Heating up to keep cool: Benefits and persistence of a practical heat acclimation protocol in elite female Olympic team sport athletes

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Abstract

Purpose

Though recommendations for effective heat acclimation (HA) strategies for many circumstances exist, best-practice HA protocols specific to elite female team sport athletes are yet to be established. Therefore, we aimed to investigate the effectiveness and retention of a passive HA protocol, integrated within a female Olympic rugby sevens team training program.

Methods

Twelve elite female rugby sevens athletes undertook 10-days of passive HA across two-training weeks. Tympanic temperature (T_{Tymp}), sweat loss, heart rate (HR), and repeated 6-s cycling sprint performance were assessed using a sport-specific heat stress test Pre-HA; after three days (Mid-HA); after 10 days (Post-HA); and 15-days post-HA (Decay).

Results

Compared to Pre-HA, submaximal T_{Tymp} was lower Mid-HA and Post-HA (both by -0.2 ±0.1 °C; $d \geq 0.71$), while resting T_{Tymp} was lower Post-HA (by -0.3 ±0.1 °C; $d = 0.81$) There were no differences in T_{Tymp} at Decay compared to Pre-HA, nor were there any differences in HR or sweat loss at any timepoints. Mean peak 6-s power output improved Mid-HA and Post-HA (76 ±36 W; 75 ±34 W, respectively; $d \geq 0.45$) compared to Pre-HA. This performance improvement persisted at Decay by (65 ±45 W; $d = 0.41$).

Conclusions

Ten days of passive HA can elicit some thermoregulatory and performance benefits when integrated into a training program in elite female team sport athletes. However, such a protocol does not provide a sufficient thermal impulse for thermoregulatory adaptations to be retained after 15-days with no further heat stimulus.

Keywords: Thermoregulation; Performance; Exercise; Team sport; Olympic Sport

Introduction

Heat acclimation (HA) can elicit physiological adaptations such as lowered core body temperature (T_c), reduced resting and exercising heart rate, plasma volume expansion, and a higher exercise sweat rate.[1] These adaptations facilitate a reduction in perceptual stress and enhanced exercise performance / capacity in the heat.[2] However, for highly-trained team sport athletes, HA may not be a high priority as they may be considered partially acclimated due to their underlying training status, or competing training priorities prohibit exercise-based HA protocols being feasible.[1] Furthermore, the lack of data specific to an elite female athlete population means that best-practice HA for female athletes remains in question.[3]

Females typically have an increased surface area-to-mass ratio, and increased sweating efficiency compared to males,[4] however, sweating capacity, and hence evaporative heat loss capacity, is lower in females compared to males for a given amount of metabolic heat generation.[5] These disparities in thermal stress are likely responsible for the longer general temporal pattern of adaptation described in females compared to males[6,7] and have distinct implications for the structure of female specific HA protocols. For example, in controlled-hyperthermia HA protocols the T_c is typically clamped to 38.5 °C, regardless of sex, meaning the absolute thermal stress to the body may not be equal between sexes.[3]

When investigating sex differences in the physiological adaptations to HA, hormonal fluctuations associated with the menstrual cycle and/or oral contraceptive use can alter thermoregulatory responses and confound findings amongst females.[4] Menstrual cycle phase and oral contraceptive use do not seem to impact thermoregulatory variables such as metabolic heat production, heat loss, or thermoeffector sensitivity during fixed or self-paced exercise in the heat.[8] However, increases in progesterone during the luteal phase has been shown to increase the T_c setpoint by ~0.3 to 0.5 °C, which is mimicked in the active pill phase, and continues into the placebo phase during hormonal contraceptive use.[4]

While the literature to date provides much needed insight into possible female specific HA protocols, the practicalities of including such protocols in an elite team sport environment remains challenging. Given that HA normally takes place in the pre-competition period, competing training priorities and logistical / practical burdens are likely to prohibit such controlled, sustained, and high-intensity exercise-based HA sessions being included at such a time.[1] Differences in menstrual cycle phase among a team can further compound these practical considerations, particularly considering that menstrual cycle phase can lead to fluctuations in T_c setpoint.[4] These competing priorities are in part, why the emergence of passive methods of HA (such as sauna bathing or hot-water immersion) have been explored. Such methods give practitioners and athletes the ability to save mechanical load for specific training modalities, and have been shown to be particularly effective when performed immediately after a temperate training session.[9,10] Indeed, hot-water immersion exposes individuals to a large uncompensable thermal stimulus,[11] and exposure to high skin temperatures has been shown to accelerate heat acclimation adaptation in females.[12]

The retention of thermoregulatory adaptations is an important consideration for elite teams when preparing to compete in the heat. There is some suggestion that physiological, perceptual, and performance changes can be well-retained across the following ~14 days after a heat stimulus is removed.[13,14] However, these suggestions are based from research involving HA protocols that may not be acceptable in an elite setting, along with being primarily performed in male and/or endurance populations.

Integrating practical HA protocols within an elite team sport training schedule is an important consideration for practitioners when preparing to compete in hot environments. Therefore, the aims of the current study were to investigate the physiological, perceptual, and performance adaptations resulting from a 10-day (primarily) passive heat acclimation protocol integrated into a female Olympic team sport training program. Furthermore, it was investigated whether any resulting adaptations could be retained after 15 days of normal training, without any further environmental heat stimulus. We hypothesised that such a protocol would elicit and retain meaningful physiological, perceptual, and performance adaptations.

Materials and methods

Participants

Data were collected from 12 female athletes from the same world-champion and Olympic gold medal winning international rugby sevens team. Menstrual cycle status was recorded using a self-reported questionnaire (see Table 1 for participant details). Data was initially grouped as; natural menstrual cycle and IUD (Natural + IUD) vs. OCP, as it is known that oral contraceptives down-regulate ovarian function, and the exogenous hormones create a stable hormone profile across the weeks of use.[15] Of the naturally cycling, all four participants indicated that they were in days 15-28 of their menstrual cycle (luteal phase; see Table 1). All participants provided informed consent prior to testing, and ethical approval for the study was obtained through the institutions Human Research Ethics Committee (HREC2018#64), in the spirit of the Helsinki Declaration.

<< Table 1 near here >>

Design

All participants undertook a 10-day HA protocol during two weeks of normal rugby sevens training in local springtime conditions (average high temperature ~18 °C). Thermoregulatory, cardiovascular, and perceptual responses to heat stress were assessed before, during, and after a specifically designed heat stress test (HST), intended to replicate the fixed intensity demands of a rugby sevens warm-up and maximal intensity of a rugby sevens game. In total, four HST were performed: Pre-HA (before the commencement of HA); Mid-HA (after 3 days of HA); Post-HA (after 10 days of HA); Decay (15 days after the end of HA). All HSTs were performed in an environmental chamber set at 35 °C, 80% RH, replicating a possible scenario expected at the Tokyo 2020 Olympic Games.[16] Participants performed all testing sessions at the same time of day to account for circadian rhythms and weekly training schedules. During the HA protocol, all participants undertook post-exercise (field-based rugby sevens training) sauna and hot water immersion (HWI) heat exposures (see below for details); whereas the pre- and mid-HA HST's functioned as exercise-based HA sessions. Participants were familiar with performing multiple 6-s cycling sprints, as this was regularly included within their normal training schedule; furthermore, participants had performed a modified intensity (80 % of that prescribed during the HSTs) familiarity session in temperate conditions prior to the commencement of the pre-HA HST. Participants were instructed to refrain from fluid consumption as much as could be tolerated during HA sessions (i.e. permissive dehydration) to induce the added stressor of dehydration.[17,18] Such permissive dehydration methods have recently been shown to lead to significant physiological adaptations during an intermittent heat stress tolerance test in females.[19] A schematic overview of the HA protocol is shown in Figure 1.

<< Figure 1 near here >>

Methodology

Heat stress test

All HST's were performed on a calibrated cycle ergometer (WattBike Ltd, Nottingham, UK) and consisted of a 24-min fixed intensity warm-up, followed by intermittent sprints with the same time structure as a rugby sevens game. In brief, the warm-up took the following structure; 7-min cycling at 2.0 $W \cdot kg^{-1}$ (submaximal); 1-min rest; 7-min cycling at 3.0 $W\cdot kg^{-1}$; 1-min rest; and 3-min cycling at 2.0 $W\cdot kg^{-1}$ with submaximal accelerations during the final 6-s of each minute, followed by a 5-min rest. The repeated intermittent sprint (R-SPRINT) section consisted of 24-s cycling at 3.0 $W\cdot kg^{-1}$, immediately followed by a 6-s maximal sprint and 40-s rest, repeated 12 times with a 2-min half-time break after interval 6. A power output of 3.0 $W\cdot kg^{-1}$ was chosen as this reflected individual mean heart rate during maximal aerobic speed running during pilot testing. Peak power output (PPO) and mean power output (MPO) during the 6-s maximal sprints were used as performance measures. Physiological and perceptual measures (as described below) were taken during seated rest (resting), after each warm-up stage, and after every third interval of the intermittent sprint section, where appropriate measurements were averaged to be used in the final analysis (i.e. warm-up and R-SPRINT).

Post-exercise sauna sessions

All participants undertook four post-exercise sauna heat acclimation sessions, with these sessions being performed within 15-min of the end of an on-field training session. These sessions consisted of passive rest in an environmental chamber set at 40 °C and 80 % RH, in the following format (10-min standing; 15-min sitting undertaking game-specific analysis; 10-min sitting undertaking quiet reflection; 10-min standing).

Hot water immersion (HWI) sessions

All participants undertook four self-directed HWI sessions. These sessions were undertaken on non-training days, away from the teams normal training base. Participants were asked to spend 45 min immersed in 40 °C water, with the first 25 min submerged to the top of chest with arms also submerged. Participants were encouraged to remain fully immersed for the remaining 20 min, however, they could be submerged to the stomach if *very uncomfortable* (≥ 8 on TC scale)[20]. It was instructed that one ≤ 3-min break could be taken, providing that there was no cold stimulus during that time (i.e. cold shower). Participants were given a portable temperature monitor (RS PRO TA298, RS Components Ltd, Auckland, New Zealand) and asked to record water temperature, along with total heat exposure time and session thermal sensation and thermal comfort.

Physiological measurements

During all HST's, tympanic temperature was measured using a validated device [21] (T_{Tymp}; Braun ThermoScan® 7 IRT6520, Braun GmbH, Kronberg, Germany). T_{Tymp} measurements were averaged to produce a value corresponding to each measurement period as described above. Heart rate (HR; Polar H10, Polar Electro Oy, Kempele, Finland) were sampled at the measurement periods described above. To estimate sweat loss, towel-dried, nude body mass (NBM) was recorded to 0.1 kg using digital scales (Tanita HD-351, Tanita Health Equipment H.K. Limited) before and immediately after each HST session, this value was adjusted for a standardised amount of ingested liquid during the HST (640 mL).

Perceptual Measurements

Rating of perceived exertion (RPE; 6-20 scale),[22] thermal sensation (1-13 point scale; 1 = unbearably cold, 10 = unbearably hot),[20] thermal comfort (1-10 point scale; 1 = comfortable, 10 = extremely uncomfortable),[20] and thirst sensation (Thirst; 1-9 point scale; 1 = not thirsty at all, 9 = very, very thirsty)[23] were collected at the same time points described above for physiological measurements during the HST's. Thermal sensation and thermal comfort were collected at the end of each HWI session.

Statistical analysis

Data was initially grouped as; natural menstrual cycle and IUD (Natural + IUD) vs. OCP groups as it is known that oral contraceptives down-regulate ovarian function, and the exogenous hormones create a stable hormone profile across the weeks of use [15]. An independent-samples t-test was run to determine whether differences existed between the Natural + IUD and OCP groups in any of the measured variables with significance set at an alpha level of 0.05. Data from the Natural + IUD and OCP groups were pooled where appropriate, and one-way repeated measures ANOVA were used to determine main effects for all variables between Pre-HA, Mid-HA, Post-HA, and Decay, along with interaction over time for all dependent measures. Normality was assessed using the Shapiro-Wilk test at each time point and Mauchly's test was used to test that sphericity had not been violated. On occasions where sphericity had been violated, the Greenhouse-Geisser correction was applied. Where a main effect was identified, effect magnitudes were determined and expressed as both mean differences ± 90% confidence limits (CL) and standardised effect sizes (Cohen's *d*) sizes whereby 90% CL were used due to the small sample size as suggested by Turner and colleagues.[24] Substantial clear effects were described using standard thresholds of < 0.20 *trivial*, 0.20 – 0.49 *small*, 0.50 – 0.79 *moderate*, and > 0.80 *large.*[25] If the 90% CL for Cohen's *d* overlapped positive and negative trivial (± 0.20) *d* values, the effect was deemed unclear. The smallest worthwhile change (SWC) for rectal temperature (as depicted in Figure 2) was determined from a recent meta-analysis.[2]

Results

During the entire 10-day acclimation process, the mean total heat exposure for each participant was 381 ± 23 min (HST: 90 min; sauna: 180 min; HWI: 115 ± 22 min). There were no significant differences in T_{Tymp}, RPE, thermal sensation, thermal comfort and thirst between Natural + IUD and OCP at any timepoints across any of the HSTs. Peak HR was greater in the OCP group at pre (by 14 ± 9 bpm; $p = 0.013$), mid (by 14 ± 7 bpm; $p = 0.001$) and post (by 13 ± 8 bpm; $p = 0.004$) HSTs. Combined group mean (±SD) physiological and perceptual variables for each HST are presented in Table 2. Both the raw mean (± 90% CL) and standardised mean differences for each comparison are presented in Table 3.

<< Table 2 near here >>

<< Table 3 near here >>

Heat acclimation sessions

During the four self-directed passive HWI sessions, self-reported exposure time was 42 ± 7 min; water temperature was 40.0 ± 1.0 °C; thermal sensation was 10.5 ± 1.2 AU; and thermal comfort was 6.8 ± 2.5 AU; all mean ± SD.

Physiological measurements

The HA intervention elicited changes in resting T_{Tymp} [$F_{(2, 22)} = 21.015$, $p < 0.001$], and submaximal T_{Tymp} [$F_{(2, 22)} = 7.557$, $p < 0.01$] over time; while there was no differences in end exercise T_{Tymp}. Submaximal T_{Tymp} was lower at Mid-HA compared to Pre-HA ($p < 0.05$; $d = -0.71$); while resting and submaximal T_{Tymp} was lower at Post-HA compared to Pre-HA (both $p < 0.01$; $d \geq -0.81$). There were no differences in resting, submaximal or end exercise T_{Tymp} at Decay compared to Pre-HA. See Figure 2, Table 2, and Table 3 for full descriptions of T_{Tymp} change across each HST. The HA intervention elicited no changes in submaximal or R-SPRINT HR or sweat loss over time; see Tables 2 and 3.

<< Figure 2 near here >>

Perceptual measurements

The HA intervention elicited changes in submaximal RPE [$F_{(2, 22)} = 11.026$, $p < 0.01$], and R-SPRINT RPE [$F_{(2, 22)} = 5.671$, $p < 0.01$] over time. The HA intervention did not lead to any changes in submaximal thermal sensation over time; however, changes in R-SPRINT thermal sensation [$F_{(2, 22)} = 4.180$, $p = 0.05$] were evident. The HA intervention elicited changes in submaximal thermal comfort [$F_{(2, 22)} = 5.896$, $p = 0.01$], but not R-SPRINT thermal comfort over time. The HA intervention did not lead to any changes in submaximal and R-SPRINT Thirst over time. Changes in these perceptual measures between HSTs are described in Tables 2 and 3.

Performance measurements

The HA intervention did not lead to any changes in 6s-MPO over time; however, changes in 6s-PPO [$F_{(2, 22)} = 10.641$, $p < 0.001$] were evident. Changes in MPO and PPO over time are described in Tables 2 and 3.

Discussion

The current study examined physiological, perceptual and performance changes during and following 10-days of (primarily) passive HA integrated into an elite female team sport training program. Our data demonstrated that meaningful changes in resting and submaximal T_{Tymp} were achieved only after the full 10-day HA protocol, however these were not well-retained after 15 days of normal training, without any further heat environmental heat stimulus. No sudomotor or cardiovascular changes were apparent during or post HA. Concurrently, meaningful performance increases (specifically R-SPRINT PPO) were evident at both mid- and post-HA and were retained after 15 days of normal training, without any further environmental heat stimulus.

The beneficial changes in resting and submaximal T_{Tymp} seen in the current study are in line with previous findings, whereby decreases in T_c after $\geq$ 9 days of HA in recreationally-trained females have been demonstrated.[6,7] Also, in agreement with the current study, both previous studies found no difference in sweat rate. While the similarities between these findings may strengthen the theory of a longer temporal pattern of HA induction in females,[6] other research with well-trained females has suggested that thermoregulatory adaptation is possible with only 5-days of controlled-hyperthermia HA.[19,26] It is likely that the differences between these studies are associated with the initial training status of the athletes, and the cumulative thermal impulse of the HA protocols.[1] Female specific HA protocols that have shown adaptation in 5-days have exceeded the teams normal peak training load,[26] included five consecutive days of 90-min controlled-hyperthermia exercise-based heat exposures,[19] or included 20-min sauna exposures before each 90-min controlled-hyperthermia exercise-based heat exposure.[12] Neither of these approaches are practical to incorporate into an elite team sports training program; hence, the current study provides the first evidence of an ecologically valid HA protocol that can elicit beneficial thermoregulatory and performance changes in such circumstances.

An important consideration for practitioners when scheduling HA into a wider training macrocycle is the time-course of adaptation retention once a heat stimulus is removed.[13] However, none of the previous research into female specific HA protocols have investigated this phenomenon,[6,7,12,19,26] making the current research novel and noteworthy. The current research indicted many positive thermoregulatory effects post-HA, yet these were mostly transient, with only R-SPRINT PPO showing any evidence of retention after 15 days without any further heat stimulus. Hence, given the relatively low intensity and training program integration of the current HA protocol, it provides a framework that could be completed close to departure for a holding camp or competition in a hot environment where additional heat impulses could be prescribed. Alternatively, the prescription of small weekly (1-3 days) ‘top-up’ heat impulses during the decay period would likely result in greater adaptation retention.[27] The efficacy of ‘top-up’ heat exposures following HA remains to be investigated in elite female athletes, which is likely to be of particular relevance considering the longer temporal adaptations in females. Of note, it was recently demonstrated that following a 10-day controlled hyperthermia HA protocol was most physiological adaptations were retained during a 28- day decay period.[28] These researchers did observe that sudomotor adaptations were lost during the decay period; however, these adaptations could be reinstated with 5-days of either active or passive HA, suggesting that in habitually trained individuals, heat re-acclimation may not be necessary within a 28-day decay period, providing that the initial thermal HA impulse was sufficient to elicit robust adaptations.[29]

The general view among the literature is that eumenorrheic females encounter a performance disadvantage when exercising in the heat during the luteal phase.[4]

Mechanistically, this view is based on threshold shifts to the vasomotor and sudomotor thermoeffectors.[5] However, compared to less-trained females, trained females exhibit altered thermoeffector responses and reduced ovarian hormone concentration and fluctuation between menstrual cycle phases, resulting in a greater capacity to deal with a heat load.[30] Importantly, most previous research concerning the impact of acute heat, or heat adaptation have been based on performance during fixed-intensity exercise, thus not allowing for behavioural thermoregulation.[31] However, even though the current study was underpowered to detect group differences, their appeared to be no significant differences between the Natural + IUD and OCP groups in physiological, perceptual, or performance metrics typically influenced by menstrual phase in less well-trained females.[4] Thus, rather than intrinsic physiological changes, behavioural changes allowing for greater effort, and correspondingly greater peak power output during self-paced repeated sprint exercise may explain the increase in RPE demonstrated in the current study.

Practical Applications

The current study indicated that passive HA was beneficial for repeated sprint performance both over short- and medium-term HA periods. In rugby sevens (and most other invasion or team sport) situations, work rate is often determined by the playing style of the opposition.[32] In this regard, an improvement in repeated sprint performance is practically important as the ability to maintain high intensities can determine outcomes within a game. Although short-term, and/or low thermal impulse acclimation protocols do not induce complete HA adaptations, the current findings demonstrate a practically useful HA protocol for elite team sport as the low intensity, passive nature of the HA protocol allows for other training priorities to take precedence. Future research should test practical re-acclimation protocols ~3 weeks after a similar HA protocol, giving further information to practitioners to support HA periodisation within a pre-competition schedule. Furthermore, future research should quantify the thermal strain of each HA session. Such quantification would allow greater understanding of the dose-response resulting from these practical scenarios, particularly any differences between post-exercise and passive exposures.

The population in the current study allows for unique and ecologically valid findings; however, the nature of undertaking research in such an elite environment involves several limitations. Namely, it precludes the use of a control group which means that we cannot disqualify that the current findings are not due to general training adaptations or a potential learning effect. Nonetheless, it is unlikely that any meaningful non-HA-specific training adaptation occurred during this time, due to the calibre and training status of athletes involved in the study.[33] Furthermore, participants were familiar with performing multiple 6-s cycling sprints, and participants had performed a modified intensity familiarity session in temperate conditions prior to the commencement of the pre-HA HST. The study design is limited by the concurrent training periodisation, whereby controlling for menstrual cycle status is not realistic within the constraints of a professional sporting context. While menstrual cycle status was recorded using a self-reported questionnaire, protected heath information limited us from reporting whether these participants were on active or placebo phases of the OCP.

The current study reported oral contraceptive use and menstrual cycle status without any subsequent methodological control. As such, the current study was underpowered to determine any differences between the Natural + IUD and OCP groups; appropriate consideration of the menstrual cycle in future research may reduce variability in experiments and aid researchers in detecting differences between treatments or groups, including in measurements of body temperature.[4]

Conclusion

The current investigation demonstrates a novel 10-day HA (primarily) passive protocol that elicits minor thermoregulatory and performance benefit when integrated into an elite team's training program. Furthermore, this data showed for the first time that such a protocol does not provide a sufficient thermal impulse for adaptations to be retained after 15-days with no further heat stimulus. Future work should endeavour to determine an ecologically valid HA protocol that is likely to promote more complete adaptations in highly trained female athletes.

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Tables

Table 1: Participant characteristics grouped by contraceptive type use (Natural + IUD and OCP); all Mean ± SD.

Contraceptive type	Participants	Day of menstrual cycle*	Age (y)	Body mass (kg)	Height (cm)
Natural	4	15, 20, 22, 25	22 ± 3	73.1 ± 7.3	168.3 ± 3.2
IUD	2	Not reported			
OCP	6	Not reported	23 ± 3	71.6 ± 6.6	168.8 ± 2.3
Mean	**12**	**-**	**22 ± 3**	**72.4 ± 7.3**	**168.6 ± 2.3**

IUD = intrauterine device; OCP = oral contraceptive pill

*Reported at the Pre-HA HST. Day of menstrual cycle at Mid-HA: 18, 23, 25, 28; Post-HA: 25, 2, 4, 7; Decay: 9, 14, 19, 22.

Table 2: Mean ± SD for variables during heat stress tests (HST) pre-, mid-, post-, heat acclimation (HA) and +15 days (decay).

		Heat Stress Test			
Variable	**Time**	**Pre-HA**	**Mid-HA**	**Post-HA**	**Decay**
Tympanic Temperature (°C)	*Resting*	37.3 ± 0.3	37.2 ± 0.3	37.0 ± 0.4***#	37.2 ± 0.2
	Sub-max	37.7 ± 0.2	37.6 ± 0.2*	37.5 ± 0.3**	37.5 ± 0.2
	End Exercise	38.6 ± 0.5	38.4 ± 0.4	38.6 ± 0.4	38.8 ± 0.4
Heart Rate (bpm)	*Sub-max*	151 ± 12	152 ± 6	149 ± 8#	151 ± 7
	R-SPRINT	171 ± 9	169 ± 8*	171 ± 7	171 ± 8
RPE (AU)	*Warm-up*	13 ± 1	14 ± 1*	15 ± 1**#	14 ± 1**
	R-SPRINT	16 ± 1	16 ± 1	17 ± 1*#	17 ± 1
Thermal Sensation (AU)	*Warm-up*	9 ± 1	9 ± 1	10 ± 1	10 ± 1
	R-SPRINT	10 ± 1	9 ± 1	10 ± 1##	10 ± 1
Thermal Comfort (AU)	*Warm-up*	5 ± 2	4 ± 1*	4.5 ± 1.3*	5 ± 1
	R-SPRINT	6 ± 2	5 ± 2*	5.7 ± 1.5	6 ± 1
Thirst (AU)	*Warm-up*	4 ± 1	4 ± 2	4.2 ± 1.2	4 ± 1
	R-SPRINT	6 ± 2	6 ± 2	5.5 ± 1.5	6 ± 1
Sweat Loss (kg)	*Mean*	1.3 ± 0.4	1.3 ± 0.4	1.3 ± 0.3	1.4 ± 0.4
Power Output (W)	*Mean*	567 ± 110	600 ± 117	602 ± 94	580 ± 120
	Peak	704 ± 145	780 ± 165**	780 ± 134**	770 ± 152*

* = different to Pre; # = different to mid; ^ = different to post. The number of symbols represent the significance level; 1 = $p \leq 0.05$, 2 = $p \leq 0.01$, and 3 = $p \leq 0.001$. AU = Arbitrary Units; RPE = Rate of perceived exertion

Table 3: Mean difference ± 90% CL; (Cohen's *d*) for variables during heat stress tests (HST) pre-, mid-, post-, heat acclimation and +15 days (decay).

Variable	Time	Mid - Pre	Post - Pre	Post - Mid	Decay - Post	Decay - Pre
Tympanic Temperature (°C)	*Resting*	-0.1 ± 0.1; (-0.34) *unclear*	-0.3 ± 0.1; (-0.81) ***large***	-0.2 ± 0.1 (-0.53) ***moderate***	0.2 ± 0.2 (0.54) *unclear*	-0.1 ± 0.2 (-0.35) *unclear*
	Sub-max	-0.2 ± 0.1; (-0.71) ***moderate***	-0.3 ± 0.1; (-0.95) ***large***	-0.1 ± 0.1 (0.36) *unclear*	0.1 ± 0.2 (0.26) *unclear*	-0.2 ± 0.2 (-0.81) ***large***
	End Exercise	-0.2 ± 0.2 (-0.44) ***small***	-0.1 ± 0.3; (-0.12) *unclear*	0.2 ± 0.3 (0.33) *unclear*	0.2 ± 0.2 (0.46) *unclear*	-0.1 ± 0.3 (-0.32) *unclear*
Heart Rate (bpm)	*Sub-max*	1 ± 6; (-0.04) *unclear*	-2 ± 7; (-0.22) *unclear*	-3 ± 2; (-0.38) ***small***	2 ± 5; (0.26) *unclear*	0 ± 5; (-0.03) *unclear*
	R-SPRINT	-2 ± 2; (-0.28) *trivial*	0 ± 4; (-0.38) *unclear*	3 ± 3; (-0.12) *unclear*	0 ± 2; (-0.02) *unclear*	0 ± 3; (-0.00) *unclear*
RPE (AU)	*Warm-up*	0.6 ± 0.5; (0.58) *moderate*	1.4 ± 0.7; (1.24) ***large***	0.8 ± 0.5; (0.67) ***moderate***	-0.4 ± 0.6; (-0.42) *unclear*	1.0 ± 0.4; (1.14) ***large***
	R-SPRINT	0.2 ± 0.7; (0.19) *unclear*	1.0 ± 0.8; (0.81) ***large***	0.8 ± 0.6; (0.62) *moderate*	-0.3 ± 0.4; (-0.23) *unclear*	0.6 ± 0.8; (0.49) *unclear*
Thermal Sensation (AU)	*Warm-up*	0.1 ± 0.4; (0.05) *unclear*	0.5 ± 0.5; (0.51) *unclear*	0.4 ± 0.5; (0.51) *unclear*	-0.2 ± 0.2; (-0.29) *unclear*	0.3 ± 0.5; (0.32) *unclear*
	R-SPRINT	-0.4 ± 0.4; (-0.45) *unclear*	0.5 ± 0.7; (0.50) *unclear*	0.9 ± 0.5; (1.01) ***large***	-0.4 ± 0.6; (-0.55) *unclear*	0.1 ± 0.5; (0.09) *unclear*
Thermal Comfort (AU)	*Warm-up*	-0.6 ± 0.4; (-0.40) ***small***	-0.5 ± 0.4; (-0.34) *trivial*	0.1 ± 0.4; (0.08) *unclear*	0.0 ± 0.5; (0.00) *unclear*	-0.5 ± 0.5; (-0.32) *unclear*
	R-SPRINT	-0.6 ± 0.5; (-0.32) *trivial*	-0.3 ± 0.5; (-0.17) *unclear*	0.3 ± 0.6; (0.16) *unclear*	0.3 ± 0.5; (0.18) *unclear*	0.0 ± 0.6; (0.00) *unclear*
Thirst (AU)	*Warm-up*	-0.2 ± 0.4; (-0.11) *unclear*	-0.1 ± 0.7; (-0.10) *unclear*	0.1 ± 0.6; (0.03) *unclear*	0.0 ± 0.3; (0.02) *unclear*	-0.1 ± 0.6; (-0.08) *unclear*
	R-SPRINT	-0.1 ± 0.4; (-0.07) *unclear*	-0.2 ± 0.6; (-0.12) *unclear*	-0.1 ± 0.6; (-0.03) *unclear*	0.2 ± 0.6; (0.10) *unclear*	0.0 ± 0.7; (-0.02) *unclear*
Sweat Loss (kg)	*Mean*	0.0 ± 0.1; (0.10) *unclear*	0.1 ± 0.1; (0.19) *unclear*	0.0 ± 0.1; (0.08) *unclear*	0.0 ± 0.2; (0.08) *unclear*	0.1 ± 0.1; (0.13) *unclear*
Power Output (W)	*Mean*	33 ± 34; (0.27) *unclear*	35 ± 31; (0.32) *unclear*	2 ± 29; (0.02) *unclear*	-22 ± 28; (-0.19) *unclear*	13 ± 38; (0.11) *unclear*
	Peak	76 ± 36; (0.45) ***small***	75 ± 35; (0.50) ***small***	0 ± 31; (0.00) *unclear*	-11 ± 34; (-0.07) *unclear*	65 ± 45; (0.41) ***small***

AU = Arbitrary Unit; RPE = Rate of perceived exertion

Figures

Week	Mon	Tue	Wed	Thu	Fri	Sat	Sun
1					Pre-HST	*No heat exposure*	HWI
2	Post-ex-sauna	Post-ex-sauna	Mid-HST	HWI	Post-ex-sauna	HWI	*No heat exposure*
3	HWI	Post-ex-sauna	Post-HST	*No heat exposure*			
4	*No heat exposure*						
5	*No heat exposure*				Decay-HST		

Figure 1: Overview of the heat acclimation timeline. HST = Heat Stress Test; HWI = passive heat acclimation session involving ~45 min hot-water immersion in ~40 °C water; Post-ex sauna = passive heat acclimation session involving 45-min passive rest in an environmental chamber set at 40 °C and 80 % RH.

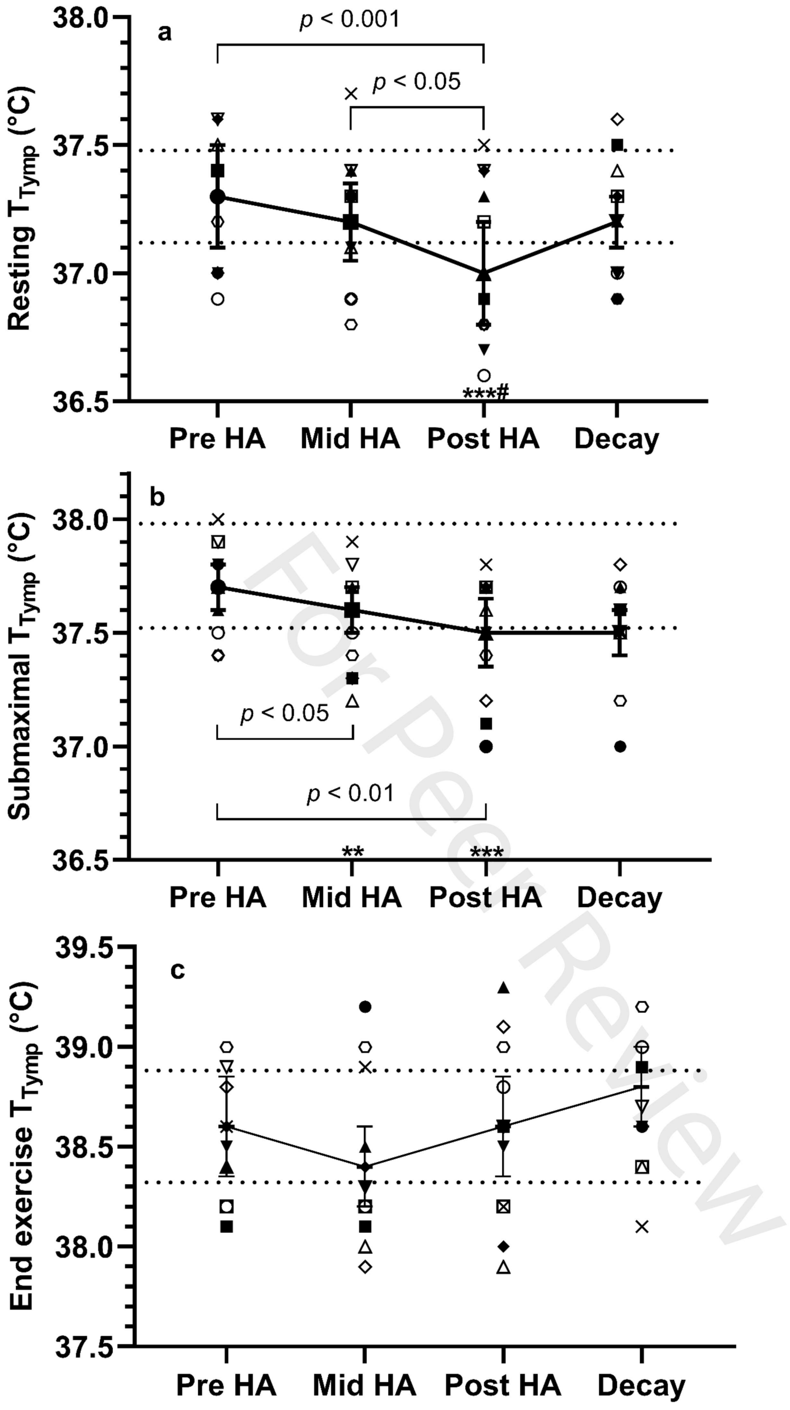

Figure 2: Resting (Figure 2a), Submaximal exercise (Figure 2b) and End exercise (Figure 2c) tympanic temperature (°C) during Heat Stress Tests Pre-HA, Mid-HA (three-days), Post-HA (nine days) and Decay (+15 days after Post-HA). The area between the dotted lines represents the smallest worthwhile change for rectal temperature. Individual data for each group is represented by; open symbols = represent Natural + IUD; closed symbols represent OCP; linked symbols represent mean ± 95% confidence limits. Where statistical significance occurred, it is indicated. Symbols above the x-axis represent standardised effect sizes (Cohen's *d*) for the following comparisons: * = compared to Pre-HA; # = compared to Mid-HA. The number of

symbols represent the size of the effect; 1 = *small*, 2 = *moderate*, and 3 = *large*. HA = Heat Acclimation