

### **Experimental trials**

Participants completed a diet diary in the 24 h prior to their first experimental trial (Fig. 1) and were asked to replicate this prior to further experimental trials. On the day of each experimental trial, participants arrived at the laboratory at 0730 h fasted. They were provided

with a standardized breakfast (0.03 MJ/kg) and a bolus of water equivalent to 7 mL/kg of body mass. At 0800 h on days -1 and +2 a venous blood sample was taken without stasis following a seated rest. Urine samples were collected on all experimental trials and analyzed for urine specific gravity (USG) using a handheld refractometer (Atago Uricon-Ne refractometer, NSG Precision cells, New York, USA). A pre-exercise nude body mass (NBM) was taken using a digital platform scale (Model 705; Seca, Hamburg, Germany) and then the participant was instrumented for the exercise protocol. At 0845 h, dressed in T-shirt, running shorts, socks and shoes participants rested for 15 min in the laboratory (18 °C) to establish baseline measures.

**Submaximal exercise.** At 0900 h dressed in running shorts, socks and shoes the participant entered the environmental chamber that was maintained at either 18 °C, 40 % RH or 33 °C, 40 % RH and completed a 40 min 65 %  $\dot{V}O_{2\max}$  treadmill run (1 % gradient; (Jones and Doust, 1996). During this time no fluids were consumed. Rectal temperature ( $T_{re}$ ), skin temperatures ( $T_{sk}$ ) and heart rate (HR) were monitored continuously and rating of perceived exertion (RPE) (Borg, 1970) and thermal sensation (TS) (Hollies and Goldman, 1977) were recorded every 5 min.  $\dot{V}O_2$ , and respiratory exchange ratio (RER) were assessed from 60 s expired gas samples collected by Douglas bag method at 9-10, 19-20, 29-30 and 39-40 min of exercise. Local forearm sweat rate was measured every 20 s for the first 15 min of exercise to assess the onset of sweating. Immediately following exercise a finger prick blood sample was taken and assessed for blood lactate concentration. A NBM was then taken to estimate whole body sweat losses, and the participant sat quietly in the laboratory in temperate conditions (18 °C) dressed in T-shirt, running shorts, socks and shoes for 60 min. A single bolus of water (5 mL/kg body mass) was consumed within the first 20 min of this rest period.



**5 km treadmill time trial.** The TT was completed immediately following a NBM after the 60 min rest period. The participant re-entered the environmental chamber dressed in running shorts, socks and shoes and completed the TT run on a motorized treadmill (1 % gradient) at self-selected speeds. Participants were instructed to run the 5 km TT as quickly as possible. No feedback other than the distance covered was provided. No fluids were consumed during the TT.  $T_{re}$  and HR were measured continuously, and on completion a NBM was recorded to estimate whole body sweat losses. The participant was then provided with water equivalent to sweat losses and was free to leave the laboratory.

### **Intervention**

The 6-day HWI and CON interventions were completed over consecutive days (days 1 to 6; Fig. 1). Participants in HWI and CON completed the same submaximal exercise protocol on each of these days in temperate conditions (18 °C) and a 40 min water immersion (HWI; 40 °C and CON; 34 °C) following its cessation. The CON intervention controlled for any training and/or hydrostatic effects on subsequent thermoregulatory measures and endurance performance.

**Submaximal exercise.** On each day participants reported to the laboratory between 0600 h and 1000 h. A pre-exercise NBM (after voiding) was taken and after fitting a rectal thermistor and HR monitor participants rested in the laboratory for 15 min to establish baseline measures. Participants then ran for 40 min on a motorized treadmill at 65 %  $\dot{V}O_{2max}$  in a temperate environment (18 °C) dressed in running shorts, socks and shoes. A bolus of water (5 mL/kg of body mass) was consumed in the first 20 min of exercise, to replicate normal training procedures, and  $T_{re}$  and HR were monitored continuously. At the cessation of exercise participants undertook the water immersion (2-3 min transition).

**Water immersion.** Following transition, participants were immersed to the neck in a water bath dressed in shorts. Those completing HWI were immersed in  $39.9 \pm 0.3$  °C water while a thermoneutral water temperature of  $34.1 \pm 0.4$  °C was used for CON. The water temperature was maintained during immersions by adding hot or cold water and allowing water to drain to maintain immersion to neck level, where necessary. The 34 °C water temperature on CON was chosen as pilot testing showed that  $T_{re}$  returned to baseline after exercise at a similar rate to sitting in temperate laboratory conditions (18 °C), and thus would not provide any additional cooling effect. During immersion, no fluids were consumed and  $T_{re}$  and HR were monitored continuously. Immersion ended after 40 min unless the participant removed themselves due to discomfort in HWI. Following immersion participants sat in the laboratory for 15 min without fluids and a NBM was taken and adjusted for fluid intake during the submaximal exercise in order to estimate whole body sweat loss. Participants were free to leave the laboratory when  $T_{re} \leq 38.5$  °C.

## **Measurement and instrumentation**

**Body temperatures.**  $T_{re}$  was measured using a flexible, sterile, disposable rectal thermistor (Henleys Medical Supplies Ltd., Herts, UK) inserted 10 cm beyond the rectal sphincter and recorded using a data logger (YSI model 4000A, YSI, Dayton, USA). Skin temperatures from four sites on the right side of the body (on the chest at a midpoint between the acromion process and the nipple, the lateral mid-bicep, the anterior mid-thigh, and lateral calf) were measured using insulated thermistors (Grant EUS-U, Cambridge, UK) and recorded using a portable data logger (Grant SQ2020, Cambridge, UK). Mean  $T_{sk}$  was calculated using a four-site weighted equation (Ramanathan, 1964).

**Sweating responses.** Changes in dry NBM were used to estimate whole body sweating rate during all intervention days and experimental trials. Local forearm sweating rate was measured by dew point hygrometry during the submaximal run of experimental trials as described (Fortes et al., 2013).

**Physiological strain.** Physiological strain index (PhSI) was calculated using  $T_{re}$  and HR data collected every 5 min throughout submaximal exercise during experimental trials, as described (Tikuissis et al., 2002). This index describes physiological strain on a 0 (no strain) to 10 (very high strain) scale.

### **Blood sample collection and analysis**

Venous blood samples were collected from an antecubital vein into an EDTA vacutainer (BD, Oxford, UK) and aliquots of whole blood were used for the immediate determination of hemoglobin in duplicate (Hemocue, Sheffield, UK) and hematocrit in triplicate (capillary tube method). Plasma volume (day -1) was estimated from body mass, as described (Sawka et al., 1992). The change in plasma volume (day +2) was estimated by correcting the initial plasma volume for the percentage change in plasma volume as described (Dill and Costill, 1974).

### **Statistical analysis**

Data in the results are presented as mean  $\pm$  standard deviation (SD), or mean and 90 % confidence interval of the change for one-tailed tests where stated, and statistical significance was accepted at  $P < 0.05$ . The meaningfulness of the within-subject differences was also calculated using Cohen's d effect size with effect sizes greater than 0.2, 0.5 and 0.8 representing small, medium and large effects. Two sample size calculations (G\*Power 3.1.2)

were performed using mean data taken from a 5-day heat acclimation study (Garrett et al., 2009) and a 5 km treadmill TT reliability study (Laursen et al., 2007). For a one-tailed t-test with alpha level set at 0.05 and power set at 0.8 a sample size of 9 participants was calculated to detect a meaningful heat acclimation induced difference in final exercising  $T_{re}$  (0.3 °C). To detect a meaningful improvement in 5 km treadmill TT performance (set at 3 %) it was estimated that a sample size of 7 participants was needed. To ensure adequate power for both key variables, and allowing for dropout, a sample size of 10 participants was used for HWI. All data were checked for normality and sphericity, and analyzed using t-tests. RER and  $\dot{V}O_2$  were analyzed using one-way repeated measures ANOVA's with Greenhouse Geisser correction to the degrees of freedom where necessary. Tukey's HSD or Bonferroni-adjusted paired *t*-test *post hoc* tests were used where appropriate. Sweating threshold was calculated by plotting individual relationships between local forearm sweating rate and  $T_{re}$ , as described (Cheuvront et al., 2009). To assess cumulative hyperthermia, area under the curve (AUC) analysis was performed on the daily  $T_{re}$  (time  $T_{re}$  was >38.5 °C) in HWI (Cheuvront et al., 2008). Pearson's correlations were performed to determine the strength of the relationship between the AUC and the change in hallmark heat acclimation variables e.g. change in resting  $T_{re}$ . All data was analyzed using SPSS version 20 (IBM Corporation, NY, USA), or GraphPad Prism Version 5.02 (GraphPad Software Inc. La Jolla, USA).

## Results

### Intervention

All participants in HWI and CON completed the 6-day intervention.  $T_{re}$  increased on average  $1.13 \pm 0.24$  °C during 40 min of daily submaximal exercise.  $T_{re}$  increased a further  $1.01 \pm 0.31$  °C during HWI and returned to the pre-exercise resting level during CON immersion ( $-1.10 \pm 0.26$  °C). Total AUC ( $T_{re} > 38.5$  °C during submaximal exercise and immersion) for the 6-day HWI intervention was  $156 \pm 83$  °C/min and for CON was  $2 \pm 4$  °C/min. Total AUC in HWI was greater on day 3 compared with day 1 ( $P = 0.05$ ) but was not different on days 4 to 6 compared with day 1; indicating no significant reduction in the total AUC. No daily differences for total AUC were observed in CON. Heat acclimation was demonstrated in HWI by an increase in whole body sweat rate by day 4 ( $P = 0.02$ ) and an increase in immersion time by day 3 ( $P = 0.04$ ; Table 1). By day 5, 9 out of 10 participants completed the full 40 min immersion in HWI: one participant was removed due to reaching the  $T_{re}$  safety limit ( $T_{re} 39.5$  °C). On all other occasions when the 40 min immersion was not completed participants removed themselves due to discomfort (Table 1). In CON, all participants completed all 40 min immersions and whole body sweat rate was unchanged from day 1 ( $0.39 \pm 0.08$  L/h).

\*\*\* Table 1 near here \*\*\*

### Experimental trials

**Resting responses.** Resting  $T_{re}$  was lower following 6-days in HWI in 9 out of 10 participants with a mean change in resting  $T_{re}$  of  $-0.27$  °C (CI:  $-0.16$  to  $-0.39$  °C,  $P = 0.001$ ,  $d = 0.75$ ; Fig.

2A). There was no change in resting  $T_{re}$  in CON (Fig. 2A). A moderate negative correlation ( $r = -0.39$ ) was observed between the total AUC for the 6-day HWI intervention and the decrease in resting  $T_{re}$ . USG was not different between experimental trials and there was a modest increase in plasma volume from day -1 to day +2 in HWI ( $3 \pm 5 \%$ ,  $P = 0.05$ ), with no change in CON ( $1 \pm 3 \%$ ,  $P = 0.31$ ).

\*\*\* Fig. 2 near here \*\*\*

**Submaximal exercise responses.** After the 6-day HWI intervention, end  $T_{re}$  during submaximal exercise was lower in 9 of 10 participants in 18 °C and 10 of 10 participants in 33 °C (Fig. 2B) where change in end  $T_{re}$  was  $-0.28$  °C (CI:  $-0.16$  to  $-0.40$  °C,  $P = 0.001$ ,  $d = 0.78$ ) in 18 °C and  $-0.36$  °C (CI:  $-0.24$  to  $-0.49$  °C,  $P = 0.0001$ ,  $d = 0.70$ ) in 33 °C. A modest negative correlation ( $r = -0.45$ ) was observed between total AUC for the 6-day HWI intervention and the decrease in end submaximal exercise  $T_{re}$  in 33 °C. CON demonstrated no change in end exercise  $T_{re}$  in either 18 °C or 33 °C (Fig. 2B). HWI decreased end exercise  $T_{sk}$  (18 °C:  $P = 0.001$ ,  $d = 0.86$ ; 33 °C:  $P = 0.001$ ,  $d = 0.60$ ; Fig. 3C) and decreased  $T_{re}$  at the onset of sweating in both 18 °C ( $P = 0.001$ ,  $d = 0.86$ ; Fig. 3A) and 33 °C ( $P = 0.02$ ,  $d = 0.57$ ). End exercise RPE (18 °C:  $P = 0.01$ ,  $d = 0.74$ ; 33 °C:  $P = 0.04$ ,  $d = 0.72$ ; Fig. 3E) and HR were lowered in 18 °C and 33 °C after 6-days in HWI (18 °C:  $-7$ , CI:  $-2$  to  $-11$  bpm;  $P = 0.02$ ,  $d = 0.52$ ; 33 °C:  $-6$ , CI:  $-2$  to  $-10$  bpm;  $P = 0.01$ ,  $d = 0.40$ ) and PhSI ( $P = 0.01$ ,  $d = 0.87$ ; Fig. 3D) and TS were lower in 33 °C ( $P = 0.01$ ,  $d = 0.70$ ; Fig. 3F). HWI had no effect on  $\dot{V}O_2$  and RER in 18 °C or 33 °C. There was no effect of CON on any of the above variables (Fig. 3A-F).

\*\*\* Fig. 3 near here \*\*\*

**5 km treadmill time trial performance.** Endurance exercise performance, assessed via a 5 km treadmill TT, was not altered in CON in either 18 °C (PRE:  $1208 \pm 191$  s and POST:  $1216 \pm 167$  s) or 33 °C (PRE:  $1321 \pm 219$  s and POST:  $1299 \pm 207$  s) indicating no training effect. One HWI participant did not complete the PRE 33 °C TT and another HWI participant's TT data was excluded from analysis due to obvious lack of effort on the POST 18 °C TT (mean % HR max was 82 % compared with 91 % for the group and 96 % for his PRE 18 °C TT). Endurance exercise performance was impaired in 33 °C compared with 18 °C before the intervention ( $P = 0.03$ ,  $d = 0.40$ ; Fig. 4A). The 6-day HWI intervention did not alter TT performance in 18 °C but improved TT performance in 33 °C ( $P = 0.01$ ,  $d = 0.42$ ; Fig. 4A and B). The 4.9 % improvement in TT performance in 33 °C in HWI restored performance to the level observed in 18 °C conditions (Fig. 4A). After the 6-day HWI intervention end TT  $T_{re}$  was lower in 33 °C ( $-0.17$  °C; CI:  $-0.04$  to  $-0.30$ ;  $P = 0.02$ ,  $d = 0.49$ ). There were no other PRE to POST differences in  $T_{re}$  in HWI or CON during the TT.

\*\*\* Fig. 4 near here \*\*\*

## Discussion

These novel findings suggest that heat acclimation can be achieved by HWI after exercise in temperate conditions on 6-days and, as such, presents a practical strategy to improve heat dissipation and endurance performance in the heat. There are two principal findings in the present study that support this recommendation. Firstly, we observed clear evidence of heat acclimation after the HWI intervention demonstrated by lower resting  $T_{re}$  ( $-0.27^{\circ}\text{C}$ ) and lower end submaximal exercise  $T_{re}$  in both  $18^{\circ}\text{C}$  ( $-0.28^{\circ}\text{C}$ ) and  $33^{\circ}\text{C}$  ( $-0.36^{\circ}\text{C}$ ). Hallmark heat acclimation responses also included a lower set point for sweating onset and reductions in  $T_{sk}$ , PhSI, RPE and TS during submaximal exercise in the heat after 6-days in HWI.

Adaptations were evident sooner than day 6 of HWI; for example, whole body sweat rate was increased by day 4 of the intervention. Secondly, the HWI intervention improved 5 km treadmill TT performance 4.9 % in  $33^{\circ}\text{C}$  restoring performance to the level achieved in  $18^{\circ}\text{C}$ . Strengths of this study include control of the time of day for the intervention and experimental trials and the inclusion of a control group. Including CON provides confidence that the adaptations shown in HWI were attributed to bathing in hot water after exercise, since the daily exercise and thermoneutral water immersions completed by CON did not affect thermoregulatory or performance outcomes. We recognize that the addition of a traditional exercise heat acclimation group would have enabled for comparisons with the HWI intervention. Additionally, though we observed a modest expansion of plasma volume in HWI we recognize the weakness of estimating this using hemoglobin and hematocrit and recommend tracer techniques be used to verify this finding.

Current recommendations state that heat acclimation should comprise repeated bouts of exercise in the heat over 1-2 weeks (Racinais et al., 2015). Here the findings suggest that HWI after exercise in temperate conditions on 6-days presents an alternative heat acclimation



strategy that overcomes some of the practical limitations of current heat acclimation strategies. Hallmarks of successful heat acclimation include a decrease in resting and exercising core temperature and an improved exercise capacity in the heat (Nadel et al., 1974). The utility of short-term exercise-heat acclimation protocols lasting 4-6 days has been investigated (Sunderland et al., 2008; Garrett et al., 2012), since most adaptations occur within the first 6 days of heat acclimation (Armstrong and Maresh, 1991) and because a shorter heat acclimation protocol is considered to integrate better into an athlete's training/tapering program. In line with other short-term (Cotter et al., 1997), and traditional, longer term exercise-heat acclimation protocols (Armstrong and Kenney, 1993), our 6-day post-exercise HWI intervention, decreased exercise  $T_{re}$  (Fig. 2B), the  $T_{re}$  at the onset of sweating (Fig. 3A),  $T_{sk}$  (Fig. 3C), PhSI (Fig. 3D) and improved exercise performance in the heat (Fig. 4A). Furthermore, the thermoregulatory benefits of HWI we observed during submaximal exercise in the heat were also apparent in temperate (18 °C) conditions (Fig. 2B). There appear to be additional acclimation advantages of the post-exercise HWI intervention because, unlike some short-term exercise-heat acclimation studies (Sunderland et al., 2008; Garrett et al., 2009; Garrett et al., 2012), we also demonstrate a reduction in resting  $T_{re}$  (-0.27 °C). The responsible mechanism requires elucidation but likely includes increased resting skin blood flow and sweating sensitivity (Taylor, 2014), and/or a potential decrease in thermoregulatory set point (Aoyagi et al., 1997); although this concept is controversial (Romanovsky, 2007). The magnitude of adaptation demonstrated in HWI in the current study, where the total AUC for  $T_{re} > 38.5$  °C was not different on day 6 vs. day 1, compares very favorably with short-term exercise-heat acclimation studies (Sunderland et al., 2008; Garrett et al., 2009; Garrett et al., 2012), including those using controlled hyperthermia ( $T_{re}$  38.5 °C) that maintain a constant adaptation impulse during daily exercise-heat stress (Garrett et al., 2012; Taylor, 2014). Work by Fox et al. and later by Regan et al. demonstrated that

whilst heat acclimation is dependent upon the degree of core temperature elevation the elevation of skin temperature is important for full heat acclimation (Fox et al., 1964; Regan et al., 1996); therefore, indicating the importance of the external thermal stress and a likely role for raised  $T_{sk}$  in the observed adaptations in HWI. Peripheral adaptations to local HWI, with and without a rise in core temperature increased local sweating responses (Fox et al., 1964), later coined “sweat gland training” (Avellini et al., 1982), but the increase in local sweating was more dramatic when both core temperature and skin temperature were elevated (Fox et al., 1964). Thus the combined elevation of  $T_{re}$  (~39.3 °C after each HWI) and  $T_{sk}$  during daily HWI in 40 °C (where  $T_{sk}$  equilibrates with water temperature) after exercise likely accounts for the additional observed benefits shown compared with short-term exercise-heat acclimation.

The majority of studies investigating the effect of heat acclimation on endurance performance have used time-to-exhaustion protocols, e.g.  $\dot{V}O_{2max}$  ramp protocols (Sawka et al., 1985; Garrett et al., 2009) or fixed intensity tests (Nielsen et al., 1997; Scoon et al., 2007). Whilst such tests have shown heat acclimation improvements of ~14 to 32 % (Nielsen et al., 1997; Scoon et al., 2007; Garrett et al., 2009), this magnitude of improvement needs to be considered in the context of the variability of time-to-exhaustion protocols, reported to be as high as 27 % (Jeukendrup et al., 1996). Only a handful of studies have assessed the effect of heat acclimation on self-paced TT performance and these used daily exercise in the heat to induce heat acclimation (Lorenzo et al., 2010; Garrett et al., 2012). To our knowledge, this is the first study to investigate the effects of a daily post-exercise HWI intervention on TT performance and here we demonstrate an improvement in 5 km TT performance of 4.9 % in the heat, where performance was restored to the level achieved in 18 °C conditions (Fig. 4A). The magnitude of performance improvement exceeds the CV (~2 %) for the 5 km TT

(Laursen et al., 2007), and thus, we contend, represents a meaningful performance improvement (Fig. 4B) attributable to the heat acclimation adaptations observed. The reduced heat strain after 6-days in HWI is also likely to benefit more prolonged endurance exercise performance in the heat, by blunting the rise in core temperature; though this requires investigation. The potential benefits of heat acclimation on endurance performance in cooler conditions received little attention until one study showed that 10 daily bouts of exercise-heat stress improved cycling TT performance by 6 % in 13 °C conditions (Lorenzo et al., 2010). Considering the decrease in thermal strain during submaximal exercise in 18 °C after 6-days in HWI (Fig. 2B) we might have anticipated, but did not observe, an improvement in 5 km TT performance in 18 °C. It is conceivable, but requires investigation, that the HWI intervention might improve endurance performance in temperate conditions that presents a greater thermal burden such as a 10 km TT (final  $T_{re}$  during 18 °C 5 km TT was only 38.6 °C).

In conclusion, hot-water-immersion immediately after exercise in temperate conditions on six consecutive days reduced heat strain during submaximal exercise in both temperate and hot conditions, and improved 5 km treadmill TT performance in the heat. For those residing and training in temperate conditions, incorporating a hot bath into the post-exercise washing routine on six consecutive days represents a simple, practical, economical and effective heat acclimation strategy to improve endurance performance in the heat.

## **Perspectives**

This heat-acclimation intervention overcomes a number of practical limitations with current exercise-heat-acclimation protocols. For example, access to a hot environment is not required, neither is precise control of exercising  $T_{re}$ , but also because a post-exercise hot bath

does not interfere with daily training and might be incorporated into post-exercise washing routines. Analogous to “live-high train-low” (Stray-Gundersen et al., 2001) we contend these findings support the concept, ‘train-cool bathe-hot’. Although this alternative heat-acclimation strategy conflicts with current athlete practice which includes post-exercise cryotherapy, the benefits of cryotherapy to improve recovery have been questioned (Leeder et al., 2012). The benefits of HWI are likely greater when core temperature is elevated following exercise, but future research that is mindful of the prior exercise-heat strain, safety and real-world limitations is required to verify this and establish whether the intervention can be optimized for military/occupational or athlete scenarios. For example, the intervention might be manipulated (e.g. reducing the water temperature, duration and/or frequency of exposures) for the military/occupational scenario where the aim is to improve tolerance and safety (reduce EHI risk) to a standard heat challenge in large groups (one-size-fits-all). For athletes wishing to optimize performance in the heat, the intervention could be manipulated to ensure constant physiological strain during exposures. Future studies are also required to investigate the decay of heat-acclimation following this intervention, in males and females, and to assess the purported benefits for cellular training adaptations (Tamura et al., 2014) and immunity (Walsh et al., 2011).

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*Conflicts of interest:* The authors of the study declare that they have no conflicts of interest.

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## Figure Legends

FIGURE 1. Schematic of study design.

FIGURE 2. Influence of a 6-day post-exercise hot water immersion (HWI) or control (CON) intervention on resting rectal core temperature ( $T_{re}$ , A) and end exercise  $T_{re}$  (B) assessed following 40 min running at 65 %  $\dot{V}O_{2max}$  in 18 °C (40 % RH) and 33 °C (40 % RH). Bars show mean at PRE and POST. Lines represent individual participants. \*\*  $P < 0.01$ , PRE greater than POST.

FIGURE 3. Influence of a 6-day post-exercise hot water immersion (HWI) or control (CON) intervention on rectal core temperature at sweating onset ( $T_{re}$ , A), whole body sweat rate (WBSR, B) and end exercise responses for mean skin temperature ( $T_{sk}$ , C), physiological strain index (PhSI, D), RPE (E) and thermal sensation (F) following 40 min running at 65 %  $\dot{V}O_{2max}$  in 18 °C (40 % RH) and 33 °C (40 % RH). Bars show mean at PRE and POST and SD. \*  $P < 0.05$  and \*\*  $P < 0.01$ , PRE greater than POST.

FIGURE 4. Influence of a 6-day post-exercise hot water immersion (HWI) intervention on 5 km treadmill TT performance (A) and % change in 5 km treadmill TT performance (B) in 18 °C (40 % RH) and 33 °C (40 % RH). Shown are mean and SD (A) and mean and 90 % CI of the difference (B). \*  $P < 0.05$  and \*\*  $P < 0.01$ .



Figure 1.

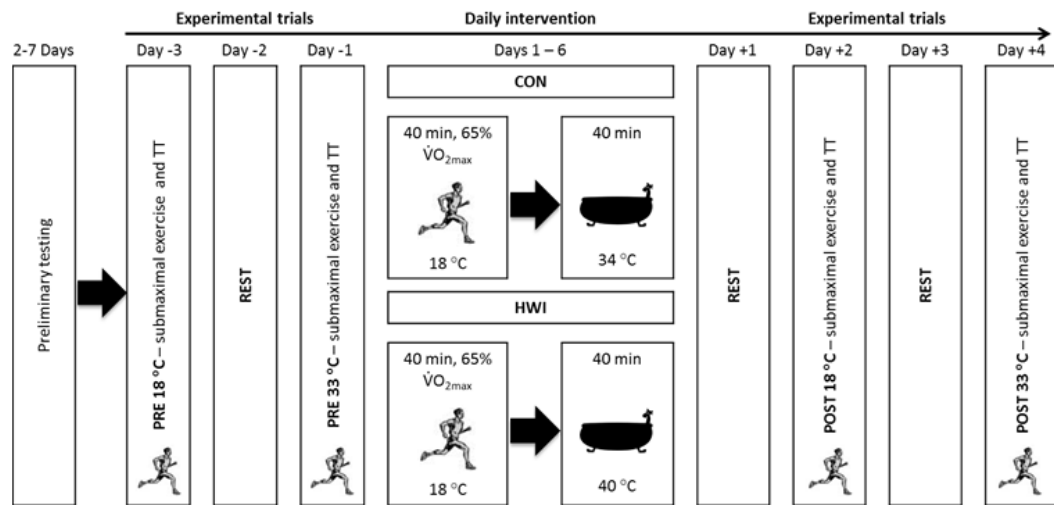


Figure 2.

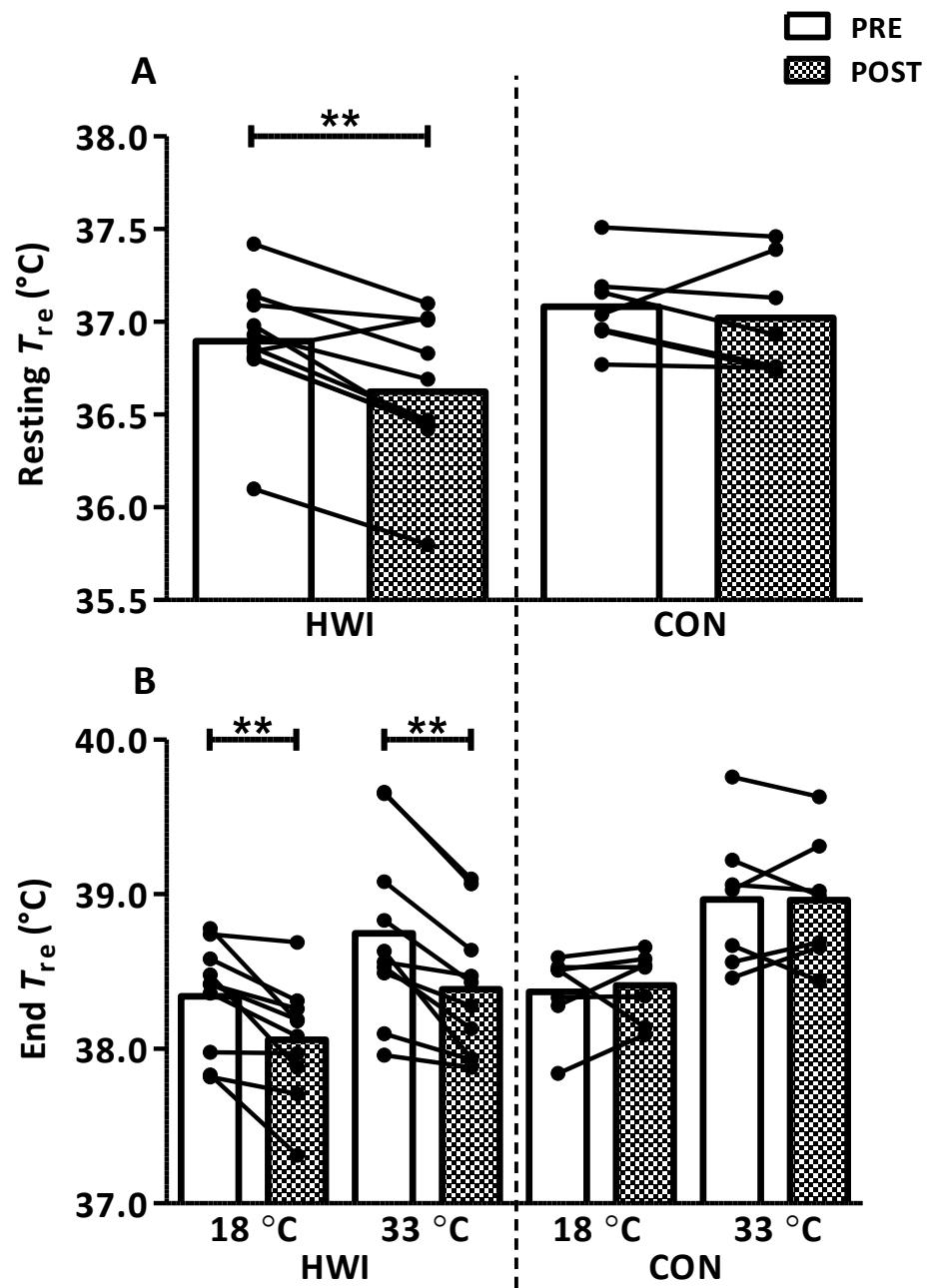


Figure 3.

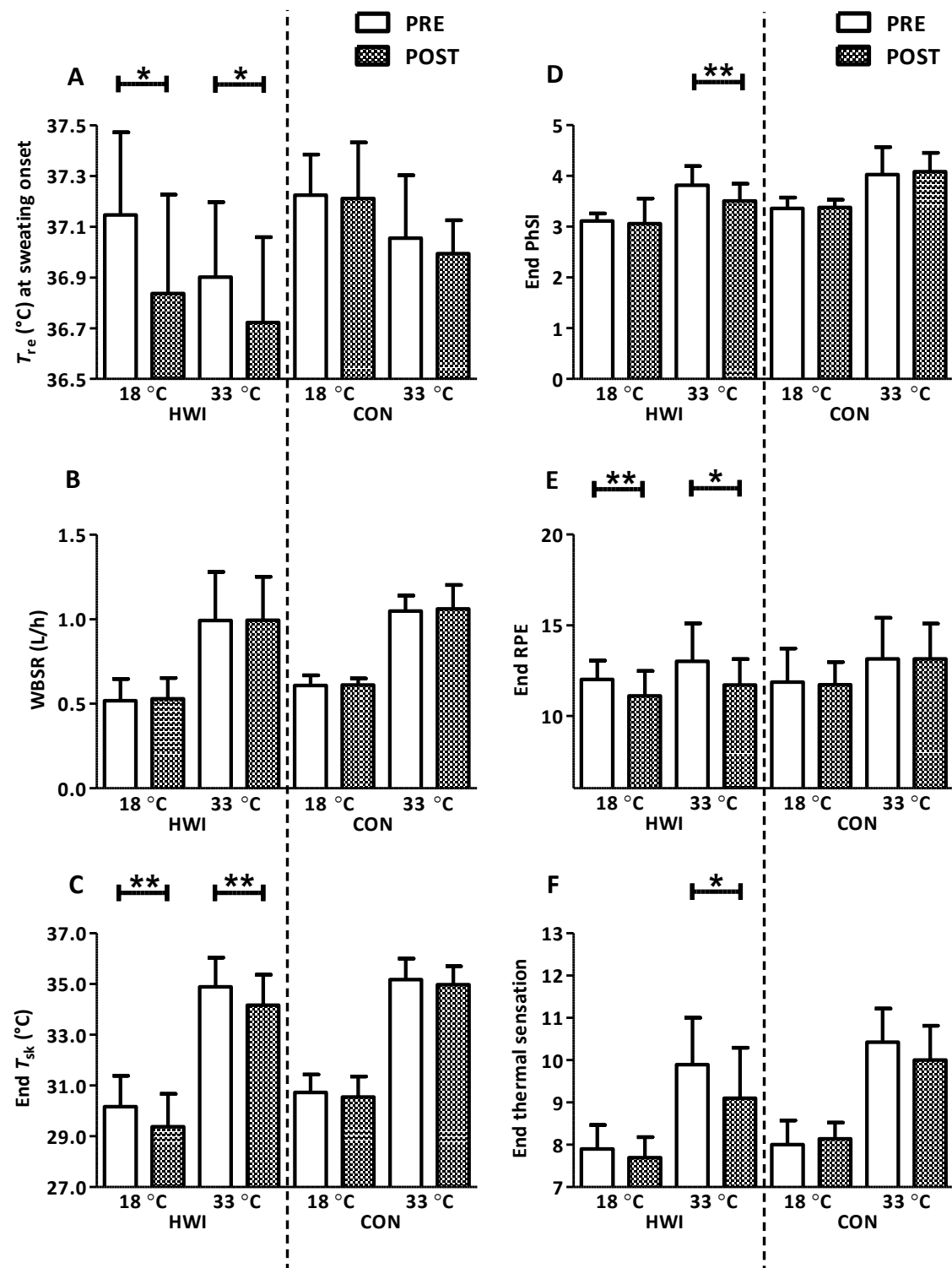
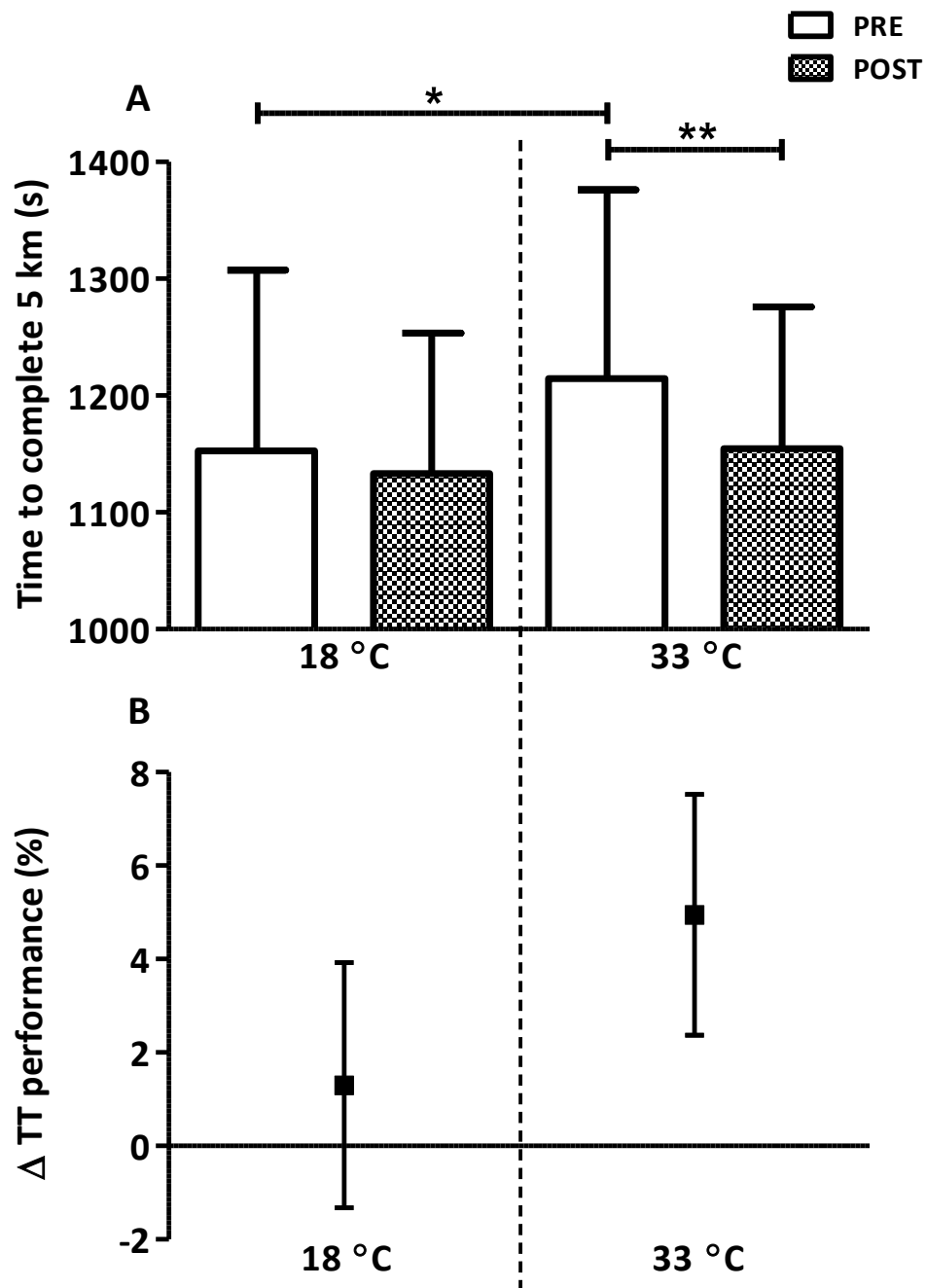


Figure 4.



**Table 1.** The influence of submaximal running at 65%  $\dot{V}O_{2\max}$  for 40 min in 18 °C and post-exercise hot water immersion in 40 °C water immersed to the neck (HWI;  $n = 10$ ) on daily thermoregulatory variables, heart rate and immersion time. Data displayed as mean  $\pm$  SD.

	HWI intervention day					
	1	2	3	4	5	6
<b><i>Submaximal exercise</i></b>						
Change in $T_{re}$ (° C)	1.11 $\pm$ 0.25	1.15 $\pm$ 0.23	1.15 $\pm$ 0.26	1.22 $\pm$ 0.17	1.12 $\pm$ 0.19	1.17 $\pm$ 0.23
Heart rate (beats/min)	142 $\pm$ 13	142 $\pm$ 15	142 $\pm$ 14	140 $\pm$ 12	139* $\pm$ 12	140 $\pm$ 11
<b><i>Hot water immersion</i></b>						
Change in $T_{re}$ (° C)	0.95 $\pm$ 0.27	0.94 $\pm$ 0.33	1.04 $\pm$ 0.40	0.99 $\pm$ 0.31	1.08 $\pm$ 0.28	1.09 $\pm$ 0.30
Immersion time (min:s)	32:50 $\pm$ 07:14	35:18 $\pm$ 06:43	38:00* $\pm$ 03:30	39:21* $\pm$ 01:25	39:36* $\pm$ 01:16	39:45* $\pm$ 00:47
Participants completing 40 min immersion ( $n$ )	4	6	7	8	9	9
<b><i>Submaximal exercise and hot water immersion</i></b>						
Sweat rate (L/h)	0.89 $\pm$ 0.30	0.98 $\pm$ 0.33	1.03 $\pm$ 0.36	1.08* $\pm$ 0.30	1.08** $\pm$ 0.26	1.14** $\pm$ 0.31

\* $P < 0.05$ , \*\* $P < 0.01$  vs. day 1.