Physiological responses to incremental exercise in the heat following internal and external precooling

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Twelve males completed three incremental, discontinuous treadmill tests in the heat [31.9(1.0) °C, 61.9(8.9)%] to determine speed at two fixed blood lactate concentrations (2 and 3.5 mmol/L), running economy (RE), and maximum oxygen uptake (VO₂max). Trials involved 20 min of either internal cooling (ICE, 7.5 g/kg ice slurry ingestion) or mixed-methods external cooling (EXT, cold towels, forearm immersion, ice vest, and cooling shorts), alongside no intervention (CON). Following precooling, participants ran 0.3 km/h faster at 2 mmol/L and 0.2 km/h faster at 3.5 mmol/L (P = 0.04, partial η² = 0.27). Statistical differences were observed vs CON for ICE (P = 0.03, d = 0.15), but not EXT (P = 0.12, d = 0.15). There was no effect of cooling on RE (P = 0.81, partial η² = 0.02), nor on VO₂max (P = 0.69, partial η² = 0.04). An effect for cooling on physiological strain index was observed (P < 0.01, partial η² = 0.41), with differences vs CON for EXT (P = 0.02, d = 0.36), but not ICE (P = 0.06, d = 0.36). Precooling reduced thermal sensation (P < 0.01, partial η² = 0.66) in both cooling groups (P < 0.01). Results indicate ICE and EXT provide similar physiological responses for exercise up to 30 min duration in the heat. Differing thermoregulatory responses are suggestive of specific event characteristics determining the choice of cooling. Precooling appears to reduce blood lactate accumulation and reduce thermoregulatory and perceptual strain during incremental exercise.

Endurance exercise is underpinned by the ability to transfer chemical energy into a given exercise velocity (Coyle, 1999). The status of this biological process can be assessed using physiological markers such as the lactate thresholds, running economy (RE), and maximum oxygen uptake (VO₂max). Under normothermic conditions, when combined with the peak treadmill velocity, these markers have been shown to account for 97.8% of the variation in 16 km run time (McLaughlin et al., 2010). McLaughlin et al. (2010) highlighted that VO₂max accounted for 90.2% of variation in running time in a group with heterogenous VO₂max values. Furthermore, Lorenzo et al. (2011) has shown the lactate turnpoint (LTP) to be a strong predictor of time trial performance in both cold (r = 0.89) and hot (r = 0.87) environments.

The addition of heat stress during endurance running is characterized by an enhanced metabolic (Parkin et al., 1999), cardiovascular (CV; González-Alonso et al., 2008), and sensory strain (Villanova et al., 1997) as core temperature (Tcorp) increases. At moderate levels of heat strain, such alterations are associated with reductions of the LTP (Lorenzo et al., 2011) and VO₂max (Nybo et al., 2014). The reduction in LTP in the heat is of particular importance given it remains a valid predictor of endurance performance in hot environments (Lorenzo et al., 2011). This decline may be associated with the shift toward carbohydrate oxidation (Fink et al., 1975; Parkin et al., 1999) and the increased blood lactate accumulation observed during heat strain (Hargreaves, 2008). At maximal exercise intensities, VO₂max is attenuated because of increased skin blood flow required for heat dissipation, which leads to a reduction in stroke volume as a consequence of cutaneous pooling, and ultimately limits muscular blood flow and oxygen delivery (González-Alonso et al., 2008). Enhanced oxygen consumption has also been reported during heat strain (Consolazio et al., 1973), although not all studies observed this effect (González-Alonso et al., 1999). Furthermore, during prolonged or intense endurance exercise, a protective reduction in central nervous system (CNS) motor output may be observed as core temperature approaches 40 °C (Cheung, 2007). Thus, thermal interventions such as precooling that reduce body temperature, thereby increasing heat storage capacity or reducing the rate of heat storage, have been shown to benefit endurance exercise in the heat.

A dichotomous approach toward precooling is apparent, with interventions either cooling externally or internally, eliciting different skin, core, and muscle temperatures, and therefore potentially different.
physiological responses. The attenuated LTP in the heat may be in part a consequence of increased muscular glycolysis (Fink et al., 1975; Parkin et al., 1999), an alteration in muscle metabolism that external body cooling has previously been shown to mediate (Kozlowski et al., 1985). Therefore, external cooling may help reduce plasma lactate accumulation and could have an effect on LTP. Similarly, by reducing \( T_{\text{CORE}} \), internal cooling may reduce the cutaneous circulation that can inhibit cardiac filling, thereby ameliorating CV strain that ultimately causes a reduction in \( \dot{V}O_{2\text{max}} \). Therefore, accurately quantifying any differences in the responses to different types of cooling is important to optimize cooling strategies.

The vast number of cooling techniques reflects the challenge of providing a large cooling impulse through a technique that remains practical for use across a number of venues. Considerable growing evidence supports the use of internal cooling through ice slurry ingestion (ICE) as an ergogen for endurance exercise in the heat (Jones et al., 2012; Siegel & Laursen, 2012; Wegmann et al., 2012). In addition to increasing heat storage capacity, direct cooling of sensitive thermoreceptors within the splanchnic region may contribute to a reduced perceived thermal strain (Villanova et al., 1997). Further, visceral cooling may preserve splanchnic flow that reduces during heat strain (Rowell et al., 1968), as well as prevent against endotoxin leakage that has been associated with impaired muscle force generation (Supinski et al., 2000) and exertional heat illness (Sawka et al., 2011). ICE typically elicits a substantial reduction in \( T_{\text{CORE}} \) of 0.3–0.6 °C (Siegel et al., 2010; Siegel et al., 2012) and has been shown to aid time trial performance in the heat, improving a 40-km laboratory cycle by 6.5% compared with no cooling (Ihsan et al., 2010). ICE appears to permit similar running time to exhaustion as the gold standard technique of cold water immersion (Siegel et al., 2012), with recent systematic reviews advocating ICE to avoid impracticalities with water immersion (Jones et al., 2012; Siegel & Laursen, 2012; Ross et al., 2013). Moreover, ICE is a simple strategy that may complement hydration and nutritional strategies during competition in the heat.

From an exogenous perspective, mixed-method whole-body external cooling (EXT) is gaining prominence following the apparent limited effectiveness of individual cooling garments on endurance performance (Ranalli et al., 2010; Jones et al., 2012). Duffield et al. (2009) combined cooling garments to enhance the cooling volume and reported a blunted rise in \( T_{\text{CORE}} \) during exercise, resulting in increased work during 30 min of intermittent sprinting. External cooling may not always elicit a reduction in \( T_{\text{CORE}} \) prior to exercise (Minett et al., 2011, 2012a, b), but appears to permit a reduced rate of heat storage during exercise by enhancing heat dissipation through an increased core-to-skin gradient (Kay et al., 1999). The reduced \( T_{\text{SKIN}} \) will also lead to reduced vasodilation of peripheral capillary beds, potentially lowering the overall CV strain. Cooling of the skin may be an important mediator of thermal sensation (TS; Schlader et al., 2011), and has been associated with a 6% increase in self-selected exercise intensity during a 30-min cycling trial when an initial reduction in \( T_{\text{CORE}} \) was not observed (Kay et al., 1999). Minett et al. (2011) has subsequently identified a dose-dependent response with skin-cooling surface area coverage, the critical factor for exercise capacity and adopting a mixed-methods approach to increase total work during intermittent sprinting in the heat, compared with no cooling (Minett et al., 2011, 2012b). As with ICE, EXT is a simple and practical technique; however, it has yet to be assessed prior to endurance running or on the physiological markers that are strongly associated with endurance exercise.

The aim of this study was to compare the physiological responses with practical and evidenced internal and external precooling techniques through the markers of lactate thresholds, \( \text{RE} \), and \( \dot{V}O_{2\text{max}} \). Our first hypothesis stated both precooling techniques would increase lactate threshold, improve \( \text{RE} \), and increase \( \dot{V}O_{2\text{max}} \) relative to no cooling. Our second hypothesis stated internal cooling would elicit the greatest improvement within these markers because of the magnitude of \( T_{\text{CORE}} \) reduction, and the size of effects previously reported for this technique.

### Methods

#### Participants

Twelve male recreational club runners volunteered as participants [mean (SD): age 38 (11) years, stature 177.8 (7) cm, mass 76.1 (5.7) kg, sum of four skinfolds 32.6 (7.1) mm, \( \dot{V}O_{2\text{max}} \) 57.5 (4) mL/kg/min. All participants met the eligibility criteria of running a sub-21 min 5-km or sub-43 min 10-km race in the previous 2 months. Each participant provided written informed consent, and institutional ethical approval was issued in accordance with the Helsinki declaration 1975 (revised 2008). Participants replicated their diet in the 12 h prior to each session refraining from alcohol, caffeine, and strenuous activity for 24 h prior to the measurements in line with similar previous research (Gibson et al., 2014). Finally, participants were asked to prepare for each trial as they would in a competition.

#### Experimental design

A randomized controlled design was used with each participant performing experimental trials under three conditions: control (no cooling, CON), internal cooling (ICE slurry ingestion, ICE), and external cooling (mixed methods, EXT). Four trials were completed, involving two graded exercise tests during each visit, and the first trial serving as a familiarization. Participants were instrumented during the familiarization. The subsequent three trials were completed in a randomized order, separated by 7–10 days. An overview of each trial is provided in Fig. 1. Briefly, trials comprised four phases: 10-min rest, 20-min precooling, 5-min warm-up, and then the graded exercise tests (GXT 1 and GXT 2), with the entire trial within a hot and humid environment [31.9 (1.0) °C, 61(8.9)% relative humidity]. In order to replicate a competition schedule, the exercise test began 15 min after cooling.
Cooling interventions

During ICE, participants ingested 7.5 g/kg body mass of ice slurry (−1 °C) during the cooling phase. Such a volume has previously been shown to elicit large reductions in $T_{\text{CORE}}$ without inducing gastrointestinal distress (Siegel et al., 2012). Of this volume, the drink consisted of two-thirds shaved ice using a snow cone maker (JM Posner, Watford, Tesco Stores Ltd, Cheshunt, UK) and one-third diluted drinking cordial (High Juice, 7.3 g carbohydrate per 100 mL of diluted drink). This cordial is a noncarbonated syrup made from fruit juice, water, and carbohydrate that was diluted one part cordial to four parts water. Slurry was dispensed in equal amounts every 5 min over the 20 min precooling period to prevent gastrointestinal discomfort, with total drink volumes typically between 500 and 600 mL.

During EXT, participants were cooled using the maneuver adopted by Minett et al. (2011). This involved wet, iced towels covering the head and neck, forearm and hand immersion in cold water (9 °C), an ice vest on the torso (Artic Heat, Queensland, Australia), and ice packs affixed to the quadriceps using cooling shorts. Towels were swapped after 10 min and hand immersion water temperature was actively maintained throughout. The same volume of squash was provided to match sugar intake between all stages, water temperature was actively maintained throughout. The same volume of squash was provided to match sugar intake between all stages, and the drink delivered to the hot environment from an ambient laboratory temperature (21 °C) during EXT and CON.

Graded exercise tests

Participants entered the environmental chamber (TISS, Hampshire, Nashua, UK) within which conditions were continuously monitored throughout the trial using a heat stress meter (HT30, Extech Instruments, Nashua, USA). Following rest and precooling phases, a 5-min warm-up was completed at 9 km/h on a motorized treadmill (Woodway ELG2, Weil am Rhein, Germany). GXT 1 was similar to that described by Jones (2006), initially a submaximal incremental speed protocol followed by GXT 2, an incremental gradient protocol to volitional exhaustion. Starting speeds were between 8 and 10 km/h (1% gradient) depending on recent running performance, with each participant completing a minimum of six stages, using speed increments of 1 km/h. Each stage was 4 min, consisting of 3 min running and 1 min for capillary blood sampling, analyzed using a YSI 2300 lactate analyzer (YSI, Hampshire, UK). Exercise continued until an exponential increase in blood lactate was observed, or the participant felt unable to complete the subsequent stage. The first capillary sample was taken 18 min after hand immersion ceased, of which 8 min was exercise. The same number of exercise stages and running speeds completed during the familiarization trial were replicated during all subsequent trials. Following a 2-min rest, GXT 2 began at a speed 2 km/h below the previous final speed with gradient increasing by 1% each min and continuing until volitional exhaustion (Jones, 2006).

Physiological measures

During the familiarization trial, anthropometric data were collected for stature, body mass, and a four-site skinfold calliper assessment (Harpenden, Burgess Hill, UK) across iliac crest, subscapular, triceps, and biceps (Durnin & Womersley, 1974). During all trials, a urine sample was requested upon arrival for assessment of hydration status. Euhydration was achieved when urine osmolality and urine specific gravity were below 700 mOsml/kg H2O and 1.020, respectively (Sawka et al., 2007). Single-use rectal probes (Henleys Medical, Welwyn Garden City, UK, Meter Logger Model 401, Yellow Springs Instruments, Missouri, USA) were inserted 10 cm beyond the anal sphincter for $T_{\text{CORE}}$ measurement. Telemetry thermistors (U-Type connected to Gen II GD38 transmitter, Eltek, Cambridge, UK) were attached to the mid-belly of the pectoralis major, biceps brachii, rectus femoris, and gastrocnemius for measurement of skin temperature ($T_{\text{SKIN}}$) with data transmitted wirelessly to a datalogger (RX250AL 1000 Series Wireless Squirrel Logger, Eltek). Heart rate was monitored continuously using a Polar 810i heart rate monitor (Kempele, Finland). Heart rate (HR), $T_{\text{CORE}}$, $T_{\text{SKIN}}$, rating of perceived exertion (RPE, Borg, 1998), and TS (0 = unbearably cold to 8 = unbearably hot; Gagge et al., 1969) were noted every 5 min during rest and precooling and at the end of each stage during exercise. The following physiological responses were calculated: lactate thresholds, RE, maximum oxygen consumption ($VO_{2\text{max}}$), and velocity at $VO_{2\text{max}}$ ($v_{VO_{2\text{max}}}$). Fixed blood lactate concentrations of 2 and 3.5 mmol/L were used to denote the lactate threshold and LTP, respectively, by solving the polynomial regression equation for blood lactate vs speed at 2 and 3.5 mmol/L as per Saunders and Green (2013). Ventilatory gases were measured using 30 s averaging from a Metalyzer Sport analyzer (Cortex, Leipzig, Germany) and the two values from the final minute of each stage used for measuring RE, ventilation ($V_{E}$), and respiratory exchange ratio (RER). Average RE across the first six exercise stages is presented, although the data from each individual stage was used for analysis. During the $VO_{2\text{max}}$ test, the highest 15-s moving average recorded represented $VO_{2\text{max}}$. A different data averaging approach was adopted because of the short recovery period between the two parts of the test to attenuate a potential effect on blunted $VO_{2\text{max}}$ values. Recovery was minimal in order to help ensure both physiological and perceptual effects of cooling would still be present while testing $VO_{2\text{max}}$. Velocity at $VO_{2\text{max}}$ ($v_{VO_{2\text{max}}}$) was calculated by multiplying $VO_{2\text{max}}$ (mL/kg/min) by...
60 and divided by the mean RE (mL O2/kg/km) determined during the first six stages of the treadmill test as per Jones (2006). Sweat rate (L/h) was calculated from the difference in pre- and post-nude body mass divided by the individual exercise duration.

Statistical analysis and derivative calculations

The following derivative calculations were completed for Mean skin temperature (T_SKIN; Ramanathan, 1964):

\[
\text{Mean } T_{\text{SKIN}} = 0.3 \times (T_{\text{CHEST}} + T_{\text{ARM}}) + 0.2 \times (T_{\text{THIGH}} + T_{\text{CALF}})
\]

Physiological Strain Index (PSI; Moran et al., 1998) whereby \( T_{\text{CORE}} \) and \( HR_0 \) denote baseline and \( T_{\text{CORE}} \) and \( HR_1 \) denotes measurement taken at the respective time:

\[
\text{PSI} = \left[ 5 \times (T_{\text{CORE}}^1 - T_{\text{CORE}}^0) \times (39.5 - T_{\text{CORE}}^0) \right] + \left[ 5 \times (HR^1 - HR^0) \times (180 - HR^0) \right].
\]

Sweat rate (L/h) was calculated from the difference in pre- and post-nude body mass divided by the individual exercise duration. All outcome variables were assessed for normality and sphericity prior to further analysis. Data were analyzed in three phases: rest, cooling, and exercise. Two-way, repeated-measures analysis of variance (ANOVA; cooling type \( \times \) time) were used to test for differences in blood lactate indices, respiratory responses, \( T_{\text{CORE}} \), \( T_{\text{SKIN}} \), PSI, RPE, and TS. One-way, repeated-measures ANOVA were used to detect differences between running time until \( \text{VO}_2\text{max} \) during GXT 2, \( \text{vVO}_2\text{max} \), thermoregulatory variables at rest, sweat rate, absolute change during cooling, and change during exercise. Where appropriate, Bonferroni-adjusted pairwise comparisons revealed where differences occurred. Data were analyzed using SPSS (Version 20, SPSS Inc., Chicago, Illinois, USA) with significance set at \( P < 0.05 \) and the data are presented as means and SD. Effect sizes for main effects and interactions are presented as partial eta squared (\( \eta^2 \)), while differences between two related samples were evaluated through Cohen’s \( d \), in accordance with Lakens (2013).

### Results

**Physiological responses**

An effect on running speed was observed across both fixed lactate concentrations (\( F = 3.78, P = 0.04, \eta^2 = 0.27 \)). Mean values at 2 mmol/L were ICE 12.3 (1.1) km/h, EXT 12.3 (1.1) km/h, CON 12.0 (1.1) km/h; and at 3.5 mmol/L, ICE 13.8 (1.0) km/h, EXT 13.8 (1.0) km/h, CON 13.6 (1.0) km/h. Bonferroni comparisons identified a difference between ICE and CON (\( P = 0.03, d = 0.15 \)), but not between EXT and CON (\( P = 0.12, d = 0.15 \)), or between ICE and EXT (\( P = 1.00, d < 0.001 \)). The mean blood lactate response is displayed in Fig. 2, while lactate and oxygen uptake during GXT 1 are plotted in Fig. 3. There was no effect of cooling on \( \text{RE} \) [ICE 230 (18) mL kg/km, EXT 230 (17) mL kg/km, CON 227 (13) mL kg/km, \( P = 0.82, \eta^2 = 0.02 \)], nor for \( \text{VO}_2\text{max} \) [ICE 57.5 (5.6) mL/kg/min, EXT 58.4 (4.7) mL/kg/min, CON 57.3 (4.9) mL/kg/min, \( P = 0.69, \eta^2 = 0.04 \)]. No statistical difference in running time until \( \text{VO}_2\text{max} \) during GXT 2 was observed (\( P = 0.707, \eta^2 = 0.03 \)). However, the mean of both precooling groups was greater than CON [368 (79) s, ICE 375 (57) s, EXT 381 (73) s], which equated to a 2% (\( d = 0.11 \)) difference following INT and 3.4% (\( d = 0.17 \)) difference following EXT. Times for EXT were 1.5% (\( d = 0.08 \)) greater than ICE. No statistical

**Comparison of internal and external cooling**

![Fig. 2. Mean lactate response over six incremental submaximal exercise stages. Total time is displayed with error bars displaying standard deviation. Each stage constituted 3-min exercise and 1-min blood sampling, with increments of 1 km/h. Horizontal dotted line indicates blood lactate concentration of 2 mmol/L from which individual running speeds were calculated to represent lactate threshold. All participants completed a minimum of six stages, with some participants completing additional stages before displaying blood lactate concentrations exceeding 3.5 mmol/L. A main effect for cooling type was observed (\( P = 0.04, \eta^2 = 0.27 \)), with differences identified between CON and ICE.](image-url)
difference was found in $\dot{V}O_2\text{max}$ ($P = 0.49$, partial $\eta^2 = 0.08$), although speed after EXT [15.4 (1.3) km/h, ICE 15.0 (1.4) km/h, CON 15.0 (1.7) km/h] equated to a 2.5% difference vs CON ($d = 0.26$) and a 2.8% ($d = 0.31$) difference vs ICE. Speed following ICE was marginally below that of CON (−0.2%, $d = 0.02$). No differences in heart rate ($P = 0.81$, partial $\eta^2 = 0.20$) were observed between groups; CON 146 (16) beats/min, ICE 145 (15) beats/min, EXT 143 (15) beats/min.

No differences were observed throughout the submaximal exercise test in either $V_E$ [mean across exercise: CON 85.6 (12.5) L/min, ICE 85.0 (12.5) L/min, EXT 85.0 (11.6) L/min, $P = 0.90$, partial $\eta^2 = 0.01$] or RER [mean across exercise: CON 0.97 (0.03), ICE 0.97 (0.03), EXT 0.98 (0.06), $P = 0.44$, partial $\eta^2 = 0.08$] were observed across six exercise stages. Similarly, no interaction effects were observed for $V_E$ ($P = 0.149$, partial $\eta^2 = 0.131$) or RER ($P = 0.11$, partial $\eta^2 = 0.16$).

Thermoregulatory responses

Figure 4(a) illustrates mean $T_{CORE}$ data for each condition. After the 10-min rest period, there were no differences between conditions [CON 37.13 (0.23) °C, ICE 37.18 (0.25) °C, EXT 37.13 (0.31) °C, $P = 0.43$, partial $\eta^2 = 0.08$]. During the cooling phase, an interaction between cooling type and time was observed ($F = 25.86$, $P < 0.001$, partial $\eta^2 = 0.70$). Ice slurry ingestion resulted in a greater reduction in $T_{CORE}$ [−0.32 (0.11) °C] than EXT [−0.05 (0.08) °C, $P < 0.001$, $d = 2.97$] and CON [−0.05 (0.07) °C, $P < 0.001$, $d = 3.03$]. No main effect for cooling type during exercise was found ($P = 0.13$, partial $\eta^2 = 0.17$), although a trend toward a higher $T_{CORE}$ for CON is apparent. The failure to detect an effect for cooling type may be explained by the presence of a cooling type × time interaction ($F = 4.38$, $P = 0.01$, partial $\eta^2 = 0.29$). As shown in Table 1, the overall change in $T_{CORE}$ across the exercise phase was greatest during ICE [1.34 (0.27) °C] compared with EXT [1.01 (0.25) °C, $P = 0.001$, $d = 1.29$]. However, there was no statistical difference vs CON [1.11 (0.29) °C, $P = 0.10$, $d = 0.82$] and between CON and EXT ($P = 0.44$, $d = 0.40$). Finishing $T_{CORE}$ for each group at $\dot{V}O_2\text{max}$ were CON 39.03 (0.45) °C, ICE 38.96 (0.55) °C, and EXT 38.88 (0.38) °C.

Figure 4(b) illustrates mean $T_{SKIN}$ data for each condition. A difference was observed between conditions at rest [CON 33.70 (0.5) °C, ICE 33.94 (0.39) °C, EXT 33.48 (0.60) °C, $P = 0.04$ partial $\eta^2 = 0.28$]; however, this effect was not detected by Bonferroni post-hoc. ANOVA revealed a main effect for cooling type ($F = 230.53$, $P < 0.001$, partial $\eta^2 = 0.96$) and a cooling

![Blood lactate vs oxygen uptake during GXT 1. Horizontal dotted line indicates blood lactate concentration of 2 mmol/L from which individual running speeds were calculated to represent lactate threshold. Error bars represent one standard deviation.](image)

**Fig. 3.** Blood lactate vs oxygen uptake during GXT 1. Horizontal dotted line indicates blood lactate concentration of 2 mmol/L from which individual running speeds were calculated to represent lactate threshold. Error bars represent one standard deviation.

<table>
<thead>
<tr>
<th>Cooling type</th>
<th>Δ Total</th>
<th>Δ per 5 min</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Ice slurry</td>
</tr>
<tr>
<td>$T_{CORE}$ (°C)</td>
<td>1.11 (0.29)</td>
<td>1.38*† (0.26)</td>
</tr>
<tr>
<td>$T_{SKIN}$ (°C)</td>
<td>0.71 (0.42)</td>
<td>0.69*† (0.46)</td>
</tr>
</tbody>
</table>

Differences ($P < 0.05$) against control are denoted by * and differences between internal and external cooling groups by †.
type × time interaction \((F = 5.74, P < 0.001, \text{partial } \eta^2 = 0.37)\) during the cooling phase with EXT displaying the lowest mean TSKIN temperatures throughout. EXT also resulted in a greater reduction in TSKIN \([-6.64 (1.46) °C]\) than ICE \([-0.17 (0.52) °C, P < 0.001, d = 6.90]\) and CON \([-0.40 (0.39) °C, P < 0.001, d = 7.62]\). There was no difference between CON and ICE \((P = 0.51, d = 0.52)\). An effect for cooling type was apparent during exercise \((F = 44.20, P < 0.001, \text{partial } \eta^2 = 0.82)\) with EXT \([\text{pre-post: 32.32 (0.6)} – 35.01 (0.59) °C]\) lower than CON \([\text{pre-post: 34.56 (0.55)} – 35.27 (0.67) °C, P < 0.001, d = 1.03]\) and ICE \([\text{pre-post: 34.94 (0.39)} – 35.28 (0.68) °C, P < 0.001, d = 1.29]\). A cooling type × time interaction was observed \((F = 44.14, P < 0.001, \text{partial } \eta^2 = 0.72)\), with a greater rate of increase within EXT resulting in no differences vs CON after stage 4 \((P = 0.058)\) and vs ICE after stage 5 \((P = 0.07)\). Consequently, EXT also displayed the largest overall change in TSKIN during exercise \([2.69 (0.61) °C]\) with post-hoc analysis revealing differences vs CON \([0.71 (0.42), P < 0.001, d = 3.86]\) and ICE \([0.69 (0.46), P < 0.001, d = 3.73]\) as shown in Table 1.

An effect for cooling on PSI during exercise was observed \((F = 6.91, P = 0.005, \text{partial } \eta^2 = 0.41)\), with differences between CON \([\text{CON 5.2 (1.6)}]\) and EXT \([\text{4.6 (1.6), P = 0.02, d = 0.36}]\), and a nonsignificant trend for ICE \([\text{4.58 (1.8), P = 0.058, d = 0.36}]\). A cooling type × time interaction was also discovered \((F = 3.98, P = 0.01, \text{partial } \eta^2 = 0.26)\) because of different rates of increase between groups, with trends displayed in Fig. 5. Sweat rates did not differ between groups \((F = 2.00, P = 0.16, \text{partial } \eta^2 = 1.66)\) with groups’ means as follows: \([\text{mean (SD), percentage of body mass}]\); CON \([1.4 (0.7) \text{ L/h (1.2%)}]\), ICE \([1.6 (0.6) \text{ L/h (1.4%)}]\), and EXT \([1.6 (0.5) \text{ L/h (1.4%)}]\).

Perceptual measures

Precooling reduced TS during exercise \((F = 20.98, P < 0.01, \text{partial } \eta^2 = 0.66)\) with CON \([6.2 (0.8)]\) higher than ICE \([5.7 (0.9), P = 0.005, d = 0.50]\) and EXT \([5.4 (0.8), P < 0.001, d = 0.98]\). However, this reduction did not remain throughout exercise, evidenced by a cooling type × time interaction effect \((F = 4.98, P < 0.001,\)
Discussion

The aims of this study were to compare the physiological responses from internal and external precooling methods during graded exercise tests in the heat. In accordance with our first hypothesis, both precooling interventions resulted in greater running speeds at fixed blood lactate of 2 and 3.5 mmol/L compared with no cooling. However, in contrast to our second hypothesis, no difference in $\text{RPE}$ ($F = 1.96$, $P = 0.54$, partial $\eta^2 = 0.06$) were observed between groups.

Effects of cooling

In this study, fixed blood lactate concentrations of 2 and 3.5 mmol/L were used to represent the lactate threshold and LTP, respectively (Saunders & Green, 2013). This approach accounted for differences in the number of stages completed, removed subjectivity of experimenter identification and provided precision to less than 1 km/h. ICE displayed a statistically significant greater running speed across both markers relative to CON, while a trend was observed for EXT. Such differences may be important given LTP remains a valid predictor of endurance performance in the heat (Lorenzo et al., 2011). Both precooling techniques displayed the same mean difference to CON at 2 (+0.3 km/h) and 3.5 mmol/L (+0.2 km/h), as well as the same overall effect size. As a result, a magnitude-based inference statistical approach may conclude that both interventions had the same effect on lactate indices (Hopkins et al., 2009). Such changes in blood lactate response following precooling are small and likely fall at the upper end of what may be considered day-to-day variation of 0.2 mmol/L for this type of test (Saunders & Green, 2013). However, the modest differences observed were consistent throughout the 23-min trial and constitute a 2% improvement in running speed at the lactate threshold, which may be meaningful as it exceeds the 1.5% coefficient of variation for speed at lactate threshold (Hopkins et al., 2001). For this participant cohort, when running at LTP pace, such a difference would equate to 31 s over 5 km. At elite level, 10 km is typically completed in under 28 min and such a change could equate to an improvement of approximately 16 seconds. Figure 3 illustrates the lactate $\text{VO}_2$ relationship during GXT 1 and provides some further evidence of a trend toward precooling eliciting a modest effect on the lactate thresholds during this test. The lack of difference in ventilation or RER suggests that under this level of heat strain cooling directly elicits an effect on lactate production or clearance rather than an altered $\text{VO}_2$ in the muscle. This is supported by the apparent mediated metabolic strain not transferring to a reduced energetic cost of running in the heat. Despite competing demands for cardiac output for both exercising skeletal muscle and cutaneous vasodilation, it is apparent that muscular blood flow is maintained at submaximal intensities (González-Alonso et al., 2008; Nybo et al., 2014). Therefore, any changes in lactate are unlikely to be explained by cooling eliciting alterations in muscular blood flow. Rather, as thermoregulatory strain leads to a reduction in visceral circulation during exercise (Rowell et al., 1968), it is more likely that by directly cooling $T_{\text{CORE}}$, ICE may elicit a heat sink away from the skin and maintain the rate of lactate-pyruvate conversion in the liver. Similarly, by substantially lowering $T_{\text{SKIN}}$, EXT may also reduce demands for skin blood flow, preserving splanchnic circulation and enhancing lactate clearance. An increase in splanchnic blood flow during exercise has...
previously been suggested as a mechanism that explains the enhancement in LTP following heat acclimation (Lorenzo et al., 2010). It should be noted that the alterations in the lactate response occurred at moderately elevated levels of T_CORE, with mean T_CORE 38.3 °C in CON after 23 min of graded exercise. Thus, these results warrant an investigation into whether a greater effect of cooling on lactate occurs when individuals are hotter, and where metabolic alterations would be expected to be more pronounced.

RE incorporates both biomechanical as well as physiological parameters and is thought to account for differences between elite athletes who display similar VO_{2max} values (Bassett & Howley, 2000). Despite indications that precooling may benefit some determinants of RE such as oxygen uptake (Lee & Haymes, 1995), stride length (Folland et al., 2006), and neuromuscular function (Siegel et al., 2011), no differences were found, supporting the results of Winne & Yates (2008) who examined RE following ice slurry precooling. Thus, beneficial effects of precooling do not appear to present through improved RE while exercising for relatively short durations in the heat.

Despite evidence that maximum oxygen consumption is attenuated in the heat (Lorenzo et al., 2011), cooling had no effect on subsequent VO_{2max}. González-Alonso and Calbet (2003) have demonstrated that under heat stress, cardiac output and mean arterial pressure reduce at maximal exercise intensities, leading to a reduction in skeletal muscle blood flow. Thus, a cooler body would be expected to mediate the decline in VO_{2max} under heat stress through reducing competition for blood flow and maintaining cardiac output and mean arterial pressure for longer. Such a mechanism is supported by the maintenance of skeletal muscle blood flow and VO_{2max} in thermoneutral, relative to hot, conditions (Périard et al., 2011). However, it would appear that thermoregulatory effects of both cooling techniques were no longer present at the point of VO_{2max}, as evidenced by the similar finishing T_CORE between cooling techniques. The time course of the current protocol shares similarities with many athletic events, whereby a precooling intervention must occur prior to a warm-up, with individuals completing 24–32 min of exercise, which may culminate in an all-out end-spurt. It remains plausible, however, that cooling may still elicit an effect on VO_{2max} if a greater cooling impulse is provided or VO_{2max} is measured during a shorter duration protocol.

Interestingly, despite reductions in body temperature and TS, RPE remained unchanged following both cooling methods. This may indicate that enhanced performance in the heat and associated pacing adjustments following precooling are closer linked to TS rather than overall perceived exertion. Such a relationship could have implications for future cooling techniques more aggressively targeting locations that determine TS.

**Comparison of internal and external cooling**

Responses to internal and external precooling

The physiological responses from each cooling technique were similar, with neither ICE nor EXT having an effect on VO_{2max} or RE, and the magnitude of the effect on lactate similar between techniques. Although a small mean difference suggests toward enhanced vVO_{2max} following EXT, no statistical difference between treatments was observed. Further, while a change of 2% (0.4 km/h) in EXT compared with ICE and CON could be interpreted as meaningful, this change is below what has been suggested to constitute a meaningful difference of 0.5 km/h (Billat & Koralsztein, 1996). Therefore, it would appear that the estimate of the maximum speed that can be maintained by oxidative phosphorylation is similar, irrespective of cooling method.

Although both precooling maneuvers produced a similar, lowered PSI throughout the trial, there were markedly different thermoregulatory responses, which may determine application. While ICE resulted in a 0.3 °C reduction in T_CORE, in keeping with other literature (Ihsan et al., 2010; Ross et al., 2011; Siegel et al., 2012), EXT did not elicit a reduction in T_CORE. This is not uncommon following external cooling techniques, as an “after-drop” may be observed, whereby T_CORE remains unchanged during cooling, before falling at the start of exercise as vasoconstriction dissipates and warm blood from the core is subsequently cooled in the periphery. While an after-drop was not observed through a reduction in T_CORE, the rate of increase in T_CORE during exercise following EXT was smaller than both ICE and CON. The lack of an after-drop may be attributable to differences in the time course and intensity of exercise following cooling compared with other research (Kay et al., 1999). Similarly, Duffield et al. (2009) did not report a reduction in T_CORE following EXT and subsequently observed a reduced T_CORE throughout exercise. Both Ucket and Joch (2007) and Duffield et al. (2010) have reported performance benefits following external cooling techniques that did not elicit an initial reduction in T_CORE, as greater pace may be achieved through a reduced rate of heat storage or T_SKIN. Such a lower T_SKIN may be associated with reduced thermal discomfort (Gagge & Gonzalez, 1974) and appears to be a key mediator of behavioral thermoregulation, contributing toward a greater selected intensity during self-paced prolonged exercise (Schlader et al., 2011). Indeed, the size of reduction in thermal strain relative to CON was greatest following EXT, in accordance with a reduced T_SKIN. ICE is thought to affect performance through an enhanced absolute heat storage capacity that prevents or delays CNS motor drive reduction, again permitting greater exercise intensity. In addition to an enhanced capacity for storing heat, functional magnetic resonance imaging has indicated that thermoreceptors within the splanchnic region may activate pleasure centres of the brain, possibly leading to a deceptive effect concerning


Comparison of internal and external cooling


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