



Mixed-Method Heat Acclimation Induces Heat Adaptations in International Triathletes Without Training Modification

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Abstract

Purpose Heat acclimation (HA) kinetics often necessitates that the intervention is conducted in the weeks immediately preceding athletic competitions, potentially interfering with a training taper. Therefore, we investigated the efficacy of a mixed-method HA protocol, superimposed over planned external training loads, during the 3-weeks prior to the 2022 U23 World Triathlon Championships.

Methods Six international triathletes completed 8 pre-competition HA sessions (5 active: running/cycling, 3 passive: hot water immersion [HWI]), across 2-weeks. Outdoor high-intensity training sessions were followed by 30–60 min HWI, whilst low-intensity cycling/running sessions were completed in a hot, humid environmental chamber. To assess heat adaptations, participants completed three 25 min heat stress tests (HST) involving iso-speed treadmill running (session 1 = HST1, session 5 = HST2, and session 8 = HST3). Physiological, haematological and wellbeing monitoring were conducted throughout HA.

Results Reduced heart rate (~ -6 beats/min) was observed within HST3 ($P=0.01$, $\eta_p^2=0.64$), versus HST1 and HST2. No changes in core temperature were observed across HSTs ($P=0.055$, $\eta_p^2=0.44$). Sweat sodium concentration was lower by HST2 at the arm (-23 ± 16 mmol/L, $P=0.02$) and back (-27 ± 17 mmol/L, $P=0.01$). White blood cell count reduced from baseline to the end of HA ($P=0.02$, $\eta_p^2=0.27$), but no changes were found in any other haematological markers (all $P>0.05$). Perceptual wellbeing measures did not change across HA (all $P>0.05$).

Conclusion By HST3, seven prior mixed-method HA sessions improved markers of heat adaptation (exercising HR and sweat concentration) within international triathletes. Mixed-method HA may be implemented without modifying training load, with no apparent detrimental effects on athlete health or training stress markers.

Keywords Adaptation · Endurance · Performance · Thermoregulation · Triathlon

Introduction

Elite athletes habitually undertake complex training programmes, whereby physical preparation is balanced against technical and tactical preparation priorities.

Physical training demands are typically monitored through internal (i.e. heart rate [HR], rating of perceived exertion [RPE]) and external training loads (i.e. duration, distance) [1, 25, 26]. In endurance sports, weekly external training volumes may exceed 20 h [8]. Whilst multisport events (e.g. pentathlon, heptathlon, triathlon), must incorporate multiple exercise modalities within this training volume. Evidently, allocation of training time is challenging for elite athletes and sessions may integrate concurrent physical, technical, tactical and skill-orientated training objectives [45].

Prior to competing in hot [16, 31], and sometimes temperate environments [35, 37], heat acclimation and acclimatization form a central focus of many elite athlete's preparation [15, 50]. The prevalence of heat acclimation/acclimatization training reflects the greater performance and physiological effects of heat adaptation, compared

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with acute heat alleviation interventions [30], such as body pre-cooling [29]. A particular characteristic of heat acclimation (HA) is that phenotypic adaptations both induce and decay across a typical training mesocycle (i.e. weeks) [11]. The HA phenotype is characterised by adaptations including decreased resting and exercising, core (T_{CORE}) and skin (T_{SKIN}) temperatures, changes in sweat rate and composition, and a reduction in exercising heart rate (HR), which is likely due to plasma volume (PV) expansion [53]. The majority of HA evidence originates from programmes comprising 5–10 days of 60–90 min exercise training sessions [16, 48, 56]. Whilst HA demonstrates effectiveness as a performance ergogen across several sports, the implementation alongside other training priorities is evidently challenging. Moreover, in endurance sports such as triathlon, completing HA in the weeks preceding competition likely coincides with carefully planned training taper phases [43, 44]. Therefore, contemporary perspectives are exploring HA prescription methods that mitigate some of these programming challenges [6].

Recent recommendations suggest acclimating ‘early’ (i.e. > 4-weeks prior to competition) and then undertaking subsequent regular maintenance thermal exposures (i.e. training sessions where individuals experience considerable thermal stimuli) [16]. This enables greater programming flexibility and training of concurrent physical and technical objectives, in the weeks preceding a competition [16, 49]. Despite negating disruption to a training taper phase, maintenance sessions necessitate a greater number of total training hours being devoted to heat preparation, compared with a traditional, concentrated programme. Other emerging approaches enabling greater flexibility around training programming include; passive heating protocols (either standalone, or post-exercise) [34, 40, 54], training twice on non-consecutive days [59, 62], completing regular training whilst overdressing [23, 60], or a combination thereof, known as mixed-methods HA [52]. These emerging approaches afford athletes the benefit of eliciting increased heat strain, without replacing planned training sessions or imposing additional external training load. Passive heat exposures demonstrate efficacy for inducing HA in sporting populations [22]. However, the ability to familiarise with representative pacing and thermal/exertional perception, that play an important role in endurance performance outcomes [27], may be limited by purely passive approaches. Thus, superimposing active and passive HA on programmed training is likely both pragmatic and efficacious for a variety of sporting scenarios, where modification to a periodised training programme may be undesirable [40, 52].

Notwithstanding the addition of heat stress to an athlete’s training, the programming of elite endurance athlete’s training already requires a delicate balance between

providing stimuli for adaptation and mitigating the risk of ‘overreaching’ and ‘overtraining’. Symptoms of both may include altered immune status, such as decreases in neutrophil function, serum and salivary immunoglobulin concentrations and white-blood cell count, in athletes [39]. Therefore, the addition of heat stress represents an additive stressor for athletes, which independently can provide a further immune challenge [21, 42]. Laboratory-based HA does not appear to compromise immune, stress or inflammatory biomarkers following short- (4 days [58]; 5 days [62] and medium-term time scales, for both passive (9 days [33]) and active protocols (7 days [20]; 8 days [10]; 10 days [62]). However, these data predominantly originate from moderately trained populations (i.e. Tier 2 [41]) who undertake modest weekly training loads. Therefore, a need exists to examine biomarker responses in elite endurance athletes (i.e. \geq Tier 4) undertaking a short-term HA intervention, in conjunction with representatively high training loads.

The aim of this study was to investigate the effect of a mixed-method HA protocol consisting of post-training passive heat stress, and low-intensity exercise in hot-humid conditions, on the physiological and health responses of six elite international triathletes. The present study occurred in the 3-weeks preceding the U23 World Triathlon Championships and no modifications were made to the planned external training loads. The intervention sought to maintain planned external training volume, and it was hypothesised that superimposing greater internal training load via HA would lead to heat adaptation, but not present undesirable physiological responses as individuals prepared for competition in heat stress.

Methods

Participants

Six international triathletes volunteered to participate in the study (3 male: 23 ± 1 years, 181 ± 8 cm, 67.0 ± 7.2 kg, sum of 7 skinfold 36.5 ± 12.8 mm, body fat percentage $16.8\% \pm 3.3\%$, $\dot{V}O_{2\text{max}}$ 69.7 ± 8.9 mL/kg/min; 3 female: 24 ± 5 years, 167 ± 6 cm, 55.8 ± 4.8 kg, 50.7 ± 4.1 mm, $22.1\% \pm 3.1\%$, 63.6 ± 2.8 mL/kg/min). Five athletes were preparing for 2022 U23 World Championships (Abu Dhabi, United Arab Emirates, air temperature: 25.1 °C, water temperature: 28.9 °C [66]). One athlete was preparing for the 2022 Asia Triathlon Cup (Ipoh, Malaysia air temperature: 27 °C, relative humidity: 79% [Integrated Surface Database, National Oceanic and Atmospheric Administration] [47]). No athletes had previously undertaken HA. Two female athletes reported the onset of menses eleven days after the study began and therefore commenced the HA protocol within the

luteal phase and ended it within the follicular phase. The third female athlete self-reported as amenorrhoeic. All participants had regularly competed in international and continental triathlon competitions for at least > 3 years. Athletes were based at the regional high-performance centre, where they typically trained for ~20–30 h per week (~6 h swimming, ~6 h running, ~10 h cycling, ~2 h strength, ~3 h recovery). Athletes provided written, informed consent and all procedures complied with the Declaration of Helsinki regarding human experimentation. Institutional ethical approval was not sought, as all measurements contained within the study were taken as part of routine physical monitoring, with training occurring as part of player's employment obligations [65].

Experimental Design

In the 3-weeks preceding the competitions, all participants undertook eight HA sessions (i.e. 'medium-term' HA: 5 active [running/cycling], 3 passive [post-exercise hot water immersion, HWI]). Exercise heat stress tests (HST) occurred at the start of the 1st, 5th and 8th HA sessions. Therefore, we report the effect of seven HA sessions (i.e. 'short-term' HA), although participants completed this 8th session, as part of competition preparation. HSTs involved 25 min of iso-speed running in the environmental chamber, at the start of the respective ~60 min HA sessions (HA session 1 = HST1, session 5 = HST2 and session 8 = HST3, thereby corresponding to the start, middle and end of the HA programme, respectively) (Fig. 1). Participants completed four HA sessions in week 1 and four sessions in week 2, before travelling to the venues 1-week in advance of competition (Fig. 1). During the World Championships, male athletes' finishing positions were between 14th and 40th, from 67 starters. Females finished between 18th and 30th, from 39 starters. Given the specifics of the U23 World Championships (i.e. age-restricted competition), it was not feasible to

recruit a representative control group. Likewise, a repeated-measures design would not account for this being the major event of the calendar around which training is periodised. Our sampling approach was therefore pragmatic, recruiting all individuals eligible for these competitions within the regional training centre. HA sessions were integrated into the existing training programme, such that planned external training load remained unaltered (Table 1). To achieve this, outdoor high-intensity training sessions were followed by 30–50 min HWI (water temperature ~39 °C), whilst low-intensity running/cycling sessions were relocated to a thermostatically controlled environmental chamber (running; 35 °C, 50% relative humidity [R.H.], cycling; 39 °C, 50% R.H., Welltech Instruments, Hong Kong China). Outdoor environmental conditions in Hong Kong during the intervention were between 19.7 and 28.5 °C (mean daily average air temperature 24.1 °C) and 80.5% R.H.

Heat Acclimation Protocol

Running speed during each 25 min HST was set to individual's 'easy' training zone (11.1 ± 0.7 km/h), representing an exercise intensity below the first lactate threshold (i.e. the first deviation from 'baseline' or 'steady-state' blood lactate concentration responses). Training zones were established during regular (normothermic) laboratory testing, which had occurred within the preceding 2 months for all athletes. During session 1, 5 min after the completion of HST1, athletes completed 6×80 m striding efforts on the treadmill, before resting in the chamber for the remaining time (~15 min). Athletes self-selected the speed, which was above 17.5 km/h for all. During sessions 5 and 8, participants completed the HST and then rested in the hot environment, exercising in 5 min blocks where required (i.e. to maintain T_{CORE} at 38.5 °C). Session 4 involved athletes completing 4×5 min intervals at an intensity corresponding to the second lactate threshold ('LT2'). Athletes had 90 s rest between running

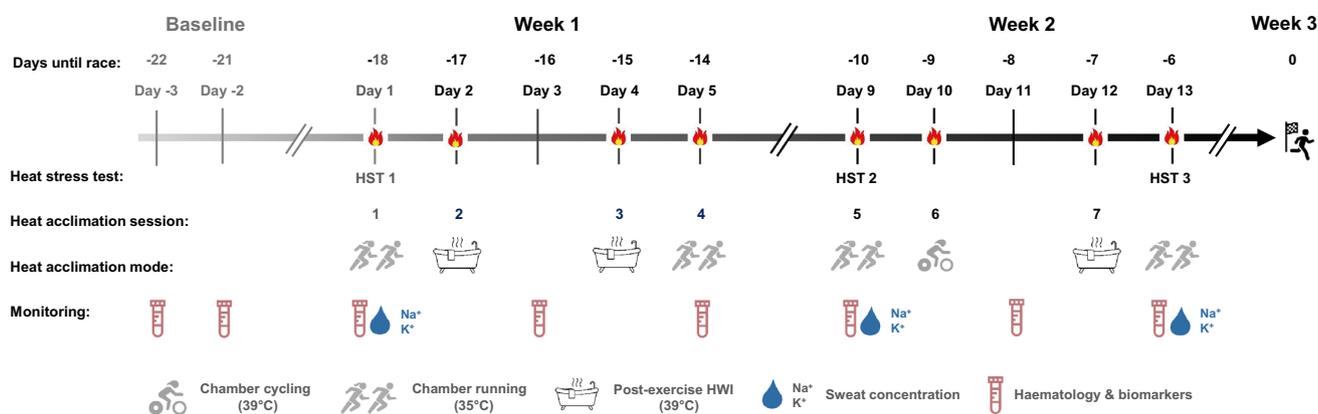


Fig. 1 Overview of HA training intervention. *HST* heat stress test, *HWI* hot water immersion

Table 1 Descriptive overview of training data preceding (weeks -3, -2, -1), and during two weeks of mixed-method heat acclimation training

Time point	Training mode	Weekly sessions (<i>n</i>)	Weekly duration (h)
Pre-3	Swim	4 ± 0	6 ± 1
	Bike	3 ± 0	5 ± 0
	Run	4 ± 1	4 ± 1
	Multi-mode	1 ± 0	2 ± 1
	Strength	2 ± 0	2 ± 0
	Weekly total	15 ± 1	21 ± 2
Pre-2	Swim	4 ± 0	4 ± 1
	Bike	4 ± 0	5 ± 0
	Run	4 ± 0	3 ± 1
	Multi-mode	1 ± 0	1 ± 0
	Strength	1 ± 0	1 ± 0
	Weekly total	15 ± 1	15 ± 3
Pre-1	Swim	5 ± 0	6 ± 1
	Bike	3 ± 0	6 ± 1
	Run	3 ± 1	3 ± 1
	Multi-mode	1 ± 0	2 ± 0
	Strength	1 ± 0	1 ± 0
	Weekly total	14 ± 1	18 ± 0
HA week 1	Swim	4 ± 1	6 ± 0
	Bike	2 ± 0	6 ± 3
	Run	3 ± 0	3 ± 0
	Multi-mode	2 ± 1	4 ± 2
	Strength	2 ± 0	1 ± 0
	Passive heat	2 ± 0	1 ± 0
	Weekly total	16 ± 1	22 ± 1
HA week 2	Swim	4 ± 0	4 ± 0
	Bike	3 ± 0	4 ± 0
	Run	5 ± 0	4 ± 0
	Strength	1 ± 0	1 ± 0
	Passive heat	1 ± 0	1 ± 0
	Weekly total	14 ± 1	16 ± 1

Non-specific recovery training sessions are not included

bouts and then rested in the hot environment (36.9 °C, 42% R.H.) until 50 min (i.e. 25 min active, 25 min passive). During session 6, participants completed the final 60 min of a scheduled 3 h ‘easy’ outdoor cycling session, in the environmental chamber (39.0 °C, 42% R.H.). For HA sessions 2 and 7, participants completed high-intensity interval training on an outdoor athletics track, and then undertook HWI (39.9 ± 0.92 °C) as soon as practically possible (< 5 min later). During HA session 3, participants completed an outdoor, cycling and running session (~ 2.5 h), before HWI afterwards. HWI duration progressed from 30 to 60 min during the HA intervention. This provided an initially

conservative, but progressive thermal stimulus through the training block.

Physiological Measures and Equipment

During all HA sessions, physiological and perceptual data were recorded at 5 min intervals. T_{CORE} was monitored using ingestible gastrointestinal temperature capsules (precision 0.1 °C, recording every min [BodyCap, Hérouville-Saint-Clair, France]). For morning sessions, capsules were provided the day before, with instructions to swallow it immediately before bed. Two hours prior to attending the laboratory, staff met with athletes to verify that the pill was still in the body. A second pill was activated and provided if the pill had been passed. This was necessary on five occasions, prior to the three HST tests (out of a possible 18). For afternoon training, the capsule was swallowed approximately 6–8 h in advance. Body temperature was also measured at the tympanic membrane (T_{TYP}) using an infrared thermometer (Thermoscan 7, Braun Instruments, Frankfurt, Germany). Measurements of T_{TYP} were recorded every 5 min throughout all sessions, aside of when participants were running. Hydration status was assessed 30 min prior to all chamber sessions, using a pocket refractometer (PAL-10S, Atago, USA). If dehydrated (urine specific gravity > 1.020), athletes consumed 500–600 mL of water or sports drink before the run. No athletes drank during the run. During all sessions, HR was monitored using OH1 arm sensors (Polar, Kempele, Finland), or via chest strap and watch (Garmin Forerunner 955, Kansas, USA). Individual athletes were consistent in their use of the same device and position across all HA sessions. Sweat sodium (Na^+) and potassium (K^+) concentrations were sampled in duplicate from the upper back and arm, during HST1, HST2 and HST3 (Fig. 1). Sterilised absorbent patches were placed on clean skin prior to entering the chamber. Samples were removed using sterile tweezers, with the experimenter wearing a surgical mask and gloves. Patches were placed into new plastic syringes, expelled onto the measurement surface and analysed immediately using portable LAQUAtwin Na-11 and K-11 pocket meters (Horiba, Kyoto, Japan).

Haematology and Biomarkers

Haematological monitoring occurred in duplicate prior to HA and then three times per week during HA (Fig. 1). Fingertip capillary blood samples were collected in 300- μl Lithium Heparin microvettes for creatine kinase and cortisol measurements. Samples were then centrifuged at 4700 r/min for 5 min, before being analysed using a biochemistry and immunoassay analyser (Abbott Architect ci4100, Abbott Illinois, USA). Further fingertip blood samples were collected to assess haematology parameters (urea, white blood

cell [WBC] count, red blood cell [RBC] count, haemoglobin [Hb] concentration, haematocrit [Hct], mean corpuscular volume [MCV], platelet count [PLT], testosterone concentration) in 200- μ L K3 EDTA microvettes. Samples were collected whilst participants were sat in an upright position, on eight occasions (Fig. 1). Whole blood samples were assessed using an automated haematology analyser (XT2000i, Sysmex, Kobe, Japan). Manufacturer stated reliability for all parameters were between 4.5% and 12.0%. All analyses were conducted within 60 min of the samples being collected. Blood samples were collected at standardised times, generally between 11 a.m. and 12 p.m., for all participants, at least 60 min after any training sessions. Estimated plasma volume (PV) changes were indicated using established formulae [12].

Perceptual Responses

RPE [4] and both thermal comfort and thermal sensation [14], were monitored every 5 min throughout all sessions. At the same time as haematological sampling were performed, athletes rated the following perceptual wellbeing responses; sleep quality, muscle soreness, psychological stress, training demands and appetite using a four-point scale, with 1 being the ‘best’ and 4 being the ‘worst’ they have previously experienced. Athletes were also asked to self-report sleep duration during this visit.

Statistical Analysis

Data are reported as mean \pm standard deviation (SD). Prior to statistical analysis, all data were checked for normality of distribution using Kolmogorov–Smirnov test and where this requirement was not met, non-parametric tests applied. Training data are reported descriptively. Changes in heat adaptation during the HST were analysed using repeated measures ANOVA (HST*time), where data at multiple time points were recorded, with Bonferroni post-hoc. Where singular data occurred, paired samples *t*-tests were used. Perceptual data during the HST were analysed using a non-parametric approach (Friedman’s ANOVA). Haematological and self-reported wellbeing data were analysed from two perspectives; (i) using the two ‘baseline’ values against the two ‘final’ values, using linear mixed models, (ii) using linear regression utilising all data points across the training period, to determine the relationship between each outcome variable over time. Partial eta-squared (η_p^2) was used as an index of the effect size within ANOVA models if significant differences were found. Analyses were conducted within SPSS statistical software (IBM Inc, USA), with $P < 0.05$. Where possible, outcome data were interpreted against absolute and relative reliability of the measure, to help interpret meaningfulness [28]. These calculations were;

Pearson’s correlation coefficient (*r*), typical error (TE: as calculated from the SD of the mean difference for each pair of trials using the formula: $TE = SD(\text{diff})/\sqrt{2}$) and, the limits of agreement (LoA: with 95% lower and upper confidence intervals [CI]) [3]. For haematological parameters, TE was calculated from the difference between the two baseline values, whilst duplicate measures of sweat concentration were used to calculate TE. Agreement from Pearson’s correlation coefficients were interpreted based upon: ‘Negligible’ = 0.0–0.3, ‘Low’ = 0.3–0.5, ‘Moderate’ = 0.5–0.7, ‘High’ = 0.7–0.9, and, ‘Very high’ = 0.9–1.0 [2]. Typical error and associated coefficient of variation (TE CV%) were categorised as: ‘Poor’ = > 10%, ‘Moderate’ = 5%–10%, and, ‘Good’ = < 5% [64]. On one occasion when T_{CORE} was not measured directly, T_{TYMP} was used with an individual-specific correction applied based upon the $T_{\text{CORE}}-T_{\text{TYMP}}$ regression equation obtained during that individual’s previous HSTs (on average 6 data points per athlete).

Results

Training Data

Training data throughout the study are reported in Table 1. Between sessions 1–7, the average total time whereby T_{CORE} exceeded 38.5 °C for each athlete was 116 ± 49 min (females 143 ± 38 min, males 88 ± 49 min). This equated to 17 ± 14 min per session. Across all sessions (i.e. including HST3 and following 30 min exposure which represents an eight HA session), the average total time with $T_{\text{CORE}} > 38.5$ °C for each athlete increased to 123 ± 57 min (females; 155 ± 50 min, males; 90 ± 49 min). However, the average duration > 38.5 °C per session remained similar (15 ± 14 min). Across all eight sessions, time > 38.5 °C was greatest in HWI sessions (total 75 ± 24 min, $n = 3$) versus active chamber sessions (total 48 ± 35 min, $n = 5$).

Heat Adaptation

Heat adaptation variables during the HST are displayed in Fig. 2. A reduction in exercising HR was observed within HST3 ($P = 0.01$, $\eta_p^2 = 0.64$), as compared to HST1 and HST2. However, no statistical changes in exercising T_{CORE} were seen across any HST ($P = 0.055$, $\eta_p^2 = 0.44$). Similarly, no differences were found in resting T_{CORE} (HST1 37.1 ± 0.2 °C, HST2 37.0 ± 0.4 °C, HST3 37.0 ± 0.2 °C, $P = 0.60$). Reduced Na^+ concentration at the arm ($P = 0.02$) and back ($P = 0.01$), were found during HST2, versus HST1, while sweat Na^+ concentration at the back was also lower in HST3 versus HST1 ($P = 0.04$). Increased sweat K^+ concentration occurred at the arm ($P = 0.01$) and back ($P < 0.01$) by HST3 versus HST2. Changes were also identified in K^+

concentration in HST3 versus HST1 (arm $P=0.04$, back $P<0.01$). Reliability data for sweat Na^+ and K^+ concentrations are displayed in Table 2. No differences were found in RPE, thermal sensation or thermal comfort across HSTs (all $P>0.05$).

Athlete Monitoring – Haematology, Biomarkers and Perceptual Wellbeing

No changes were found in creatine kinase, cortisol, urea, RBC count, Hb concentration, Hct, PV, PLT, testosterone concentration or the testosterone: cortisol ratio (all $P>0.05$, Fig. 3). A reduction in WBC count was detected between baseline ($7.3 \pm 1.8 \cdot 10^9/\text{L}$) and end ($6.0 \pm 1.0 \cdot 10^9/\text{L}$, $P=0.02$, $\eta_p^2=0.27$). An effect was also observed via regression ($P=0.01$, $\beta=-0.117$), although the mean difference ($-1.36 \cdot 10^9/\text{L}$) was smaller than the calculated TE ($1.44 \cdot 10^9/\text{L}$). An increase in MCV was found from baseline ($87.7 \pm 3.2 \text{ fL}$) to

end ($88.4 \pm 3.3 \text{ fL}$, $P=0.01$, $\eta_p^2=0.33$, regression: $P=0.047$, $\beta=0.042$). Perceptual wellbeing measures (i.e., sleep quality [pre 2.0 ± 0.8 , post 1.8 ± 0.6], muscle soreness [pre 2.3 ± 0.5 , post 2.2 ± 0.4], psychological stress [pre 1.7 ± 0.7 , post 1.9 ± 0.5], training demands [pre 1.6 ± 0.5 , post 1.8 ± 0.6], appetite [pre 1.6 ± 0.7 , post 1.8 ± 0.6] and sleep duration [pre $7:47 \pm 1:01 \text{ h}$, post $8:26 \pm 1:08 \text{ h}$]) all did not change during HA (all $P>0.05$).

During HA, there was stronger agreement between T_{CORE} and T_{TYMP} at rest (17 observations, $T_{\text{CORE}}=37.0 (0.3)^\circ\text{C}$, $T_{\text{TYMP}}=36.8 (0.4)^\circ\text{C}$, $r=0.62$, $\text{TE}=0.20^\circ\text{C}$, $\text{LoA}=0.55^\circ\text{C}$ [-0.34 to 0.76]) and immediately after treadmill running in the hot environment (32 observations, $T_{\text{CORE}}=38.3 (0.4)^\circ\text{C}$, $T_{\text{TYMP}}=37.5 (0.4)^\circ\text{C}$, $r=0.60$, $\text{TE}=0.26^\circ\text{C}$, $\text{LoA}=0.72^\circ\text{C}$ [0.04 – 1.48]), than during HWI (90 observations, $T_{\text{CORE}}=38.6 (0.6)^\circ\text{C}$, $T_{\text{TYMP}}=38.2 (0.9)^\circ\text{C}$, $r=0.61$, $\text{TE}=0.50^\circ\text{C}$, $\text{LoA}=1.37^\circ\text{C}$ [-0.99 to 1.76]). Overall, the agreement across all situations was relatively strong (139

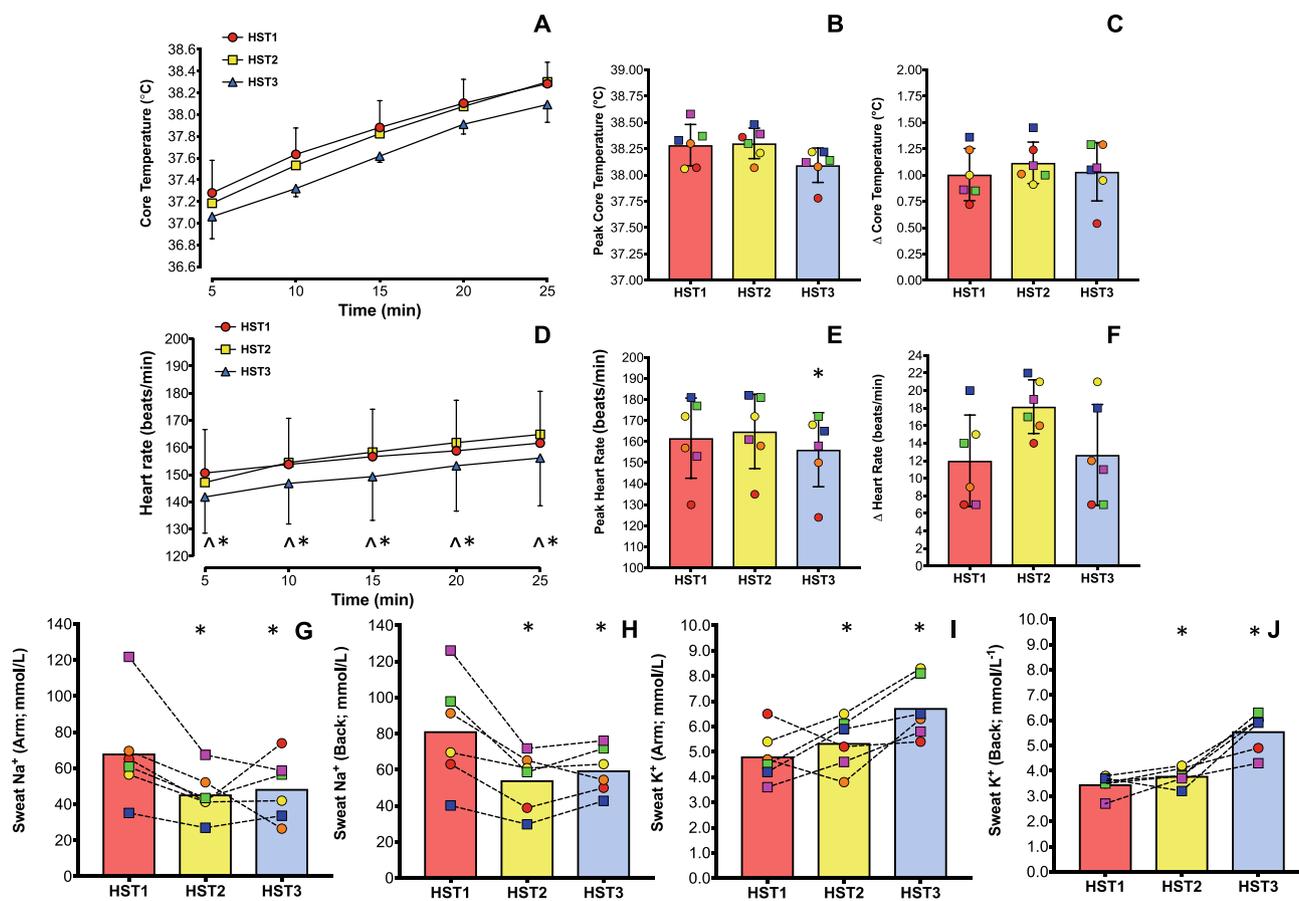


Fig. 2 Panel of physiological markers during the HA training intervention. **A** core temperature over time during each HST, **B** peak core temperature at the end of each HST, **C** change in core temperature during each HST, **D** heart rate over time during each HST, **E** peak heart rate at the end of each HST, **F** change in heart rate during each HST, **G** arm sweat sodium concentration, **H** back sweat sodium concentration, **I** arm sweat potassium concentration, **J** back

sweat potassium concentration. Coloured bar represents group mean (red=HST1, yellow=HST2, blue=HST3). Coloured shapes represent individual athletes, circles represent male athletes, squares represent female athletes. * denotes a difference from HST1 within datapoint ($P<0.05$), ^ denotes difference from HST2 within datapoint ($P<0.05$)

Table 2 Reliability of sweat concentration measurement

Variable	Location	<i>n</i>	Mean bias (SD)	<i>r</i>	TE (mmol/L)	TE (CV%)	LoA (mmol/L)
Sweat Na ⁺	Arm	16	-2.1 (5.6)	0.97	3.9	7.5	10.9 (-13.0 to 9.0)
	Back	19	5.1 (6.7)	0.96	4.8	7.6	13.2 (-8.0 to 18.0)
Sweat K ⁺	Arm	16	-0.6 (0.9)	0.82	0.6	11.0	1.7 (-2.3 to 1.2)
	Back	19	0.0 (0.4)	0.92	0.3	7.3	0.9 (-0.9 to 0.8)

Na⁺ sodium, K⁺ potassium, TE typical error, LoA limits of agreement

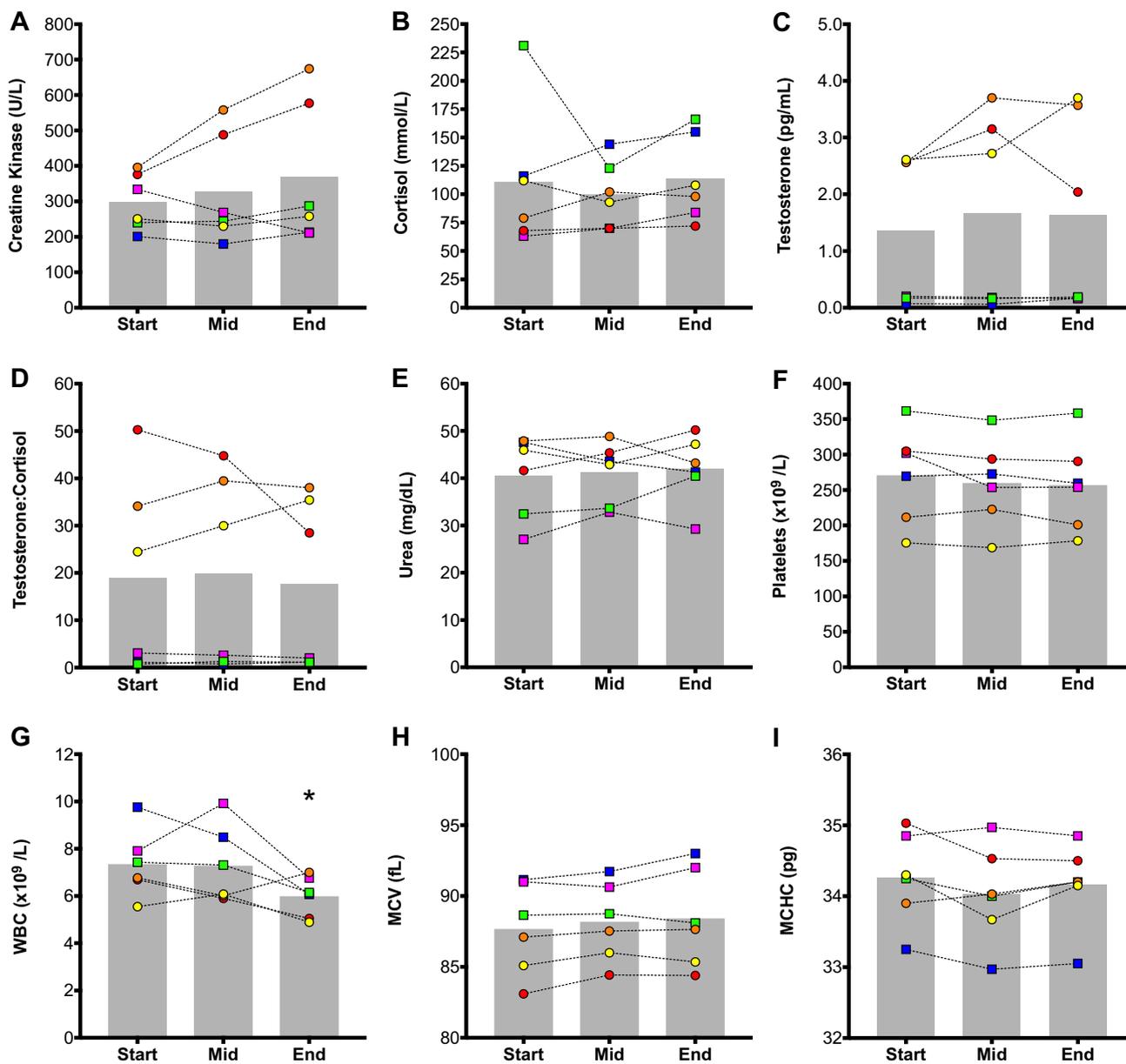


Fig. 3 Panel of measured biomarkers during the HA training intervention. **A** creatine kinase, **B** cortisol, **C** testosterone, **D** testosterone to cortisol ratio, **E** urea, **F** platelet count, **G** white blood cell count (WBC), **H** mean corpuscular volume (MCV), **I** mean corpuscular haemoglobin concentration (MCHC). *Start* average of 1st and 2nd measurement (Day-3 and -2), *Mid* average of 4th and 5th measure-

ment (Day 3 and 5), *End* average of 9th and 10th measurement (Day 7 and 9). Grey bars represent group mean. Coloured shapes represent individual athletes, circles represent male athletes, squares represent female athletes. * denotes difference within End timepoint compared to Start and Mid ($P < 0.05$)

observations, $T_{\text{CORE}} = 38.3 (0.8)^\circ\text{C}$, $T_{\text{TYMP}} = 38.9 (0.9)^\circ\text{C}$, $r = 0.73$, $\text{TE} = 0.44^\circ\text{C}$, $\text{LoA} = 1.22^\circ\text{C}$ [-0.77 to 1.67]).

Discussion

This study investigated the physiological responses and health effects of a mixed-method HA programme, integrated into the preparation of six elite triathletes preparing for the U23 World Championships ($n = 5$) and the Asia Triathlon Cup ($n = 1$). Planned external training loads remained unchanged throughout the lead-in to the Championships. Evidence of partial heat adaptation was observed by way of reduced exercising HR during HST3 (Fig. 2) and lower sweat Na^+ concentrations from HST2 (Fig. 3). Exercising T_{CORE} was lower in four out of six athletes in HST3, which also aligns with partial adaptation (Fig. 2). Concurrently, no adverse effects on haematological immune and health markers were observed (Fig. 3), nor were detrimental athlete perceptual responses identified following the superimposition of heat stress to planned training. This indicates the addition of HA to an existing training programme was well-tolerated and provides meaningful change in a population of elite athletes. These partial adaptations serve as a reference for designing HA interventions in other similarly high-demanding elite endurance training programmes.

The mean reduction in exercising HR (~ -6 beats/min) appears highly meaningful in this population [58] and is comparable to prior research implementing medium-term HA [56]. In the context of elite endurance athletes, such a reduction represented the breadth of a HR training zone for most of this cohort (e.g. training zone of 154–160 beats/min). This has the potential to change the interpretation of internal training load, before and after HA (i.e. run in HR zone 3, is now in zone 2). For example, quantitative training load metrics based on HR data, such as TRIMP [7] or acute: chronic ratio [46] may be altered following HA. Therefore, the consideration of both internal and external load appears pertinent, around HA interventions [26, 51]. Indeed, an alternative emerging approach may be the internal: external training load ratio, providing greater flexibility in training session content, so long as there is consistency in the training monitoring metrics [51]. Reduced exercise HR following HA is usually attributed to an expansion of blood PV, although changes in cardiac contractility and sudomotor function may also contribute [53]. However, we did not observe any changes in PV. Similarly, no statistically significant reduction in T_{CORE} was found at the group level. However, the group mean reduced by 0.22°C (at rest) and 0.20°C (exercising, at end of HST) between HST1 and HST3. A physiologically meaningful reduction in exercising or resting T_{CORE} following HA has been proposed as being $>0.2^\circ\text{C}$ [56, 62]. Interestingly, four athletes displayed

a reduction in exercising T_{CORE} of this magnitude or greater though it is acknowledged that the change in menstrual cycle phase across two participants during the analysis timeframe means typical deductive reasoning as to the effects of HA on T_{CORE} reduction is not possible. We also found positive reductions in sweat Na^+ concentration following HST2 (arm: -23 mmol/L, back: ~ -27 mmol/L), which were maintained at HST3 (arm: -20 mmol/L, back: -22 mmol/L) compared to HST1. This change in sweat Na^+ retention during HA is in agreement with previous literature (-20 mmol/L) [9, 62, 63] and exceeded our measurement error (Table 2), thus appears meaningful and has the potential to attenuate the negative implications of reduced systemic sodium during competition. These changes are indicative of improved reabsorption ability of the eccrine sweat glands [5]. A modest increase was observed in sweat K^+ concentration (HST2 arm: $+0.6$ mmol/L, back: $+0.4$ mmol/L, HST3 arm: $+1.9$ mmol/L, back: $+2.1$ mmol/L). This may be linked to a reduction in sweat Na^+ , but has also been suggested to reflect the accumulation of K^+ within the sweat patch following an increased local sweat rate [36]. As we did not measure sweat rate, we could not verify this mechanism.

In the context of the haematological and biomarker dataset (Fig. 3), changes following the superimposition of heat to the planned training would have indicated a systemic perturbation that may represent ‘overtraining’ or compromised immune status [39]. An 18% reduction in WBC was observed. Leukopenia can be associated with an increased risk of infection in athletes undertaking high training loads [19], however all athlete’s final WBC measures ($\sim 6.0 \times 10^9/\text{L}$) remained within a normal range for athletes ($5.7\text{--}7.4 \times 10^9/\text{L}$) [24]. Accordingly, the superimposition of heat to the planned training was not considered to negatively influence WBC. Across the intervention, no significant, nor meaningful changes were observed in the other biomarkers assessed e.g. urea, RBC count, MCV, PLT, cortisol and testosterone. This supports the conclusion that the superimposition of heat did not push the athletes towards a state of ‘overtraining’ or compromise these selected markers, which can characterise immune status. For example, no observable reduction in testosterone, nor an increased testosterone: cortisol ratio, which may occur in response to training stress [39]. In agreement with our hypothesis, the mixed-method HA programme does not therefore appear to have compromised immune function in these athletes. Taken together, these real-world, elite athlete haematological and biomarker training data align with laboratory data from active and passive HA studies collected on sub-elite and active individuals [10, 33, 58, 62]. Overall, the addition of heat stress to an existing training programme resulted in encouraging, albeit partial heat adaptation, within international endurance athletes.

The individual responses that we observed could be reflective of differences in HA state between individuals

[61]. Indeed, athletes may already operate at a greater HA state than untrained individuals and require a greater HA stimulus to reduce T_{CORE} e.g., running at a higher intensity than implemented during HSTs and/or greater total volume. We also acknowledge the potential for measurement bias in HST1, given one of these two participants who did not demonstrate a reduced T_{CORE} took a second pill earlier in the morning having passed the original through the night. As such, fluids consumed prior to the HST may still influence the derived temperature [57].

Inducing heat adaptation without modifying prescribed training load represents an exciting development in HA prescription. Our HA session volume (4 sessions per week, 7 prior to HST3) may be considered modest, compared with related literature (13 sessions in 18 days [52]; 8 sessions in 8 days [55]) or laboratory-derived guidelines (> 6 days [16]). Similarly, the duration of HA sessions (max 60 min) is considerably shorter than many controlled hyperthermia protocols (90 min [17]) and below the historical recommendation to attain 60 min above a ‘target’ T_{CORE} [13]. As a result, average time > 38.5 °C was 15–17 min, which compares with 63 min in a prior short-term laboratory based HA study with trained runners [31]. It is important to note that the mixed-method approach we adopted was designed to support concurrent training aims. Therefore, session design and external training intensities were not prescribed solely targeting specific physiological responses (e.g. exceed 38.5 °C as soon as possible [18]). Rather, the external training demands of sessions were predetermined, as the coach sought to train towards multiple objectives across the three disciplines. Adding a further stressor to a high-volume (> 20 h) endurance training programme logically raises the risk of training maladaptation, such as overreaching. Thus, we adopted a pragmatic and conservative approach to progressively introduce the HA stimulus, avoiding high-intensity training in the heat and utilising initially shorter (30 min) HWI sessions. Interestingly, our shorter active HA sessions and lower weekly total heat exposure than prevailing HA guidelines [16, 48, 56], are comparable to that which has recently been successfully applied over a chronic timeline (5 × 50 min over 5 weeks) in highly trained endurance athletes [38]. Given the modest nature of our HA prescription, these data serve as a reference for short-term HA that achieves partial heat adaptation in elite endurance athletes across 2-weeks, without compromising biomarker status or overreaching. The absence of negative interactions with ongoing training, wellbeing and immune markers, indicates merit in extending our progressive prescription model across a longer timescale.

Our data demonstrate the relevance of a practitioner ‘toolbox’ approach, whereby partial HA can be achieved through a variety of pragmatic techniques (active and passive), so long as the primary potentiating stimuli are achieved (i.e.

targeting prolonged, elevated T_{CORE} and profuse sweating) [16]. Interestingly, time $T_{\text{CORE}} > 38.5$ °C was greatest in HWI sessions (total 75 ± 24 min) compared to active chamber sessions (total 48 ± 35 min). This is a consequence of the HWI sessions following outdoor high-intensity training. Thus, T_{CORE} is already elevated and HWI primarily served to maintain thermal strain. Conversely, active sessions were predominantly completed from a rested state, where a larger heat gain was required to achieve this level of thermal stress. These are important observations helping to guide integrated HA prescription into high-volume training programmes comprising 15–20 sessions per week (Table 1). Our findings have relevance for the application of HA across a number of endurance sports, and provide encouraging evidence for other sports such as team-sports, whereby heat stress may not be a primary determinant of fatigue, but a performance decrement is observed relative to normothermic conditions [32].

Although we monitored heat adaptation across predefined training sessions, this resulted in heat adaptation being assessed in the low-intensity domain during modest levels of thermal and cardiovascular stress, potentially masking the magnitude of adaptations. This approach was necessitated by a need for conservative exercise prescription, given these athletes had not undertaken HA previously and were preparing for important upcoming competitions. Therefore, planned external training loads remained unchanged during our intervention, with pre-existing training sessions relocated to the hot environment. Enhancing thermal strain during future HA interventions is now possible for this cohort with a view to increasing the magnitude of adaptation. Hydration variability within the range described as euhydrated and variability of blood sampling times (i.e. ± 60 min) may also conceal true magnitude of some haematological changes. For example, some athletes may not have been sufficiently rehydrated following training, which could help explain the absence of a detected change in PV. Nevertheless, consistency of sampling in such a busy training week is difficult to achieve. Thus, the true magnitude of adaptation may have been greater than we report, given the challenges of control in an applied environment. Practitioners should also note the circumstance with poorest agreement between T_{CORE} and T_{TYMP} was during passive heating. This is an observation which has recently been made when using a portable steam sauna [64], whereby similar to HWI, an external source of heat interacts across the majority of the skin surface area excluding the measurement site of T_{TYMP} . Therefore, we advocate the development of individual- and situation-specific regression equations (e.g. immediately post exercise only or high/low levels of heat stress), given resting data demonstrated stronger agreement than post-exercise or during HWI.

Conclusion

Superimposing mixed-method HA on top of an existing training programme was well-tolerated by elite endurance athletes and provided partial heat adaptation in this population. This intervention serves as a reference for pragmatic HA strategies that other elite endurance athletes may utilise, when undertaking similarly highly demanding training programmes. Heat adaptation can be achieved through numerous approaches that are complimentary and not dichotomous to concurrent training.

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Data Availability Data may be made available upon request to the corresponding author.

Declaration

Conflict of interest On behalf of all authors, the corresponding author states that there is no conflict of interest.

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