

Title:

Short term heat acclimation improves the determinants of endurance performance and 5,000 m running performance in the heat.

Running title:

Heat acclimation and endurance running.

Carl James^{1,2}, Alan Richardson¹, Peter Watt¹, Ashley Willmott¹, Oliver Gibson³ and Neil Maxwell¹

Author affiliations:

¹ Environmental Extremes Laboratory, Centre for Sport and Exercise Science and Medicine (SESAME), University of Brighton, Eastbourne, UK.

² National Sports Institute of Malaysia, Institut Sukan Negara, Bukit Jalil Stadium, Kuala Lumpur, Malaysia.

³ Centre for Human Performance, Exercise and Rehabilitation (CHPER), Brunel University London, UK.

Dr Carl James

Dr Alan Richardson

Dr Peter Watt

Mr Ashley Willmott

Dr Oliver Gibson

Dr Neil Maxwell

carlalexanderjames@gmail.com (corresponding author)

A.J.Richardson@brighton.ac.uk

P.Watt@brighton.ac.uk

A.Willmott@brighton.ac.uk

Oliver.gibson@brunel.ac.uk

N.Maxwell@brighton.ac.uk

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Abstract

This study investigated the effect of 5 days controlled hyperthermia heat acclimation (STHA) on the determinants of endurance performance and 5 km performance in runners, relative to the impairment afforded by moderate heat stress. A control group (CON), matched for total work and power output ($2.7 \text{ W}\cdot\text{kg}^{-1}$), differentiated thermal and exercise contributions of STHA on exercise performance. Seventeen participants (10 STHA, 7 CON) completed graded exercise tests (GXT) in cool (13°C , 50% RH, pre training) and hot conditions (32°C , 60% RH, pre and post training), as well as 5 km time trials (TT) in the heat, pre and post training. STHA reduced resting ($p=0.01$) and exercising ($p=0.04$) T_{CORE} alongside a smaller change in thermal sensation ($p=0.04$). Both groups improved the lactate threshold (LT, $p=0.021$), lactate turnpoint (LTP, $p=0.005$) and $\dot{V}\text{O}_{2\text{max}}$ ($p=0.031$) similarly. Statistical differences between training methods were observed in TT performance (STHA $-6.2[5.5]\%$, CON; $-0.6[1.7]\%$, $p=0.029$) and total running time during the GXT (STHA; $+20.8[12.7]\%$, CON; $+9.8[1.2]\%$, $p=0.006$). There were large mean differences in change in $\dot{V}\text{O}_{2\text{max}}$ between STHA $+4.0(2.2) \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ($7.3[4.0]\%$) and CON $+1.9(3.7)\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ($3.8[7.2]\%$). Running economy deteriorated following both training programmes ($p=0.008$). Similarly, RE was impaired in the cool GXT, relative to the hot GXT ($p=0.004$). STHA improved endurance running performance in comparison to work matched normothermic training, despite equality of adaptation for typical determinants of performance (LT, LTP, $\dot{V}\text{O}_{2\text{max}}$). Accordingly, these data highlight the ergogenic effect of STHA, potentially via greater improvements in $\dot{V}\text{O}_{2\text{max}}$ and specific thermoregulatory and associated thermal perception adaptations absent in normothermic training.

Key words

Heat acclimation; hyperthermia; endurance; $\dot{V}\text{O}_{2\text{max}}$; thermoregulation

Introduction

A deleterious effect of heat stress on endurance performance is well established (Galloway & Maughan 1997). This impairment extends to the primary physiological determinants of endurance performance; $\dot{V}O_{2max}$ (Sawka et al. 1985) and blood lactate indices (Lorenzo et al. 2011), whilst the influence on running economy (RE) is contentious (Saunders et al. 2004). Considerable, recent evidence documents the effectiveness of transient thermal adaptations, proffered through heat acclimation (HA) training, in alleviating physiological and thermal strain (Gibson et al. 2015a; Mee et al. 2015; Willmott et al. 2016), as well as the ergogenic potential for endurance performance in the heat (Lorenzo et al. 2010; Garrett et al. 2012; Racinais et al. 2015). Appropriately, given the multi-dimensional nature of heat strain (Nybo et al. 2014), the HA phenotype arises from physiological adaptation across multiple systems, notably pertaining to; sudomotor function (Lorenzo & Minson 2010), cardiovascular stability (Rowell et al. 1967), skeletal muscle metabolism (Febbraio et al. 1994), cutaneous blood flow (Lorenzo & Minson 2010), central thermoregulatory control (Buono et al. 1998) and cellular function (McClung et al. 2008). In turn, the most widely observed and prominent HA adaptations include decreased resting and exercising, core (T_{CORE}) and skin (T_{SKIN}) temperatures, alongside a reduction in exercising heart rate (HR) in the heat, which likely arises through a combination of increases in plasma volume (PV), cardiac contractility and sudomotor function (Sawka et al. 2011; Périard et al. 2015). Collectively, such adaptations ameliorate the deleterious cardiovascular strain that arises from peripheral vasodilation in the hyperthermic individual, which forms a predominant limitation during maximal aerobic exercise in the heat (González-Alonso et al. 2003; Périard et al. 2011). Improved perception of both heat strain and exertion during heat stress may be observed following HA (Gonzalez & Gagge 1976), alluding to a role for behavioural alterations to improve free-paced, sub-maximal exercise in the heat (Flouris & Schlader 2015). Whilst the majority of adaptations typically occur after ~10 daily exertional heat exposures (Pandolf 1998; Garrett et al. 2009), the rapid induction of ~75% of adaptations after 4–6 days (Armstrong & Maresh 1991) helps explain the prominence of time-efficient short-term

acclimation strategies (STHA; ≤ 7 days) for athletes who will compete in the heat (Garrett et al. 2011; Chalmers et al. 2014).

In addition to improving endurance time trial performance in the heat in sports such as cycling (Lorenzo et al. 2010; Racinais et al. 2015) and rowing (Garrett et al. 2012), HA may enhance physiological determinants of endurance performance such as $\dot{V}O_{2max}$ and the lactate turnpoint (Lorenzo et al. 2010, Lorenzo et al. 2011). However, the effects of both HA and environmental heat stress on the determinants of endurance performance model (Bassett & Howley 2000) is not well documented. This model is widely applied to monitor and/or predict endurance performance across a range of sports and athletes whereby $\dot{V}O_{2max}$ represents the upper limit of aerobic metabolism, beneath which the lactate turnpoint and running economy interact to determine the sustainable exercise intensity (Coyle 1995; Bassett & Howley 2000; Jones & Carter 2000; McLaughlin et al. 2010). Changes in $\dot{V}O_{2max}$ may be highly influential on endurance performance in the heat, with Racinais et al. (2015) recently demonstrating an initial reduction and subsequent improvements in both $\dot{V}O_{2max}$ and time trial performance, as individual's arrive and then adapt to a hot environment. The authors suggested this may reflect the maintenance of a relative exercise intensity ($\% \dot{V}O_{2max}$) during endurance exercise in the heat following adaptation.

A lack of consistency within HA regimes exists, with different durations (short, medium, long term), environmental conditions and models (fixed intensity, self-regulated, controlled hyperthermia) adopted, making it difficult to extrapolate results between studies (Tyler et al. 2016). The controlled hyperthermia approach appears well supported for inducing thermal adaptations (Patterson et al. 2004; Garrett et al. 2014; Gibson et al. 2015b) and achieves this through a smaller exercise volume than fixed-intensity, constant work training (Gibson et al. 2015a). Despite the efficacy of HA for improving cycling performance under heat stress, there is a dearth of research concerning running time trials and particularly following STHA. Compared with cycling, running elicits a higher metabolic heat production (MHP), reduced convective cooling and potentially greater

individual variability of economy of movement (Millet et al. 2009) therefore, the effects of both heat stress and STHA on running performance may differ to cycling. Indeed, given the propensity for heightened heat strain in running (Chan et al. 2008), larger effects following HA may be apparent in runners.

Exercise training within HA may promote adaptation independently of thermal strain, making it hard to differentiate the precise roles of thermal and exertional strain during HA training. Therefore, matched exercise training is important in order to optimise HA practices, indicating how exercise intensity and/or passive thermal exposures should be applied. Furthermore, training using the same exercise modality to the performance trial may simply result in improvements arising from an increased training volume. Therefore, this study investigated the effect of a cycling, controlled hyperthermia, STHA programme on the determinants of endurance performance and TT performance in runners, in comparison to a work matched control group, as well as quantifying the decrement to runners elicited by moderate heat stress. It was hypothesised STHA would elicit larger improvements in the determinants of endurance performance and 5 km time trial performance than normothermic training.

Methods

Participants

Seventeen amateur runners volunteered as participants (Table 1). Ten participants (9 male, 1 female) completed STHA, whilst seven (male) participants completed control training (CON). All participants trained at least three times per week, with mean (\pm SD) recent 5 km performances of 20:51 (1:41) in the STHA group and 19:48 (1:39) in CON within the previous month. Testing occurred in the UK Spring, therefore participants were not heat acclimated and were entering the competition season. The female participant completed pre-tests and training during the follicular phase of the menstrual cycle, with post-tests during the first 5 days of luteal phase. Each participant provided written informed consent and institutional ethical approval was issued in accordance with the Declaration of Helsinki (2013). Participants avoided intense exercise, alcohol and caffeine for 48 hours before testing and arrived hydrated, verified through urine analysis using a handheld osmometer ($<700 \text{ mOsmol.kg}^{-1} \text{ H}_2\text{O}$, Osmocheck™ Pocket, Vitech Scientific Ltd, UK) and a refractometer (<1.020 , Specific Gravity Refractometer Model 32, Atago; USA) in accordance with Sawka et al. (2007). Participants completed a 24-hour food diary prior to each test and indicated sleeping hours, motivation, muscle soreness and stress on 5-point Likert scales upon arrival. Finally, participants were asked to prepare for each trial as a competition and suspend their normal training regime for the duration of the study (~ 16 days), to ensure training load could be accurately quantified and sufficient recovery between tests would be achieved.

Table 1 here

Experimental design

A mixed model, independent groups design was adopted, with participants assigned to either STHA or CON training. The CON group were individually matched against the STHA group for anthropometry, $\dot{V}O_{2\max}$ and recent running performance, in order to elicit similar relative and

absolute training intensities. All participants completed instrumented familiarisations of both the graded exercise test (GXT) and 5 km time trial (TT), separated by at least 7 days later to control against the induction of heat adaptations (Barnett & Maughan 1993). Participants then completed a normothermic GXT (13°C, 50% RH), a TT in the heat (32°C, 60% RH) and a hot GXT (32°C, 60% RH). All trials separated by 48 hours. Following five consecutive days of training, participants repeated the hot GXT and hot TT. All pre and post trials were completed within 1 week of training, a period in which adaptation has been shown to be maintained following a similar STHA protocol (Garrett et al. 2009).

Graded exercise test

During the familiarisation visit, stature, body mass and a skin fold assessment (Harpenden, Burgess Hill, UK) across iliac crest, subscapular, triceps and biceps (Durnin & Womersley 1974) were recorded. All trials were conducted within a thermostatically controlled environmental chamber (WatFlow control system TISS, Hampshire, UK), with conditions verified using a heat stress meter (HT30, Extech Instruments, USA).

In the STHA group, markers of heat acclimation, in accordance with recent literature (Sawka et al. 2011; Périard et al. 2015), were assessed from resting and exercise responses during GXTs in hot conditions, pre and post training. Plasma volume change was estimated from haematocrit and haemoglobin concentration (Dill & Costill 1974). Resting responses were assessed after 30 min of lying in the supine position, in hot conditions.

The graded exercise test was split into two parts; GXT 1 and GXT 2, as described by Jones (2006). GXT 1 was a discontinuous, submaximal incremental speed protocol involving 3 minutes of exercise per stage and 1 min for capillary blood sampling. Each participant completed a minimum of six stages, using speed increments of 1 km.h⁻¹ on a motorised treadmill (Woodway ELG2, Weil am Rhein, Germany). Following GXT 1, participants rested in the hot environment for 10 min before GXT

2, an incremental gradient protocol to volitional exhaustion. GXT 2 began at a speed $2 \text{ km}\cdot\text{h}^{-1}$ below the previous final speed with gradient increasing by 1% each min. Participants were not permitted to drink and were blinded to all feedback.

Pre and post exercise nude body mass permitted sweat loss estimation, whilst T_{CORE} was measured using disposable rectal probes (Henleys Medical, UK), inserted 10 cm beyond the anal sphincter and connected to a meter logger (Model 401, Yellow Springs Instruments, Missouri, USA). Telemetry thermistors (U-Type connected to Gen II GD38 transmitter, Eltek, UK) were attached to the pectoralis major, biceps brachii, rectus femoris and gastrocnemius with data transmitted wirelessly to a datalogger (RX250AL 1000 series Wireless Squirrel Logger, Eltek) for measuring T_{SKIN} , as per James et al. (2014). Heart rate was monitored continuously using a Polar 810i heart rate monitor (Kempele, Finland).

During the GXTs, HR, T_{CORE} , T_{SKIN} , rating of perceived exertion (RPE, Borg, 1998) and thermal sensation (0=unbearably cold to 8=unbearably hot, Gagge et al. 1969) were noted at the end of each 3 min stage. Running speeds at 2 and 4 $\text{mmol}\cdot\text{l}^{-1}$ were calculated by solving the polynomial regression equation for blood lactate concentration versus speed at 2 and 4 $\text{mmol}\cdot\text{l}^{-1}$, denoting the lactate threshold (LT) and lactate turnpoint (LTP) respectively, following the methods outlined by Saunders & Green (2013). Fingertip blood samples were analysed immediately (YSI 2300 analyser, YSI, Ohio, USA). Ventilatory gases were measured using a Metalyzer 3B analyser (Cortex, Leipzig, Germany), with the two 30 s averages from the final min of each stage used for running economy (RE) and the respiratory exchange ratio (RER). Average RE ($\text{mL O}_2\cdot\text{kg}^{-1}\cdot\text{km}^{-1}$) was calculated across the first five exercise stages. During GXT 2, the highest 30 s moving average represented $\dot{V}\text{O}_{2\text{max}}$. Velocity at $\dot{V}\text{O}_{2\text{max}}$ ($v\dot{V}\text{O}_{2\text{max}}$) was calculated by multiplying $\dot{V}\text{O}_{2\text{max}}$ ($\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) by 60 and dividing by the average RE (Jones 2006). Mean T_{SKIN} and the Physiological Strain Index (PSI) were calculated following the methods outlined by Ramanathan (1964) and Moran et al. (1998), respectively.

Time trial

Participants completed one treadmill TT familiarisation in the heat, as recommended for trained runners (Laursen et al. 2007). For experimental TTs pre and post the training week, following a 10 min rest phase, participants completed a 5 min, self-selected warm-up that was consistent across trials. Standardised instructions were given at the start of the trial and nothing thereafter; 'give your all', 'pace yourself throughout the trial' and 'adjust speed as you see fit' as per Stannard et al. (2011). Participants began all trials with the treadmill belt set to their average speed from the familiarisation and were free to adjust speed immediately and *ab libitum* thereafter (increment 0.2 km.h⁻¹). The treadmill gradient was fixed at 1% to reflect the additional energy expenditure experienced during outdoor running (Jones & Doust 1996). Distance was displayed continuously, however participants were blinded to all other feedback and did not drink during the trials.

Training

The STHA group completed five, 90 min daily training sessions in the heat (36.6 [0.8]°C, 59 [9]% relative humidity [RH]) using controlled hyperthermia and permissive dehydration (Garrett et al. 2014). Participants cycled (Monark, e724, Vansbro, Sweden), with power output initially prescribed relative to body mass at 2.7 W.kg⁻¹ (Gibson et al. 2016) in 5 min blocks and thereafter adjusted to the maximum tolerable power in order to achieve the target T_{CORE} (38.5°C) within 30 min. Where participants could not maintain 2.7 W.kg⁻¹ across consecutive days, such as those who did not habitually cycle, the required power output was reduced and thus may be better characterised as 'maximum tolerable'. Upon attaining a T_{CORE} of 38.5°C, cycling exercise was again completed in 5 min blocks of fixed intensity, as necessary in order to ensure T_{CORE} remained above 38.5°C during the following 60 min, as performed previously in our laboratory (Mee et al. 2015; Gibson et al. 2015a). The power output during this 60 min period was chosen by the participant.

The novel prescription of exercise based on power output, relative to body mass, as opposed to % $\dot{V}O_{2max}$ (Nielsen et al. 1993; Lorenzo et al. 2010; Castle et al. 2011), removes the necessity for an

initial cycling $\dot{V}O_{2max}$ test and demonstrates a greater relationship with increased T_{CORE} than other training prescription variables (Gibson et al. 2016). Furthermore, controlled hyperthermia heat acclimation maintains thermal strain as adaptation occurs (Taylor 2014) and mitigates systematic differences in MHP that may present across individuals of varying $\dot{V}O_{2max}$ (Jay et al. 2011). Training occurred at the same time of day, with the majority in the morning (07:00-10:00 h) and one participant in the evening (18:00-20:00 h). No fluid intake was permitted throughout the training sessions (Garrett et al. 2014; Neal et al. 2015).

The purpose of the CON group was to understand the contributing roles of exertional and thermal strain as independent stimuli for subsequent aerobic performance in the heat, rather than assess HA state, given the volume of evidence supporting the efficacy of this type of programme for establishing HA (Garrett et al. 2012; Garrett et al. 2014; Gibson et al. 2015c; Mee et al. 2015). Therefore, individually matched for total work completed and power output against the STHA group, the CON group completed five consecutive days of training in temperate laboratory conditions (20.0 [0.9]°C, 43 [7]% RH). As total work was matched between groups, training typically lasted ~40 min, reflecting the initial 30 min of exercise during STHA training and two further 5 min blocks during the subsequent 60 min. The CON group used the same equipment and procedures as in STHA, although cooling fans were permitted, in addition to the reduced environmental temperature.

Statistical analyses

All outcome variables were assessed for normality and sphericity prior to further analysis. Heat acclimation criteria for the STHA group were analysed using paired samples T-tests or one-way repeated-measures ANOVA, with Bonferroni correction applied during post-hoc analysis where significant differences were identified. RM ANOVA was used to analyse variables that have repeated measures over time within both the pre HA test and the post HA test, such as T_{SKIN} or HR. Where variables provide a single value, such as resting HR or sweat loss, paired samples t-tests were used. Physiological and performance data from the GXTs and TTs were analysed using mixed-model, 2-way

ANOVA (Group*Time), with Bonferroni t-test. Data from STHA and CON were pooled for comparisons between GXTs in cool and hot environments, to quantify the effect of heat stress. Data were analysed using SPSS (Version 21, SPSS Inc, Illinois, USA) with statistical significance set at $p < 0.05$ and data presented as means and standard deviation (\pm SD). Effect sizes for main effects and interaction effects are presented as partial eta squared (partial η^2), differences between related and independent samples were evaluated through Cohen's d_{av} (d_{av}) and Cohen's d (d) respectively, in accordance with Lakens (2013).

Results

Participants

Self-reported motivation, muscle soreness and stress responses did not differ between groups or trials ($p > 0.05$). One participant from STHA was unable to complete the post training TT due to injury, so for TT comparisons $n=9$. Furthermore, equipment failures during two trials reduced the STHA group to $n=9$ for RE and $n=8$ for $\dot{V}O_{2max}$ and $v\dot{V}O_{2max}$.

Heat acclimation adaptation

A variety of thermal adaptations were observed during GXT 1, post the 5 day, controlled hyperthermia training programme (Table 2 and Figure 1). Notably, this included reduced resting (-0.15°C , $p=0.01$), exercising (-0.21°C , $p=0.04$) and change in T_{CORE} (-0.25°C , $p=0.01$). Furthermore, reduced mean exercising HR ($-3 \text{ b}\cdot\text{min}^{-1}$, $p=0.02$) and RER (-0.08 , $p=0.03$) were observed alongside expansion of the blood plasma volume (5.7%, $p=0.03$). Finally, the change in thermal sensation during GXT 1 was smaller following STHA ($p=0.04$), alluding to a mediated perceived stress during the submaximal run. There was no change in average thermal sensation ($p=0.26$) or average RPE ($p=0.34$) following HA.

Table 2 here

Figure 1 here

Training

There were no differences ($p>0.05$) between groups for total work completed, total exercising time, power output (W) and relative power output ($\text{W}\cdot\text{kg}^{-1}$) during the training week. STHA training elicited a markedly greater physiological strain compared with CON in terms of; peak session HR, average session T_{CORE} , time above 38.5°C , average peak session T_{CORE} , sweat loss volume and sweat loss relative to body mass. Notwithstanding, CON training elicited a mean HR that equated to 81 (± 5)% of maximum HR and mean RPE was 16 (± 1) across the ~ 41 min training. Training responses are displayed below in Table 3.

Table 3 here

Time trial performance

Environmental conditions (WBGT) during the hot TTs did not differ between groups before or after the training week (Pre training; $p=0.07$, Post training; $p=0.429$) or within participants for both the STHA group ($p=0.787$) and CON group ($p=0.436$). Before training TT performance was not different between groups (STHA; Pre 1476 [173] s, CON; 1405 [178] s, $p=0.436$, $d_{\text{av}}=0.40$). However following training, 5 km time in STHA was 1378 [116] s and CON 1396 [177] s, with an interaction effect ($p=0.029$, partial $\eta^2=0.296$) revealing a greater improvement following STHA (-6.2 [5.5]%) than CON (-0.6 [1.7]%). Average HR during the TT was not different following training for either group ($p=0.617$), with no interaction ($p=0.336$). Alongside improved TT performance, finishing T_{SKIN} was lower in STHA following training (Pre; 35.7 [0.35] $^{\circ}\text{C}$, Post; 34.6 [0.31] $^{\circ}\text{C}$, $p=0.001$, $d_{\text{av}}=1.07$), but not in CON (Pre; 35.7 [0.43] $^{\circ}\text{C}$, Post; 35.5 [0.38] $^{\circ}\text{C}$, $p=0.564$), revealing an interaction effect ($p=0.041$, partial $\eta^2=0.283$). Finishing blood lactate concentration was not different between STHA and CON pre ($p=0.323$) or post ($p=0.138$), but was higher in STHA after training (Pre; 4.2 [0.5] $\text{mmol}\cdot\text{l}^{-1}$, Post; 6.8 [0.6] $\text{mmol}\cdot\text{l}^{-1}$, $p<0.001$), but not CON (Pre; 5.0 [0.5] $\text{mmol}\cdot\text{l}^{-1}$, Post; 5.4 [0.6] $\text{mmol}\cdot\text{l}^{-1}$, $p=0.541$). No main effects pre-post (Time), or interaction effects (Time*Group) were observed for RPE (Time; $p=0.821$, Time*Group; $p=0.821$), thermal sensation (Time; $p=0.820$, Time*Group; $p=0.085$), finishing

T_{CORE} (Time; $p=0.142$, Time*Group; $p=0.142$), PSI (Time; $p=0.05$, Time*Group; $p=0.992$). Individual and group $\dot{V}O_{2\text{max}}$ and TT performances are shown below in Figure 2.

Figure 2 here

Determinants of endurance performance in the heat after training

Environmental conditions (WBGT) during the hot GXTs did not differ between groups before or after the training week (Pre; $p=0.372$, Post; $p=0.894$) or within participants for both the STHA group ($p=0.505$) and CON group ($p=1.000$). The change in the determinants of endurance performance following training, when measured in hot conditions, are shown in Figure 3. A main effect for time (pre:post training) in $\dot{V}O_{2\text{max}}$ was observed ($p=0.004$, partial $\eta^2=0.517$), indicating both STHA and CON enhanced $\dot{V}O_{2\text{max}}$ in the heat, however no Group*Time interaction effect was observed ($p=0.228$). The mean increase in $\dot{V}O_{2\text{max}}$ pre to post HA in STHA was 4.0 (2.2) $\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ (7.3 [4.0]%, $d_{\text{av}}=0.47$) and for CON 1.9 (3.7) $\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ (3.8 [7.2] %, $d_{\text{av}}=0.30$). Maximum HR did not differ following training ($p=0.147$) and there was no interaction between groups ($p=0.671$). Similarly, a main effect was observed for LT ($p=0.021$, partial $\eta^2=0.306$), whereby change in STHA was 0.4 (0.6) $\text{km}\cdot\text{h}^{-1}$ (4.0 [6.0]%, $d_{\text{av}}=0.24$) and 0.4 (0.6) $\text{km}\cdot\text{h}^{-1}$ (3.4 [5.3]%, $d_{\text{av}}=0.24$) in CON, with no interaction ($p=0.923$). For LTP there was a main effect for time ($p=0.005$, partial $\eta^2=0.413$), whereby change in STHA was 0.3 (0.4) $\text{km}\cdot\text{h}^{-1}$ (2.5 [2.9]%, $d_{\text{av}}=0.20$) and 0.2 (0.3) $\text{km}\cdot\text{h}^{-1}$ (1.8 [2.2]%, $d_{\text{av}}=0.16$) in CON, but no interaction effect ($p=0.699$). A main effect was also observed for $v\dot{V}O_{2\text{max}}$ ($p=0.031$, partial $\eta^2=0.332$), where change in STHA was 0.5 (0.8) $\text{km}\cdot\text{h}^{-1}$ (3.5 [5.3]%, $d_{\text{av}}=0.24$) and 0.3 (0.8) $\text{km}\cdot\text{h}^{-1}$ (2.6 [5.4]%, $d_{\text{av}}=0.13$) in CON, but no interaction ($p=0.553$). However, running time during GXT 2 revealed both a main effect for time ($p=0.002$, partial $\eta^2=0.532$) and interaction effect ($p=0.006$, partial $\eta^2=0.457$). Following STHA, the mean running time during GXT 2 increased by 78 (43) s (20.8 [12.7]%, $p<0.001$, $d_{\text{av}}=2.09$), compared with 18 [44] s (+9.8 [1.2] %, $p=1.000$) in CON. Finally, a main effect for RE was observed ($p=0.008$, partial $\eta^2=0.459$), with a greater amount of oxygen consumed per kilometre following training, but no interaction ($p=0.341$). For STHA, the mean difference pre to post was 7.3

(7.3) mL.kg⁻¹.km⁻¹ (3.5 [3.5]%, d_{av} =0.59), whilst for CON it was -2.4 (5.0) mL.kg⁻¹.km⁻¹ (1.1 [2.3]%, d_{av} =0.12).

Figure 3 here

Effect of heat stress

The pooled data revealed elevated physiological strain at the end of GXT 1 under heat stress (32.1 [1.2]°C, 57 [6]% RH), compared with cool conditions (12.8 [0.9]°C, 52 [7]% RH), with marked reductions in the determinants of endurance performance (Table 4). Blood lactate concentration was elevated during submaximal running in hot, compared with cool conditions as shown by a -4.4 (6.2)% reduction in both LT and -4.8 (4.5)% in LTP. Other impairments included; -7.7 (5.9)% in $\dot{V}O_{2max}$, -4.0 (4.4)% in $v\dot{V}O_{2max}$ and -19 (10)% in running time to exhaustion during GXT 2. RE improved in hot conditions, compared with cool, with a reduction of 5.3 (4.3)% in oxygen consumption per kilometre.

Table 4 here

Discussion

Compared with cool conditions, heat stress impaired $\dot{V}O_{2max}$ (-7.7%), LT (-4.4%), LTP (-4.8%), $v\dot{V}O_{2max}$ (-4%) and running time during GXT 2 (-19%), but improved RE (+5.3%). Both high intensity normothermic and heat acclimation training improved all of these variables, when measured in a hot environment, aside of RE. Despite these improvements across both training groups, TT performance only improved following STHA (+6.5%) and not following CON (+0.6%). There was a trend towards a greater increase in $\dot{V}O_{2max}$ following STHA (+7.3%), than CON (+3.8%). Moreover, specific thermal adaptations were only inducted following STHA, such as reduced exercising T_{CORE} and improved thermal perception. Therefore, these data allude to roles for $\dot{V}O_{2max}$ and specific thermal adaptations attained using STHA for improving performance in hot conditions, rather than the traditional determinants of endurance performance. Finally, a controlled hyperthermia, cycling STHA programme appears effective for improving aerobic running performance in hot and humid conditions.

Heat acclimation

Despite the greater prevalence of LTHA strategies within environmental physiology research, STHA appears more practical for athletes to combine with competitive schedules and these data demonstrate multi-system adaptations commensurate with the HA phenotype. There were notable improvements in exercising HR ($-3 \text{ b}\cdot\text{min}^{-1}$), RER (-0.08) and T_{CORE} (-0.21°C), alongside an enhanced PV ($+5.7\%$) and reduced elevation of thermal sensation (-0.4). Previous research would suggest 70-75% of the adaptations typically observed from LTHA to be induced in this timescale (Garrett et al. 2011; Mee et al. 2015; Gibson et al. 2015a). The attenuated increase in exercising T_{CORE} , relative to pre-acclimation (Figure 1B), represents reduced heat storage during GXT 1 (post), which alongside the reduction observed in resting T_{CORE} , theoretically affords a greater heat storage capacity. The unchanged average sweat rate following STHA is consistent with previous research (Mee et al. 2015; Garrett et al. 2009; Garrett et al. 2014), however that this occurred alongside a smaller change in T_{CORE} alludes to increased sudomotor sensitivity. The modest, but consistent, reduction in exercising HR (Figure 1A) is indicative of enhanced cardiac output through increased stroke volume, to which thermal adaptations such as reduced T_{CORE} and T_{SKIN} may contribute by reducing the cutaneous vasodilation demand, as well as localised factors such as increased cardiac contractility and/or venous tone (Périard et al. 2015). However, within this timescale the largest contributing factor is likely hypervolaemia, with the observed PV expansion of 5.3% in keeping with other STHA literature (Patterson et al. 2004, Garrett et al. 2009; Garrett et al. 2012). The level of dehydration during HA training may stimulate plasma volume expansion (Garrett et al. 2011), however despite a greater average dehydration of $\sim 3.2\%$ of body mass per session versus Garrett et al. (2012; 2014) of ~ 1.8 - 2.1% , we did not observe a greater plasma volume expansion. The reduction in RER during exercise (Figure 1D) may represent a lower relative intensity ($\% \dot{V}\text{O}_{2\text{max}}$) during the GXT following STHA (Jones & Carter 2000), but may also be explained by a reduction in energy derived through glycogenolysis under heat stress, resulting in a relative maintenance of fat oxidation. Similar effects have previously

been reported following HA (Gibson et al. 2015b) and appear to arise from a reduced exercising body temperature and associated reduced plasma adrenaline levels (Febbraio et al. 1994).

These data demonstrate the efficacy of a novel exercise prescription method for HA, initially based on power output relative to body mass ($2.7 \text{ W}\cdot\text{kg}^{-1}$) (Gibson et al. 2016) and subsequently maintained the maximum tolerable power. Thermal strain was controlled between individuals with the target T_{CORE} of 38.5°C consistently reached within 30 min by the majority of participants (27 ± 4 min), across all days. This method also eliminated a prior cycling maximal test and better controls MHP than $\% \dot{V}O_{2\text{max}}$ (Gibson et al. 2016), therefore maintaining relative thermal strain independently of any progressive increase in aerobic fitness. Therefore, researchers and practitioners should consider prescribing exercise intensity based upon relative, before maximum tolerable power, for time-efficient acclimation training.

Effect of training on GXT and time trial performance

Both training methods demonstrated improvements across the determinants of performance, aside of RE. However, an additional benefit of heat training was only observed in the TT (post; -6.2%), running time to exhaustion during GXT 2 (post; +20.8%) and $\dot{V}O_{2\text{max}}$ (post; +7.3%). The improvements in CON for $\dot{V}O_{2\text{max}}$, LT, LTP and $\dot{V}O_{2\text{max}}$ in hot conditions (Figure 3) appear novel, and may reflect a higher training intensity ($\sim 81\%$ HR maximum), given that previous research has compared against lower intensity ($50\% \dot{V}O_{2\text{max}}$) control training (Lorenzo et al. 2010; Gibson et al. 2015a). These physiological benefits in CON arose independently of thermal strain during HA, and do not represent increased familiarity with running, given cycling training was completed. However, ultimately these improvements do not improve endurance performance in the heat. Previous research highlights the importance of maintaining the core:skin gradient, which otherwise narrows as T_{CORE} and T_{SKIN} increase, increasing skin blood flow demands for heat dissipation (Gonzalez-Alonso & Calbet 2003). As the core:skin gradient narrows, increased blood flow to the skin results in a reduction in cardiac output, which in turn impairs $\dot{V}O_{2\text{max}}$ (Périard et al. 2011; Lee et al. 2015). Furthermore, elevated T_{SKIN}

influences thermal comfort and RPE, which are associated with voluntary reductions of exercise intensity in the heat (Schlader et al. 2011a, Schlader et al. 2011b, Barwood et al. 2015). Indeed, the interrelation of T_{SKIN} , cardiovascular strain and RPE has previously been highlighted by Schlader et al. (2011a), reinforcing the potential for HA to influence performance through a variety of mechanisms (Nybo et al. 2014). This relationship is supported in the current data by the maintenance of average HR, RPE and thermal sensation across trials, despite running 6.2% faster following STHA. Notwithstanding, $\dot{V}O_{2\text{max}}$ likely explains some performance improvement, as there appears to be a trend for a greater increase in $\dot{V}O_{2\text{max}}$ from STHA than CON, despite no interaction effect. The delta change and effect size for STHA (7.3%, $d=0.47$), compared with CON (3.8%, $d=0.30$), indicate meaningful change. Furthermore, the mean difference between the change in STHA and change in CON ($2.1 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) exceeds a meaningful change ($2 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$, Tanner & Gore 2013). There was considerable variability in the $\dot{V}O_{2\text{max}}$ changes in CON (Figure 2), which may be partially attributable to genetics (Bouchard et al. 1999). However, we also observed that the individual who experienced the largest increase in $\dot{V}O_{2\text{max}}$ had one of the lowest $\dot{V}O_{2\text{max}}$ of all participants ($58 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$), suggesting the high intensity training provided a larger stimulus for lesser trained individuals (CON group mean $63 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$). Thus STHA may be an appropriate training intervention for acutely enhancing $\dot{V}O_{2\text{max}}$, with an enlarged PV the most likely mediator, within this sub-elite cohort.

The degree to which the improved running performance in the heat can be attributed to the trend towards an improved $\dot{V}O_{2\text{max}}$ is unclear. Both Périard et al. (2011; 2015) and Schlader et al. (2011a) have highlighted how $\dot{V}O_{2\text{max}}$ may be a primary determinant of self-paced endurance performance in the heat, based on the maintenance of relative exercise intensity. This supports the traditional model of endurance performance, where $\dot{V}O_{2\text{max}}$ may set the upper limit for performance in endurance events (Bassett & Howley 2000) and strongly predicts performance in a heterogeneous population (McLaughlin et al. 2010). Furthermore, previous research has shown an 8% improvement in 1 hour cycling TT performance (38°C) to parallel an 8% increase in $\dot{V}O_{2\text{max}}$ in the heat following HA (Lorenzo et al. 2010). However, Lorenzo et al (2010) also reported a greater increase in the LTP

following HA, compared with control training, whilst both Lorenzo et al. (2010) and our data demonstrates PV expansion, a lower T_{SKIN} and larger core-to-skin gradient, indicating $\dot{V}O_{2max}$ alone cannot explain improved performance in the heat. Accordingly, we observed a trend for increased $\dot{V}O_{2max}$ in CON (+3.8%), but TT performance did not improve (+0.6%). The modest improvements in blood lactate thresholds following STHA are not without precedence (Chalmers et al. 2016), but appear surprising, given greater changes previously reported (Lorenzo et al. 2010; Chalmers et al. 2014; Neal et al. 2015), potentially arising through reduced body heat storage maintaining splanchnic circulation, thus preserving lactate clearance and reducing glycogenolysis (Febbraio et al. 1994). It is possible the ~2-3% improvement in both LT and LTP from CON has arisen due to the intensity of exercise, with an average intensity during normothermic cycling training at ~81% of HR maximum, maintained for ~40 min daily across 5 days, which is likely to be at or above LTP for most participants, an intensity that promotes adaptation when replicated over four weeks, 2-4 times weekly (Keith et al. 1992). The apparent impairments to RE following both training programmes demonstrate greater oxygen consumption during GXT 1 (post). Increases in submaximal oxygen consumption may present alongside increases in $\dot{V}O_{2max}$ and/or lactate thresholds, due to the interrelation of the determinants of endurance performance (Midgley et al. 2007), with a greater absolute aerobic energy provision at the same submaximal intensities. Accordingly, there is a trend for greater oxygen consumption in the STHA group (3.5%, $d_{av}=0.59$) compared with CON (1.1%, $d_{av}=0.12$), reflecting the changes observed in $\dot{V}O_{2max}$ following STHA. In summary, despite CON eliciting improvements across the determinants of endurance performance, TT performance was unaffected, reinforcing that self-selected running speed is determined by a combination of physiological, thermoregulatory and perceptual factors (Nybo et al. 2014).

Effect of heat stress

Heat stress broadly enhanced physiological strain, characterised by increased HR, RER, T_{CORE} , T_{SKIN} , thermal sensation and RPE during incremental running. Whilst the determinants of endurance

performance effectively predict performance in normothermic conditions (Joyner 1991; McLaughlin et al. 2010), the impairment afforded to runners exercising under moderate heat stress is not well defined. Such information could benefit preparation for competitions under heat stress, as well as those who complete field testing in the heat.

The largest decrement was observed in $\dot{V}O_{2\max}$ (-7.3%), with smaller reductions across LT, LTP and $v\dot{V}O_{2\max}$ (all <-5%), whilst RE was improved (~+5%). Furthermore, the total running time during the $\dot{V}O_{2\max}$ test (GXT 2) reduced by 19%. Although T_{CORE} at $\dot{V}O_{2\max}$ was elevated in the hot condition (38.9°C), compared with cool (38.5°C), the largest difference between conditions was in T_{SKIN} , with mean T_{SKIN} 35.3 °C in hot conditions, compared with 28.3°C in the cool, which was 7% greater at the end of GXT 1. This supports previous assertions that when T_{CORE} is elevated, a reduction in $\dot{V}O_{2\max}$ is more aligned to the T_{SKIN} elevation, as a consequence of skin blood flow requirements (Arngrímsson et al. 2004). A reduced maximal cardiac output is considered the primary limitation to $\dot{V}O_{2\max}$ during heat stress (Rowell 1966), as peripheral vasodilation compounds venous return (Gonzalez-Alonso & Calbet 2003), reinforced by recent evidence demonstrating no change in myocardial systolic or diastolic function during maximal exercise in the heat, compared with normothermic conditions (Smith et al. 2015). This is despite alterations to the Frank–Starling relationship and cardiac contractility having previously been reported during passive heat stress (Wilson et al. 2009). It should be noted the decrement to $\dot{V}O_{2\max}$ is progressive, therefore the ~8% impairment to $\dot{V}O_{2\max}$ may vary when exercising harder or longer than ~24 min of incremental running, or under different environmental conditions. Similarly, reductions in the exercise intensity and fractional utilisation at LT and LTP have previously been reported (Tyka et al. 2009; De Barros et al. 2011; Lorenzo et al. 2011), however different methodologies preclude synthesis of typical delta change.

In contrast to the relative unanimity surrounding the effects of heat stress on $\dot{V}O_{2\max}$ and blood lactate thresholds, the effect on RE is less well defined. Enhanced (MacDougall et al. 1974), reduced (Rowell et al. 1967) and unchanged (Rowell et al. 1969; Maron et al. 1976) submaximal $\dot{V}O_2$ have all

been reported during submaximal exercise in the heat. Elevated T_{CORE} has been associated with small increases in metabolic rate and therefore $\dot{V}O_2$ during prolonged submaximal exercise (Shvartz et al. 1977; MacDougall et al. 1974). However, oxygen kinetics remain unchanged under heat stress (Koga et al. 1997; Nybo et al. 2001), which is pertinent given the incremental protocol in the current study. An increased energy demand may reflect increased peripheral circulation, sweat gland activity, hyperthermic hyperventilation and an increased mitochondrial metabolic rate, as well as a reduction in efficiency within the mitochondria (Brooks et al. 1971; MacDougall et al. 1974). Of note, hyperventilation may increase $\dot{V}O_2$ by 0.4-0.6 mL.kg⁻¹.min⁻¹ when ventilation is 117-147 L.min⁻¹ (Aaron, Johnson, et al. 1992; Aaron, Seow, et al. 1992). In spite of these alterations, a warmer muscle is more efficient through a range of mechanisms (Racinais & Oksa 2010), including enhanced neural drive (Racinais et al. 2004) and reduced viscosity of the muscles and joints (Hill 1927). Therefore, increased biomechanical efficiency under environmental heat stress (Rowell et al. 1969; Saunders et al. 2004), particularly in Type 1 muscle fibres (De Ruyter & De Haan 2000), may explain the reduced $\dot{V}O_2$ during GXT 1 in the hot condition, with this benefit potentially dissipating and/or outweighed by energy demanding thermoregulatory responses to the progression of heat strain (Bailey & Pate 1991). Accordingly, ventilation rate was not greater during hot GXT 1. It is also plausible that the cool condition elicited a small reduction in muscle temperature, affording a small impairment to muscle efficiency and requiring greater oxygen consumption for a given running speed, although we acknowledge muscle temperature was not measured.

In summary, this experiment supports the use of 5 days of controlled hyperthermia, with high intensity exercise, for improving endurance running performance in the heat. STHA improves endurance running performance in comparison to work matched normothermic training, despite equality of adaptation for typical determinants of performance (LT, LTP, $\dot{V}O_{2\text{max}}$). Accordingly, these data highlight the ergogenic effect of STHA, potentially via greater improvements in $\dot{V}O_{2\text{max}}$ and specific thermal adaptations, that may include reduced T_{SKIN} , T_{CORE} and perceived strain, alongside plasma volume expansion, all of which do not present following normothermic training.

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1 **Table captions**

2 Table 1: Comparison of experimental groups. Data are mean (\pm SD).

3 Table 2: Effect of 5 days STHA on recognised HA criteria. Data are mean (\pm SD).

4 Table 3: Mean (\pm SD) training responses during STHA and CON training programmes.

5 Table 4: Effect of heat stress on physiological variables (mean \pm SD). Exercising measures taken at
6 end of GXT 1, after 24 min of running during the final stage of (incremental) GXT 1 test. '**' $p < 0.05$.

7 **Figure captions**

8 Figure 1: Clockwise from top left: Pre and post STHA HR response (A), exercising T_{CORE} (B), thermal
9 sensation (C) and RER (D) during GXT 1. Data are mean (\pm SD)

10 Figure 2: Average (grey columns) and individual data of percentage change in $\dot{V}O_{2\text{max}}$ and 5 km time
11 trial performance following heat acclimation (STHA) and normothermic training (CON).

12 Figure 3: Mean (\pm SD) percentage difference between hot and cool trials pre and post STHA and CON
13 for the individual determinants of endurance performance. Error bars represent one standard
14 deviation.

15

1 Tables

2 Table 1: Comparison of experimental groups. Data are mean (\pm SD).

	Heat acclimation	Control	<i>p</i>
	<i>n</i> = 10	<i>n</i> = 7	
Age	34 (16)	27 (3)	0.279
Body mass (kg)	73.2 (9.2)	70.2 (8.9)	0.512
Stature (cm)	176 (7)	179 (6)	0.346
$\dot{V}O_{2max}$ (mL.kg⁻¹.min⁻¹)	58.9 (6.7)	62.4 (5.9)	0.280
Maximum heart rate (b.min⁻¹)	187 (11)	189 (9)	0.720
Sum of skinfolds (mm)	25.8 (3.9)	23.5 (5.3)	0.326
Body surface area (m²)	1.89 (0.14)	1.88 (0.13)	0.878
Recent 5 km (s)	1253 (103)	1188 (100)	0.214

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1 Table 2: Effect of 5 days STHA on recognised HA criteria. Data are mean (\pm SD).

Criteria	Pre	Post	Δ Pre - post	<i>p</i>	<i>d</i> _{av}
Resting HR (b.min ⁻¹)	52 (5)	49 (8)	-2 (4)	0.115	0.36
Exercising HR (b.min ⁻¹)	163 (14)	159 (13)	-3 (4)	0.023*	0.26
Resting T _{CORE} (°C)	36.97 (0.33)	36.83 (0.32)	-0.15 (0.12)	0.014*	0.45
Exercising T _{CORE} (°C)	38.12 (0.36)	37.91 (0.43)	-0.21 (0.25)	0.039*	0.54
Δ Exercising T _{CORE} (°C)	1.26 (0.27)	1.00 (0.28)	-0.25 (0.20)	0.006*	0.91
Resting T _{SKIN} (°C)	34.6 (0.3)	33.8 (1.1)	-0.77 (0.95)	0.046*	1.13
Exercising T _{SKIN} (°C)	35.2 (0.8)	34.9 (0.7)	-0.32 (0.52)	0.205	0.44
Δ Exercising T _{SKIN} (°C)	0.1 (1.0)	0.0 (1.1)	-0.14 (0.99)	0.670	0.13
Exercising blood glucose (mmol.l ⁻¹)	4.63 (0.62)	4.80 (0.48)	0.17 (0.40)	0.240	0.31
Δ Exercising blood glucose (mmol.l ⁻¹)	1.47 (0.77)	1.02 (0.80)	-0.44 (0.54)	0.040*	0.56
Exercising RER	1.02 (0.15)	0.94 (0.07)	-0.08 (0.12)	0.032*	0.70
Δ Exercising RER	0.22 (0.06)	0.18 (0.05)	-0.03 (0.05)	0.025*	0.59
Sweat loss (L)	1.35 (0.3)	1.39 (0.39)	0.05 (0.20)	0.503	0.13
Plasma volume	-	-	5.7 (7.1) %	0.031*	1.06
Exercising TS	6.0 (0.8)	5.8 (0.6)	-0.2 (0.6)	0.262	0.29
Δ Exercising TS	2.0 (0.5)	1.6 (0.4)	-0.4 (0.6)	0.042*	0.86
Exercising RPE	14.1 (0.9)	13.8 (1.1)	0.0 (2.0)	0.342	0.31

2 NB. Δ = change in variable from stage 1 to stage 6. * denotes *p*<0.05.

1 Table 3: Mean (\pm SD) training responses during STHA and CON training programmes.

	STHA	CON	<i>p</i>	<i>d_{av}</i>
Environmental conditions ($^{\circ}$C, % RH)	37 (0.8), 59 (9)%	20 (1), 43 (7)%	-	-
Total work completed (kJ)	2443 (657)	2530 (336)	0.502	0.36
Session duration (min)	90 (0)	41 (2)	-	-
Exercise duration (min)	36 (6)	41 (2)	0.289	0.57
Power output (W)	201 (33)	203 (20)	0.895	0.07
Relative power output ($W \cdot kg^{-1}$)	2.7 (0.3)	2.9 (0.2)	0.181	0.73
Heart rate at 20 min (% maximum)	89 (3)	82 (5)	0.003*	1.68
Peak heart rate (% maximum)	94 (3)	85 (6)	0.002*	1.77
Peak RPE	19 (1)	17 (1)	0.011*	1.42
Session T_{CORE} ($^{\circ}$C)	38.5 (0.2)	37.8 (0.2)	<0.001*	4.08
Time $T_{CORE} >38.5^{\circ}$C (min)	63 (5)	2 (2)	-	-
Peak session T_{CORE} ($^{\circ}$C)	39.1 (0.2)	38.2 (0.2)	<0.001*	4.91
Sweat loss (L)	2.3 (0.7)	0.5 (0.2)	<0.001*	3.43
Sweat loss (% body mass)	3.2 (1.1)	0.6 (0.2)	<0.001*	3.24

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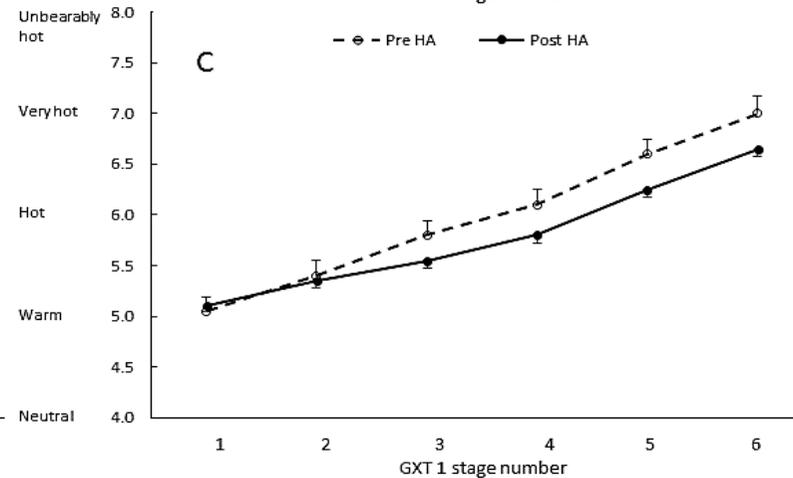
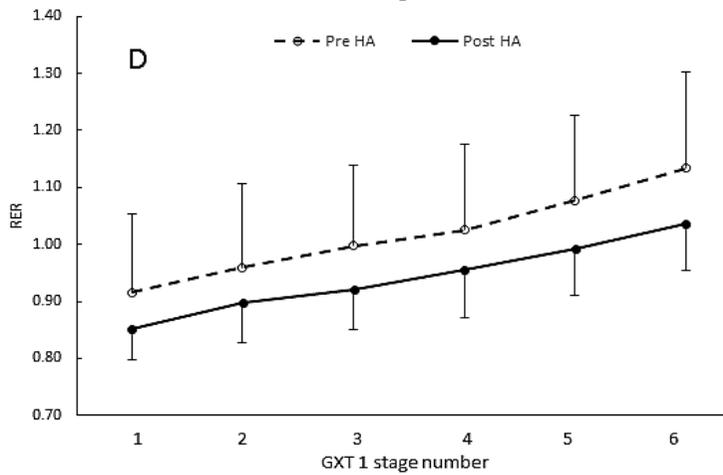
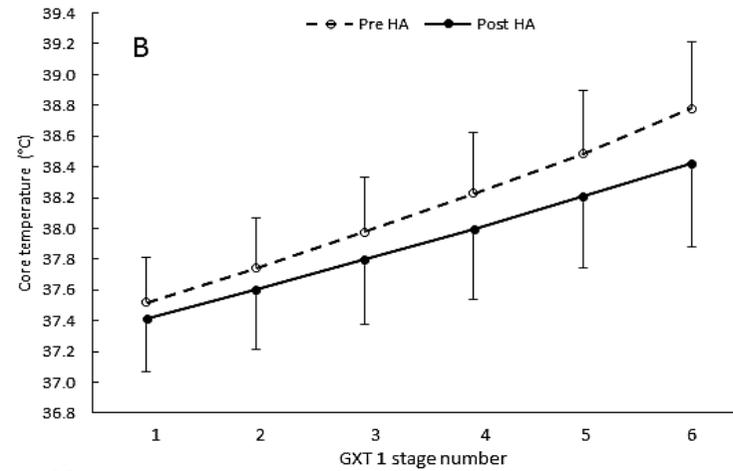
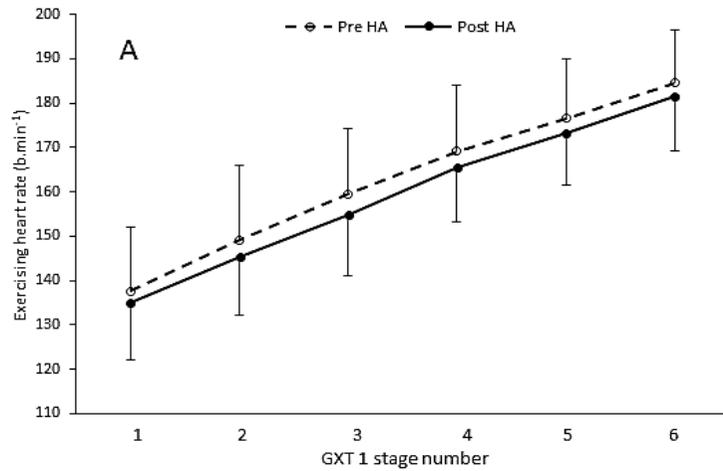
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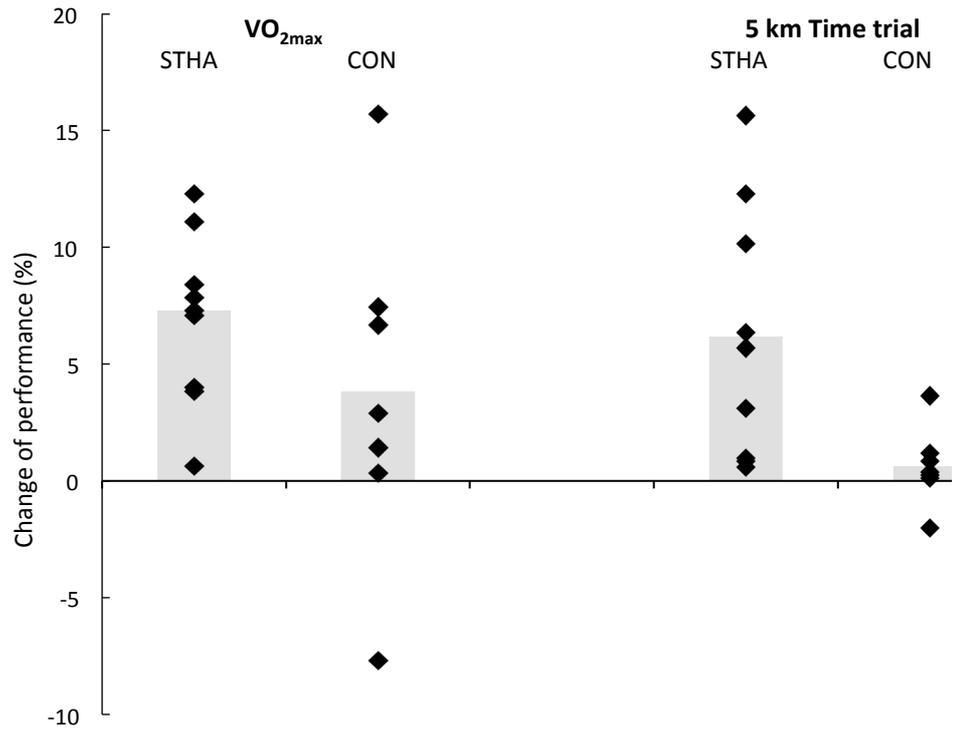
1 Table 4: Effect of heat stress on physiological variables (mean \pm SD). Exercising measures taken at
 2 end of GXT 1, after 24 min of running during the final stage of (incremental) GXT 1 test. '**' $p < 0.05$.

	Cool (13°C)	Hot (32°C)	<i>p</i>	<i>d_{av}</i>
Exercising HR (b.min ⁻¹)	177 (12)	184 (12)	<0.001*	0.60
Exercising T _{CORE} (°C)	38.46 (0.41)	38.70 (0.38)	0.031*	0.61
Exercising T _{SKIN} (°C)	28.34 (1.37)	35.34 (1.08)	<0.001*	5.69
Exercising blood glucose (mmol.l ⁻¹)	5.39 (0.80)	5.95 (1.20)	0.033*	0.57
Exercising blood lactate (mmol.l ⁻¹)	4.85 (1.17)	6.21 (1.46)	<0.001*	1.04
Exercising RER	1.04 (0.07)	1.12 (0.13)	0.081	0.73
Exercising ventilation (L.min ⁻¹)	125 (17)	129 (20)	0.185	0.23
Sweat loss (L)	0.64 (0.24)	1.31 (0.27)	<0.001*	2.63
Exercising TS	5.4 (1.0)	6.9 (0.8)	<0.001*	1.60
Exercising RPE	15.9 (1.3)	17.7 (1.4)	<0.001*	1.36
$\dot{V}O_{2max}$ (mL.kg ⁻¹ .min ⁻¹)	61.0 (6.2)	56.3 (7.1)	<0.001*	0.70
Lactate threshold speed (2 mmol.l ⁻¹) (km.h ⁻¹)	12.3 (1.9)	11.7 (1.8)	0.008*	0.31
Lactate turnpoint speed (4 mmol.l ⁻¹) (km.h ⁻¹)	14.4 (2.0)	13.7 (1.7)	<0.001*	0.40
Running economy (mL.kg ⁻¹ .km ⁻¹)	227 (17)	215 (16)	<0.001*	0.75
$v\dot{V}O_{2max}$ (km.h ⁻¹)	16.1 (2.1)	15.8 (2.3)	0.030*	0.23
Running time GXT 2 (s)	506 (44)	408 (56)	<0.001*	1.96

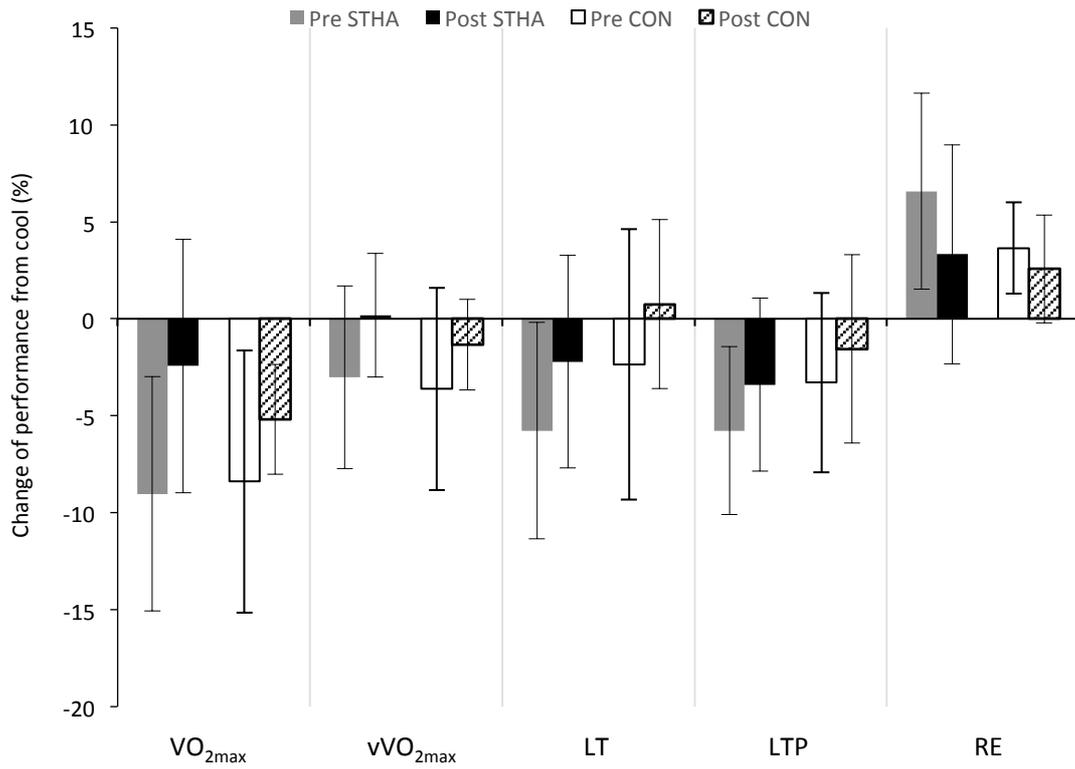
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1 Figures





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 2 Figure 2: Average (grey columns) and individual data of percentage change in $\dot{V}O_{2max}$ &
 3 trial performance following heat acclimation (STHA) and normothermic training (CON).



1

2 Figure 3: Mean (\pm SD) percentage difference between hot and cool trials pre and post STHA and CON
 3 for the individual determinants of endurance performance. Error bars represent one standard
 4 deviation.

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