Title

Intermittent sprint performance in the heat is not altered by augmenting thermal perception via L-Menthol or Capsaicin mouth rinses.

Short title

Thermal perception and intermittent sprint performance in the heat

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Number of figures/tables

Figures – 5

Tables – 1

Total word count

4,164

Abstract word count

250

Key words

Cooling; Heat stress; Sprint performance; Temperature; Thermal comfort; Thermal sensation; Thermal perception.

Abbreviations

Core temperature (T_{rec})

Cycling intermittent sprint protocol (CISP)

Heart rate (HR)

Intermittent Sprint Performance (ISP)

Maximum aerobic power output (W_{max})

Mean Power (MP)
Nude body mass (NBM)
Peak oxygen uptake ($\dot{V}O_{2peak}$)
Peak power (PP)
Rating of perceived exertion (RPE)
Skin temperature (T_{skin})
Thermal comfort (T_{com})
Thermal sensation (T_{sens})
Time trial (TT)
Transient receptor potential channels (TRP)
TRP ion channel Melastatin 8 (TRPM8)
TRP ion channel Vanilloid 1 (TRPV1)
Whole Body Sweat rate (WBSR)
Work done (WD)

Abstract

Purpose. Cooling sensations elicited by mouth rinsing with L-Menthol have been reported as ergogenic. Presently, responses to L-Menthol mouth rinsing during intermittent sprint performance (ISP) in the heat are unknown and the impact of increased thermal perception on ISP via Capsaicin has also not been quantified. This experiment aimed to identify whether eliciting cooling/warming sensations via L-Menthol/Capsaicin would alter ISP in the heat.

Method. Fourteen participants [mass=72±9 kg, $\dot{V}O_{2peak}$=3.30±0.90 L.min^{-1}], undertook four experimental trials, involving 40 min of ISP in hot conditions (40.2±0.6°C, 42±2%R.H.) with mouth rinsing (25 mL, 6 sec) at the protocol onset, and every 10 min thereafter. Cooling (0.01%L-Menthol; MEN), warming (0.2%Capsaicin; CAP), placebo (0.3 sham-CHO; PLA) and control (water; CON) mouth rinses were utilised. Performance was quantified via power (PP) and work done (WD) during sprints. Heart rate (HR), core (T_{rec}) and skin (T_{skin}) temperature, perceived exertion (RPE), thermal sensation (T_{sens}) and comfort (T_{com}) were measured at 10 min intervals. Sweat rate (WBSR) was calculated from ∆mass.

Result. PP reduced over time (P<0.05), however no change was observed between trials for PP or WD (P>0.05). T_{com} increased over time and was lower in MEN (2.7±1.1; P<0.05) with no difference between CAP (3.1±1.2), PLA (3.2±1.3) and CON (3.1±1.3). RPE, T_{sens} HR, T_{rec}, T_{skin} increased over time (P<0.05) with no between trial differences (P>0.05).

Conclusion. Despite improved thermal comfort via L-Menthol, ISP did not improve. Capsaicin did not alter thermal perception or ISP. The reduction in ISP over time in hot conditions is not influenced by thermal perception.

Introduction

In comparison to equivalent exercise demands in temperate conditions, heat stress impairs endurance (Kenefick et al. 2007) and intermittent (Girard et al. 2015) exercise performance. In cycling (Racinais et al. 2015b) and running (Guy et al. 2014) modalities, continuous intensity/aerobic endurance-type exercise performance diminishes with increased environmental temperature along a continuum once the magnitude of heat stress increases above preferable ambient temperatures of 10-15°C (Galloway and Maughan 1997). Impaired performance of intermittent sprint activity replicating team sports e.g. football/soccer, rugby league and union, field hockey, basketball and netball, during heat stress has been characterised (Hayes et al. 2014), albeit to a lesser extent than continuous exercise in spite of intermittent/repeated sprint exercise providing a greater heat strain (Maxwell et al. 1996). It has been reported that football/soccer players performing at the 2014 FIFA World Cup demonstrated a reduction in the number of sprints and total distance covered under high heat stress vs moderate or low heat stress (Nassis et al. 2015), an observation shared by others (Konefal et al. 2014; Watanabe et al. 2017). Conversely, within that same study it was identified that technical parameters (rate of successful passes) were equal or enhanced in high heat stress vs moderate or low stress matches (Nassis et al. 2015) suggesting that 'pacing' of high intensity work is occurring either in response to physiological or perceptual stimuli (Girard et al. 2017).

Detrimental performance under heat stress is closely related to the elevations in cardiovascular strain under heat stress (González-Alonso et al. 2008), which occur to attenuate elevations in core/deep body temperature via elevated skin blood flow and redistribution of cardiac output (González-Alonso 2012), ultimately to maximise evaporative cooling (Candas et al. 1979). Two experimental games of football played in hot (~43°C) or temperate (~21°C) conditions evidenced these responses with muscle and core temperature ~1°C higher in the hot games (Mohr et al. 2012; Nybo et al. 2013). These temperature responses were not accompanied by elevated cardiovascular strain. However, this is likely a response to aforementioned reductions in physical performance whereby distance covered (-7%) and high intensity running (-26%) were lower in the hot conditions, presumably to moderate the magnitude of cardiovascular strain i.e. the players were incurring similar physiological strain for reduced work. In a follow up study utilising the same data (Nybo et al. 2013), no difference in maximal voluntary contraction, voluntary activation and peak twitch torque, nor the magnitude of glycogen depletion occurred in the football matches played in the hot or temperate conditions, suggesting these pathways are not directly contributing to reduced physical performance in the heat. Thermally driven pacing during intermittent team sport type activity has been evidenced in the laboratory whereby relative to temperate conditions, hot-wet and hot-dry conditions elicit an earlier and greater reduction in peak power output during 40 min of high intensity

sprints (Hayes et al. 2014). The conscious/subconscious pacing of performance becoming apparent given equality of performance during the final sprint/end spurt across conditions inspite of earlier reductions in performance (Hayes et al. 2014). It has previously been identified that cooling at a physiological and perceptual level (Castle et al. 2006; Duffield et al. 2010), can enhance self-paced exercise performance under heat stress in laboratory conditions. This suggests in these scenarios that attenuated physiological or perceived temperature is ergogenic and therefore has the potential to improve intermittent sprint performance. Conversely, under conditions of equivalent core temperature, deception of temperature (Castle et al. 2012) and subsequent reductions in RPE leads to an improvement in performance, suggesting perception of temperature is at least in part responsible for performance detriments rather than the physiological temperature alone (within the range of internal temperatures elicited by these experiments).

Eliciting alterations in thermal perception in the absence of any physiological differences in temperature has been termed ‘non-thermal cooling’ or ‘non-thermal warming’. Non-thermal cooling can be administered by topical, facial applications of L-Menthol [an activator of the transient receptor potential (TRP) ion channel Melastatin 8 (TRPM8) (Montell and Caterina 2007)] to elicit alterations in thermal sensation to that of fan cooling, and Capsaicin [an activator of the TRP ion channel Vanilloid 1 (TRPV1) (Montell and Caterina 2007)] has been used to elicit non-thermal warming eliciting equivalent changes in thermal perception to that of a heater (Schlader et al. 2011). Non-thermal cooling sensation of L-Menthol improved mean power output to the same extent as the actual cooling (+21% during RPE-clamped exercise), whilst Capsaicin and actual warming reduced power output relative to cooling trials (Schlader et al. 2011).These data inform that self-paced fixed intensity exercise performance such as time trials or time to task failure/exhaustion in the heat can be modulated by thermoregulatory behaviour. Given the difficulties in applying non-thermal cooling via facial applications during fixed intensity exercise performance the ergogenic potential of non-thermal cooling via L-Menthol in the form of a mouth rinse has received significant attention. A recent review concluded that a mouth rinse or a beverage containing L-Menthol during endurance exercise in the heat is beneficial for performance (Stevens and Best 2017). It was stated that ergogenic benefits for performance are contingent on altering thermal perception/perceived exertion (Mündel and Jones 2010; Stevens et al. 2016, 2017; Flood et al. 2017; Jeffries et al. 2018). This notion is supported by the observation that protocols which did not report altered perceptual responses following oral L-Menthol do not demonstrate a performance enhancement (Sönmez et al. 2010; Riera et al. 2014, 2016). At present only one experiment has investigated the use of L-Menthol [combined with cool fluid (0.2 or 3.0°C)] during interval-type activity (Five repetitions of 4 km cycle and 1 km running TT performance) whereby no ergogenic improvement was attributable to L-Menthol (Tran Trong et al. 2015). To the authors knowledge, no data exists investigating the use of L-Menthol during intermittent sprint activity replicating that of team sports. This is surprising given

153 the natural breaks in play during team sport performance would facilitate mouth rinsing at the same
154 time as habitual drinking (Garth and Burke 2013), something not as plausible in a continuous, fixed
155 intensity competitive endurance event such as track running/cycling.
156
157 The aim of this experiment was to determine whether eliciting a cooling sensation via oral L-
158 Menthol, or a warming sensation via oral Capsaicin would alter intermittent sprint performance in
159 the heat in comparison to control and placebo oral solutions. It was hypothesised that L-Menthol
160 would reduce (enhance) thermal sensation and improve performance, whilst Capsaicin would
161 increase (diminish) thermal sensation and decrease performance.

Methods

Participants

Fourteen healthy, non-heat-acclimated, trained team sports players volunteered to participate in the study (Participant characteristics are presented in Table 1). Initially sixteen participants volunteered and commenced the experiment, however two withdrew having completed two and three visits for reasons unrelated to the experiment. Confounding variables of caffeine and alcohol consumption 24 hours prior to testing and prolonged thermal e.g. exercise heat acclimation protocols, repeated sauna or hot tub use, or hypoxic exposures e.g. altitude training in the 6 weeks prior to testing, were all controlled for in line with previous work involving exercise-heat stress and intermittent sprinting (Hayes et al. 2014; Gibson et al. 2014). Following institutional ethics approval (2732-MHR-Jul/2016-3430-2) and full description of experimental procedures, all participants completed medical questionnaires and provided written informed consent following the principles outlined by the declaration of Helsinki of 1975, as revised in 2013.

Insert table 1 near here please

Experimental design

The protocol consisted of five visits. The first visit was an incremental test to determine peak oxygen uptake ($\dot{V}O_{2peak}$) and maximum aerobic power output (W_{max}). During the same visit, the subjects completed a familiarisation trial of 20 min of a cycling intermittent sprint protocol [CISP; The full CISP is 40 min in duration and involves, 20x2 min blocks of 10 sec passive rest, a 6 sec sprint against 7.5% of body mass, and 104 sec of active recovery at a power eliciting 35% $\dot{V}O_{2peak}$ determined from regression of $\dot{V}O_2$ against power from the incremental test (Castle et al. 2011)] in hot conditions [40°C, 50% relative humidity (RH)]. This served to minimise the negative effects of initial heat exposure and any subject learning effect associated with the protocol (Hayes et al. 2014). Then, a minimum of 48 hours later, in a randomised and cross-over design, participants commenced the experimental trials (visits 2-5). Female participants performed experimental trials during the follicular phase of the menstrual cycle (Mee et al. 2015; Lei et al. 2016). Trial order was counterbalanced using a Latin square based upon 16 participants. Each experimental visit to the laboratory was separated by a minimum of 48 hrs to allow for a full-recovery between trials, and to mitigate against physiological adaptation to the heat (Gill and Sleivert 2001). All exercise tests were carried out on a friction braked cycle ergometer (Monark 724, Vansbro, Sweden), operating in a pedal-rate independent mode.

Experimental trials involved participants performing a CISP in a heat chamber maintained at ~40°C, ~50% RH. In the experimental trials, subjects periodically swilled an L-Menthol, Capsaicin, water or

orange-flavoured placebo solution at four time points. Each mouth rinse was 25 mL in volume and administered at rest and at 10 min intervals thereafter i.e. after every 5th sprint during the 10 s passive rest phase of the CISP protocol. The participants were instructed to swill/gargle the solution around the mouth for 5 sec and then expectorate the solution into a bowl without swallowing. The Menthol solution (MEN) was a 0.01% concentration of L-Menthol crystals ground and dissolved in distilled water (≥99% food grade L-Menthol, Sigma Adrich, UK) in line with previous experiments (Mündel and Jones 2010; Stevens et al. 2016). The Capsaicin solution (CAP) was administered at a concentration of 0.2% with the Capsaicin containing red pepper sauce (Tabasco Habanero Sauce, McIlhenny Co., Avery Island, CA, USA) diluted in distilled water. These differences in concentrations were used based upon pilot data which identified them to elicit an equal magnitude of perceptual change in thermal sensation of the oral cavity + 0.5 (MEN) or - 0.5 (CAP) on the scale (Toner et al. 1986) at rest, in thermoneutral conditions. Orange-flavoured fruit squash (Tesco Ltd, UK) solution mixed with distilled water to create a 0.5% (0.3g/100 mL) placebo-CHO solution (PLA) (Carter et al. 2004). Distilled water served as the control (CON). To minimise the impact of drink temperature on perceptual, or physiological responses (Lee and Shirreffs 2007), all fluids were maintained to the temperature within the heat chamber (~40°C), this also served to ensure that the L-Menthol crystals remained dissolved in water and ensured minimal pre/per-cooling effect given the close proximity of the rinse temperature to that of the oral cavity.

Preliminary testing

Prior to the initial assessment of $\dot{V}O_{2peak}$ and W_{max} in the preliminary trial, standing height (cm) was measured via a stadiometer and nude body mass (kg) was recorded following self-reported measurement in a private bathroom (SECA 875 scale, Birmingham, UK). These data were used to calculate body surface area (Du Bois and Du Bois 1916). Body density was also calculated using calipers (Harpenden, Burgess Hill, UK) and a four site skinfolds calculation (Durnin and Womersley 1974). This was later used to calculate body fat [%, (Siri 1956)].

The incremental test was conducted in temperate lab conditions (21°C, 50% RH). Starting intensity was set at 80 W with resistance subsequently applied to the flywheel to elicit a 24 W.min^{-1} increase at the constant cadence of 80 rpm. Expired metabolic gas was measured using online gas analysis (Oxycon Pro, Jaeger GmbH or Metalyser Sport, Cortex, Leipzig, Germany); the $\dot{V}O_{2peak}$ was considered as the highest $\dot{V}O_2$ obtained in any 10 sec period. Heart rate (HR; bmin^{-1}) was recorded continually during all exercise tests by short range telemetry (Polar Electro Oyo, Temple, Finland). Saddle position was adjusted by the participant to their preferred cycling position and remained unchanged for all experimental trials. Experimental workloads (i.e. active recovery at 35% of

$\dot{V}O_{2peak}$) were subsequently calculated using linear regression utilising power: $\dot{V}O_2$ data collected following the incremental test.

After 25 ± 5 min recovery, a 20 min stabilisation period (seated rest), and ten stages (20 min) of the CISP were completed in the heat chamber (~40°C, ~50% RH). Following stabilisation, a standard warm-up [5 min at 95 W [80 rpm] and two 30 sec bouts at 120 W [100rpm] with 30 sec rest in between (Hayes et al. 2012, 2014)] was followed by the 10 x 2 min blocks of the CISP i.e. ten instances of 10 sec passive rest, a 6 sec sprint against 7.5% of body mass, and 104 sec of active recovery at a power eliciting 35% $\dot{V}O_{2peak}$ (Castle et al. 2011).

Experimental procedure

Each experimental trial was conducted at the same time of day (within participants) to control for the effects of circadian variation in performance (Drust et al. 2005). To minimise differences in starting muscle glycogen concentrations between visits, subjects recorded their diet in the 24-hour period before the second visit and were instructed to follow the same diet before each subsequent visit. Energy or macronutrient intake was not quantified in the experiment. Participants consumed 500 mL of water 2 hours before all preliminary and experimental exercise sessions to ensure adequate hydration. Once participants were deemed euhydrated [i.e. urine osmolality (Alago Vitech Scientific, Pocket PAL-OSMO, UK) was < 700 mOsm kg^{-1} H_2O (Sawka et al. 2007)], they were subsequently able to commence further preliminary, and experimental procedures. Following confirmation of adequate hydration, in private, participants measured their nude body mass (NBM) inserted a single-use disposable rectal thermistor (Henleys Medical, UK, Meter logger Model 401, Yellow Springs Instruments, Yellow Springs, Missouri, USA) 10 cm past the anal sphincter to facilitate the measurement of rectal temperature (T_{rec}). Skin temperature (T_{sk}) was measured using a data logger (Squirrel Meter Logger, Grant Instruments, Cambridge, UK) and skin thermistors secured at four sites (pectoralis major muscle belly, lateral head of triceps brachii, rectus femoris muscle belly, and lateral head of the gastrocnemius) on the right-hand side of the body using 6 cm x 7 cm transparent Tegaderm patches (3M, UK). Weighted mean skin temperature was determined using a four-site formula (Ramanathan 1964). Heart rate was recorded continually in the same manner as the preliminary test.

Participants then mounted a cycle ergometer located inside the environmental chamber where conditions were consistent across trials (~40°C, ~50% RH) and performed the warm up followed by the full 40 min CISP. During the CISP, the Monark Anaerobic software (Monark Anaerobic Wingate Software, Version 1.0, Monark, Vansbro, Sweden) recorded peak power (PP), mean power (MP), and work done (WD) at a sampling frequency of 50 Hz, during each sprint within of each 2 min

stage of the CISP. Peak power was recorded as the highest recorded power output value for each sprint, MP was determined as the average power output from all values recorded during the 6 s sprint. WD was calculated cumulatively over the whole protocol. Physiological measurements were recorded at rest (0 min) and then 60 sec into the active recovery phase after every 5th sprint (every ~10 min) thereafter. Perceptual responses were also recorded at 10 min intervals with this frequency least likely to cause non-experimental artefacts (Corbett et al. 2009). Perceptual responses included whole body thermal comfort (T_{com}) and thermal sensation (T_{sen}) determined on a five (from 1, comfortable, to 5, very uncomfortable) and seventeen (from 0.0, unbearably cold, to 8.0, unbearably hot) point scale respectively (Toner et al. 1986), and the rating of perceived exertion (RPE) measured using a 15-point Borg scale (from 6, very very light, to 20, very very hard) (Borg 1982). Following completion of the protocol, the participants towel dried and recorded NBM for the later calculation of whole body sweat rate (WBSR, L.hr^{-1}).

Data analysis

Data are presented as mean ± SD (n = 14) unless otherwise indicated. All statistical analyses were carried out using the SPSS software (Version 25). All outcome variables were first checked for normality and sphericity. The Greenhouse Geisser correction for the *F* statistic and related degrees of freedom was used when data violated sphericity. The data from the CISP was 'blocked" into an average of each 10 min series of sprints/recovery sprints. A two-way repeated measures analysis of variance (ANOVA) was performed to determine differences in dependent variables associated with performance (PP, MP; W.kg^{-1}) during each of the four 10 min blocks between the four mouth rinse trials. Perceptual measures of RPE, T_{Sen}, T_{Com}, and physiological measures of T_{rec}, T_{skin}, and HR were compared between the four trials, and across five timepoints (0, 10, 20, 30, 40 min). WBSR and WD was analysed using a one-way repeated measures ANOVA between the four mouth rinse trials. Main and interaction effects were followed up with Bonferroni adjusted post hoc comparisons. Partial Eta squared (η_p^2) as used as an estimate of the effect size for main and interaction effects [0.01 = small, 0.06 = medium, 0.013 = large (Cohen 1992)], Cohen's d_{av} ($d_{av;}$) was used as an estimate of the effect size [0.2 = small, 0.5 = medium, 0.8 = large (Cohen 1992)] for post-hoc comparisons. The threshold for rejecting the null hypothesis was set at $p<0.05$.

Results

Effect of mouth rinse on sprint performance

There was no main effect of mouth rinse, or interaction effect between mouth rinse and time, on peak power (Figure 1), mean power or total work done (Figure 2) (PP mouth rinse: $F_{(2,27)}$<0.1, p=0.967, η_p^2=0.003, mouth rinse x time: $F_{(4,54)}$=0.4, p=0.776, η_p^2=0.034, MP mouth rinse: $F_{(3,39)}$<0.1, p=0.981, η_p^2=0.005, mouth rinse x time: $F_{(4,51)}$=0.9, p=0.447, η_p^2=0.067, Work done (kJ) mouth rinse: $F_{(3,39)}$=0.4, p=0.735, η_p^2=0.032).

Insert figure 1 near here please

Peak power (W.kg^{-1}) and mean power (W.kg^{-1}) decreased during the first 3 blocks (main effect of time, PP: $F_{(1.3,16)}$=10.6, p=0.003, η_p^2=0.450, MP: $F_{(1,15)}$=14.3, p=0.001, η_p^2=0.524). PP and MP were significantly lower during sprints 6-10 compared to sprints 1-5 (PP: p=0.027, d_{av}=0.3, MP: p=0.018, d_{av}=0.3), and significantly lower during sprints 11-15 compared to sprints 6-10 (PP: p=0.002, d_{av}=0.3, MP: p<0.001, d_{av}=0.3). Sprint performance data are presented in Figure 1 and Figure 2 with data tables included as electronic supplementary material.

Insert figure 2 near here please

Perceptual responses to mouth rinse

There was a main effect of mouth rinse on T_{Com} ($F_{(3,39}$=2.9, p=0.046, η_p^2=0.183), where T_{Com} was lower after L-Menthol (2.7±1.1) than after carbohydrate mouth rinse (3.2±1.3, p=0.042, d_{av}=0.4). There was no difference between any other mouth rinse conditions and no interaction effect between mouth rinse and time ($F_{(6,82)}$=0.8, p=0.548, η_p^2=0.061).

T_{Com} ($F_{(2,31)}$=121.0, p<0.0001, η_p^2=0.903), T_{Sen} ($F_{(2,20)}$=216.1 p<0.0001, η_p^2=0.943) and RPE ($F_{(2,28)}$=399.1, p<0.0001, η_p^2=0.968) increased throughout each trial where all perceptual responses were higher than at the previous time point (T_{Com}; all p<0.003, d_{av}>0.6, T_{Sen}; all p<0.0001, d_{av}>0.7, RPE; all p<0.0001, d_{av}>0.7). Perceptual responses to mouth rinse are displayed in Figure 3 with data tables included as electronic supplementary material.

Insert figure 3 near here please

There was no main effect of mouth rinse and no interaction between mouth rinse and time on T_{Sen} or RPE (T_{Sen} mouth rinse: $F_{(3,39)}$=0.6, p=0.639, η_p^2=0.042, mouth rinse x time: $F_{(5,66)}$=1.2, p=0.313, η_p^2=0.085, RPE mouth rinse: $F_{(2,26)}$=0.6, p=0.543, η_p^2=0.046, mouth rinse x time: $F_{(12,156)}$=1.2, p=0.289, η_p^2=0.084).

337

338 Physiological responses to mouth rinse

339 There was no main effect of mouth rinse, or interaction effect between mouth rinse and time, on T_{rec},
340 $T_{skin,}$ HR or WBSR (T_{rec}: mouth rinse: $F_{(3,39)}$<0.1, p=0.967, η_p^2=0.003, mouth rinse x time: $F_{(3,44)}$=1.1,
341 p=0.355, η_p^2=0.079, T_{skin} mouth rinse: $F_{(3,39)}$=0.1, p=0943, η_p^2=0.010, mouth rinse x time: $F_{(3,44)}$=0.9,
342 p=0.465, η_p^2=0.064, HR mouth rinse: $F_{(3,39)}$=0.4, p=0.769, η_p^2=0.028, mouth rinse x time: $F_{(4,56)}$=0.5,
343 p=0.769, η_p^2=0.035, WBSR mouth rinse: $F_{(3,39)}$=0.2, p=0.925, η_p^2=0.012). Physiological responses are
344 presented in Figure 4 and Figure 5 with data tables included as electronic supplementary material.

345

346 ***Insert figure 4 near here please***

347

348 Physiological responses are presented in Figure 4 and Figure 5 with data tables included as electronic
349 supplementary material. T_{rec} (°C), $T_{skin,}$ (°C) and HR (b.min^{-1}) increased over time (T_{rec}: $F_{(1,15)}$=150.7,
350 p<0.001, η_p^2=0.921, T_{skin}: $F_{(2,21)}$=78.4, p=0.001, η_p^2=0.858, HR: ($F_{(2,26)}$=294.6 p<0.001, η_p^2=0.958).

351

352 T_{rec} was higher at every time point than the previous time point (all p<0.001, all d_{av}>0.8). The same
353 trend occurred for T_{skin} and HR (all p<0.02, d_{av}*>0.4)*, though the differences between 20 minutes and
354 30 minutes (T_{skin}: p=0.051, d_{av}=0.3, HR: p=0.316, d_{av}=0.3) were not statistically significant.

355

356 ***Insert figure 5 near here please***

Discussion

The aim of this experiment was to determine whether eliciting a cooling sensation via oral L-Menthol, or a warming sensation via oral Capsaicin would alter intermittent sprint performance in the heat in comparison to control and placebo oral solutions. Identical physiological responses (Figures 4 and 5) between mouth rinse conditions demonstrate equality of the physical capacity to perform intermittent sprints in the heat. Enhanced thermal comfort [-0.5 vs PLA ($p < 0.05$), and -0.4 vs CAP and CON] following L-Menthol stimulation of the TRPM8 ion channel (Figure 3), suggested that the participants were more 'perceptually tolerant' of the physiological heat stress, therefore in line with the work of others, had the potential to perform better given a reduction in perceived heat stress. This improved thermal perception via L-Menthol did not however alter intermittent sprint performance (Figure 1 and Figure 2). Opposing the experimental hypothesis, the Capsaicin mouth rinse elicited no detrimental effect on performance, perception or physiological responses with all data comparable to the placebo and control. Accordingly, the targeted TRPV1 channel was insufficiently modulated by our Capsaicin rinse protocol.

Our data contrasts evidence of performance enhancement during fixed intensity tasks following oral menthol interventions from cycling time to exhaustion/task failure experiments [+9% (Mündel and Jones 2010), +8% (Flood et al. 2017) and +6% (Jeffries et al. 2018)] and preloaded running time trials of 3 km and 5 km [TT; -3% (Stevens et al. 2016) and -3.5% (Stevens et al. 2017)]. The common theme amongst the studies demonstrating a benefit of Menthol are reports of improved thermal perception (Stevens et al. 2016, 2017; Flood et al. 2017; Jeffries et al. 2018), or reduced perceived exertion (Mündel and Jones 2010). Our data adds to the equivocal findings relating to the use of oral Menthol interventions which have reported no ergogenic effects during performance trials e.g. 400 m running TT (Sönmez et al. 2010), 20 km (Riera et al. 2014) and 30 km cycle TT (Riera et al. 2016). It is noteworthy that improved thermal perception (Riera et al. 2014) and perceived exertion (Riera et al. 2016) do not necessarily improve performance. In support of these equivocal data relating to the importance of thermal /exercise perception on performance following oral menthol, it has also been observed that menthol spray to the torso offers no ergogenic effect during 16.1 km (Barwood et al. 2015) or 40 km cycle TT performance (Barwood et al. 2012), or during a pre-loaded 5 km running TT (Barwood et al. 2014) despite improved thermal sensation. Combining Menthol with neck cooling also elicits no difference in exercise performance (time to task failure) in comparison to abdominal or non-menthol neck cooling, or in comparison a control trial, in spite of improved thermal sensation [vs no intervention (Bright et al. 2018)]. The role of improved thermal or exertional perception via menthol remains equivocal with no consensus on which perceptual metric for thermal/exertion feelings is most affected by L-Menthol despite their proposed importance during continuous, fixed-intensity tasks (Flood 2018). Our data, highlights that during intermittent sprint exercise, in spite of improved thermal perception of equivalent magnitudes to

others [specifically thermal comfort, but not thermal sensation as per (Flood et al. 2017)], menthol elicits no ergogenic benefit.

Only one experiment has considered the role of L-Menthol, and subsequent manipulations in thermal perception on intermittent exercise performance (Tran Trong et al. 2015). The intermittent exercise in this study was unaffected by L-Menthol, however the long, higher intensity intermittent bouts in this study do not reflect the activity profiles of field-based team sports making comparisons with the current protocol inappropriate. A number of other experiments have implemented supramaximal intermittent sprint exercise (similar to the CISP) and altered either thermal perception i.e. sensation or comfort, or physiological temperature. During intermittent sprint exercise in the heat for example, ice slurry consumption reduced core temperature (start of protocol -0.5°C, end of protocol -0.3°C) and thermal sensation (-3) but did not influence the total distance covered or speed during jog, run and sprint phases of the protocol (Gerrett et al. 2017). As skin temperature and RPE were not different, the authors concluded, in congruence with previous work (Duffield and Marino, 2007), that combined alterations in core and skin temperature are necessary for influencing intermittent sprint performance in the heat (Gerrett et al. 2017). The CISP has been utilised in experiments where various manipulations of temperature perception and actual temperature have occurred. It has been observed that, in comparison to control conditions, a cooling intervention using ice packs (local cooling of exercising muscle) can reduce muscle temperature (ice packs -0.6°C vs control), core temperature (-0.2°C) and heart rate (-7 $b.min^{-1}$ vs control) at the end of a CISP performed in hot conditions (Castle et al. 2006). The result of these temperature manipulations was increased PP (+4%) and WD (+2%) in the ice packs only condition (Castle et al. 2006). This performance data, coupled with no differences in either RPE or T_{sens} between cooling and control trials, suggests that the performance ability during intermittent sprints in the heat is more closely related to physiological responses, and particularly local muscle temperature, rather than perceived temperature. This provides an explanation for why, despite improved thermal comfort with L-Menthol, intermittent sprint performance was unchanged in the present study. Support for this mechanism can be found when examining the independent impact of heat acclimation, and pre-cooling interventions on CISP in the heat. Only when implementing a heat acclimation intervention that induces reductions in core temperature (-0.4°C), heart rate (-18 $b.min^{-1}$) *and* thermal sensation (-1.0) did PP improve in the heat (+2%) with pre-cooling (thermal and non-thermal cooling) offering no additional benefit (Castle et al. 2011). This appears a different mechanism to that observed during 5,000m running in the heat where pre-cooling demonstrates an ergogenic effect independently (TT duration -3.7%), and when used in conjunction with heat acclimation (TT duration -7.0%) greater performance enhancement than heat acclimation alone (TT duration -6.6%) (James et al. 2018). These findings highlight that whilst physiological intervention is a priority for intermittent sprinting in the heat (Castle et al. 2006, 2011), physiological and perceptual manipulations should

be conferred for continuous intensity, endurance performance in the heat (James et al. 2017), suggesting there may still be benefits associated with oral menthol in this domain (Stevens and Best 2017; Flood 2018).

Experimental Considerations and Future Directions

In light of the small adjustments to thermal comfort, and no observed change in thermal sensation, it could be suggested that despite replicating dosages utilised in previous experimental work (Mündel and Jones 2010; Stevens et al. 2016) the concentration of L-Menthol (0.01%) was insufficient to elicit changes in behavioural thermoregulation. This comment can also be made in regard to the exploratory inclusion of our Capsaicin rinse. Recent experimental work has subsequently identified that the concentration of L-Menthol (concentrations range 0.05-0.105% L-Menthol at 0.05% increments when dissolved in Ethanol, rather than water) being mouth rinsed does not alter thermal perception within individuals (Best and Berger 2017). Accordingly, although the rinse frequency in the present study replicates the timing used by Mündel and Jones (Mündel and Jones 2010) a more frequent rinse in line with Stevens et al., (Stevens et al. 2016) may be ergogenic during intermittent sprint and warrants further investigation. Understanding the impact of alterations in L-Menthol mouth rinse temperature may be required to understand the role this plays in intermittent sprint performance given our experiment utilised a drink temperature similar to deep body temperature to minimise the influence of visceral temperature modulation (Morris et al. 2018), but (for experimental control) this temperature was greater than that likely consumed in actual competition and this may be one reason for different findings compared to studies using cooler mouth rinses (Stevens and Best 2017). It is likely that the Capsaicin ‘dose’ used in this experiment was suboptimal to achieve our experimental aim. The use of Capsaicin mouth rinse (to make mechanistic inference rather than as an ergogenic aid) to increase thermal perception therefore requires further refinement allied to application, concentration and frequency given the lack of a response across dependent variables, an opposing finding to that of topical Capsaicin (Schlader et al. 2011). The present experiment did not utilise sufficient techniques to understand the mechanisms, and level at which L-Menthol cooling is ergogenic. To elucidate this, techniques such as peripheral nerve stimulation and transcranial magnetic stimulation (Goodall et al. 2014; Twomey et al. 2017) to determine central and peripheral components to the reduction in workload associated with intermittent sprint exercise in the heat, with and without alterations in perception via L-Menthol. The intensity of continuous endurance activity is closer to the submaximal intensity proposed as subject to influence via precooling (Duffield and Marino 2007), than the supramaximal sprint efforts within the CISP suggesting perceived temperature plays a lesser role in modulated short sprint exercise of a fixed number and frequency. Performance during submaximal phases of an intermittent sprint protocol which is most likely to be influenced by cooling, rather than the sprint itself (Duffield and Marino 2007), Regrettably, this was not quantifiable in the present experiment given the fixed -intensity nature of

our active recovery protocol but the notion that peak sprint performance is less likely to be influenced by actual/perceptual cooling was observed (Figure 1). These comments raise an important point relating to the protocol used in the present study. Whilst the CISP is a reliable and valid protocol to determine the physiological responses to intermittent sprinting (Hayes et al. 2012, 2014), the specific task structure is such that the CISP is closed in nature. This closed task potentially creates an experimental artefact whereby participants are not able to sprint freely (in frequency or duration). Future work should consider the benefits of an 'open-ended' task e.g. devising a protocol with an undefined sprint duration, or participant regulated sprint frequency, performed on a non-motorised treadmill where participants could pace independently in response to non-thermal cooling interventions in the heat. This approach is such that it may facilitate different responses due to the elevation in pacing associated with these tasks, differentiating it from the CISP which implements fixed duration and fixed frequency of sprinting. In light of alterations in both low and high intensity running with heat stress (Konefal et al. 2014; Nassis et al. 2015; Watanabe et al. 2017), changes in these movement velocities could also be quantified using this approach in a similar manner to other work (Gerrett et al. 2017). Finally, the CISP is only 40 min in duration and therefore only replicates the first half of a field-based team sport. Future work should therefore extend the task duration to more closely replicate the team sport of interest (Turner et al. 2014), in elite team sport players.

Practical Implications

These data suggest that the ergogenic potential of L-menthol associated with endurance tasks does not extend to intermittent sprint performance (of 40 min) in very hot temperature conditions (~40°C). This is in spite of the magnitude in alteration in thermal perception in the present experiment being congruous with others (Stevens et al. 2017; Flood et al. 2017) meaning our null finding relating to performance was not simply a result of a null finding relating to altering thermal perception as is the case with the Capsaicin data. As such, rather than seeking perceptual manipulations alone, individuals seeking to use an intervention to enhance intermittent sprint performance e.g. team sport players, should preference either pre cooling, or mid (per) cooling interventions as an acute ergogenic aid (Castle et al. 2006; Luomala et al. 2012; Sunderland et al. 2015), or a heat acclimation strategy e.g. the preferable isothermic approach (Racinais et al. 2015a; Pryor et al. 2018), to induce enhanced physiological responses to intermittent sprinting as part of a chronic intervention (Sunderland et al. 2008; Castle et al. 2011) in a manner that minimises training disruption (Gibson et al. 2015).

Conclusion

Mouth rinsing with L-Menthol improves thermal comfort during 40 min of intermittent sprint exercise in the heat but this does not appear to alter ISP. Capsaicin did not alter thermal perception or ISP. No mouth rinse utilised changed the physiological responses to ISP in the heat. Alterations to the intervention e.g. mouth rinse frequency/duration subsequently leading to a greater change in thermal perception, or an open loop ISP may induce ergogenic responses however based on the present data, the reduction in ISP over time in hot conditions is not influenced by altering thermal perception.

Acknowledgements

We would like to express our gratitude to the participants who voluntarily took part in the experimental study and to Tom Howes, Craig Stout, Atharva Tere, Dominic Mitchell and Michael Polycarpou for their support with the data collection.

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Figure titles

Figure 1. Mean±95%CI Peak power output during the sprint blocks across the menthol, capsaicin, carbohydrate and water mouth rinse conditions. * Denotes difference from previous sprint block with no difference between groups

Figure 2. Mean±95%CI Total work done during sprints across the menthol, capsaicin, carbohydrate and water mouth rinse conditions

Figure 3. Mean±95%CI Thermal comfort (A), Thermal sensation (B), and Rating of perceived exertion (C) across the menthol, capsaicin, carbohydrate and water mouth rinse conditions, for all time points. * Denotes difference from previous sprint block. † denotes significant difference in menthol.

Figure 4. Mean±95%CI Core temperature (A), Skin temperature (B), and Heart rate (C) across the menthol, capsaicin, carbohydrate and water mouth rinse conditions, for all time points. * Denotes difference from previous sprint block with no difference between groups

Figure 5. Mean±95%CI Sweat rate across the menthol, capsaicin, carbohydrate and water mouth rinse conditions

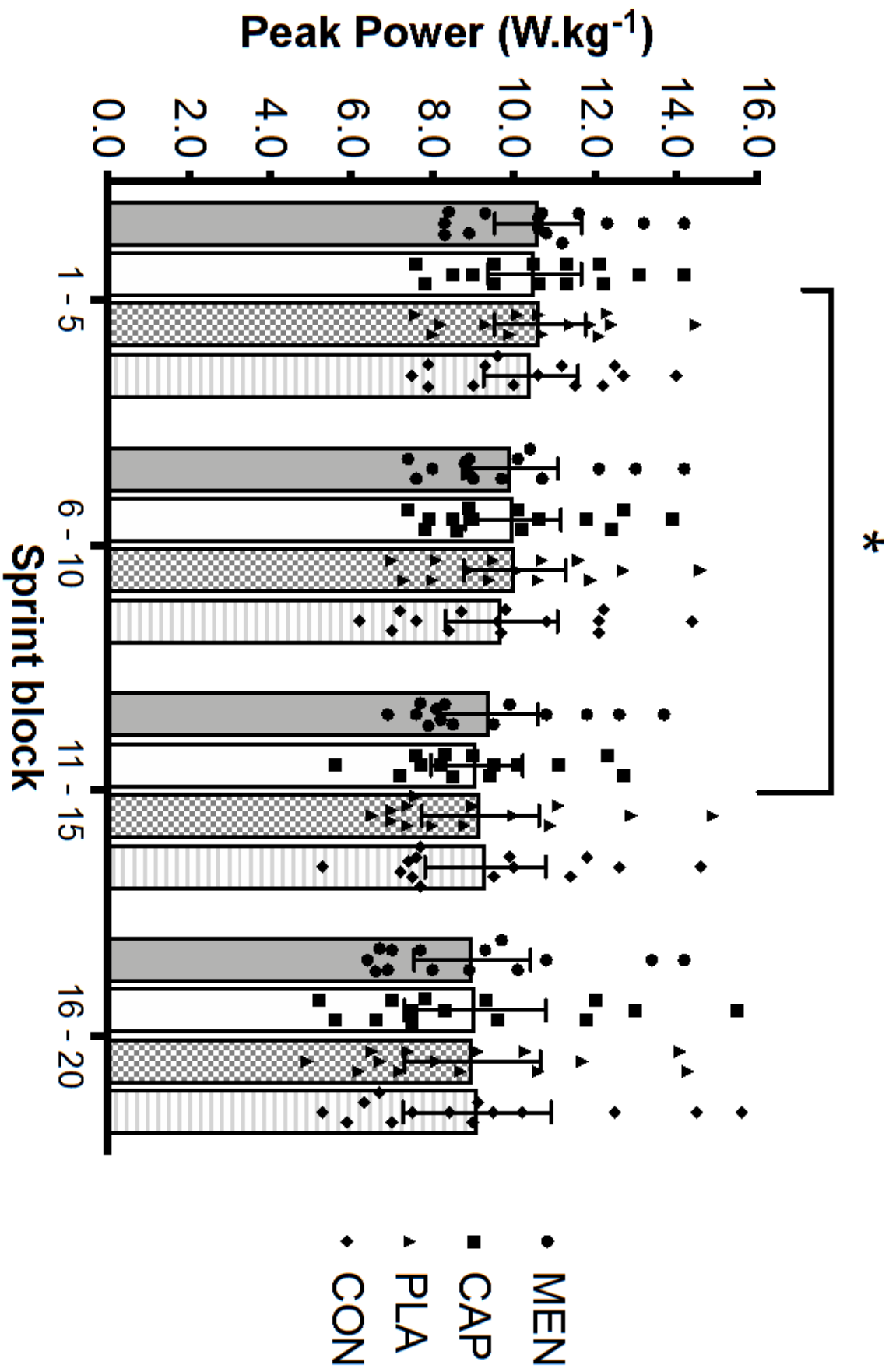
714 **Table**

715 Table 1. Mean±SD Participant characteristics (n = 14; 11 males, 3 females)

Variable	Mean ± SD
Age (years)	24 ± 3
Height (cm)	175 ± 12
Mass (kg)	71.6 ± 8.8
Body surface area (m^2)	1.86 ± 0.17
Body mass index ($kg.m^2$)	23.4 ± 2.2
Body fat (%)	11.6 ± 3.1
Maximal oxygen uptake ($L.min^{-1}$)	3.29 ± 0.89
Maximal oxygen uptake ($mL.kg^{-1}.min^{-1}$)	46.2 ± 12.9
Power at maximal oxygen uptake ($W.kg^{-1}$)	3.8 ± 1.3
Power eliciting 35% $\dot{V}O_{2peak}$ ($W.kg^{-1}$)	1.3 ± 0.5

716

717 Figure 1
718


Peak Power (W.kg-1)
0.0
2.0
4.0
6.0
8.0
10.0
12.0
14.0
16.0
Sprint block
1 - 5
6 - 10
11 - 15
16 - 20
*
MEN
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719

720 Figure 2

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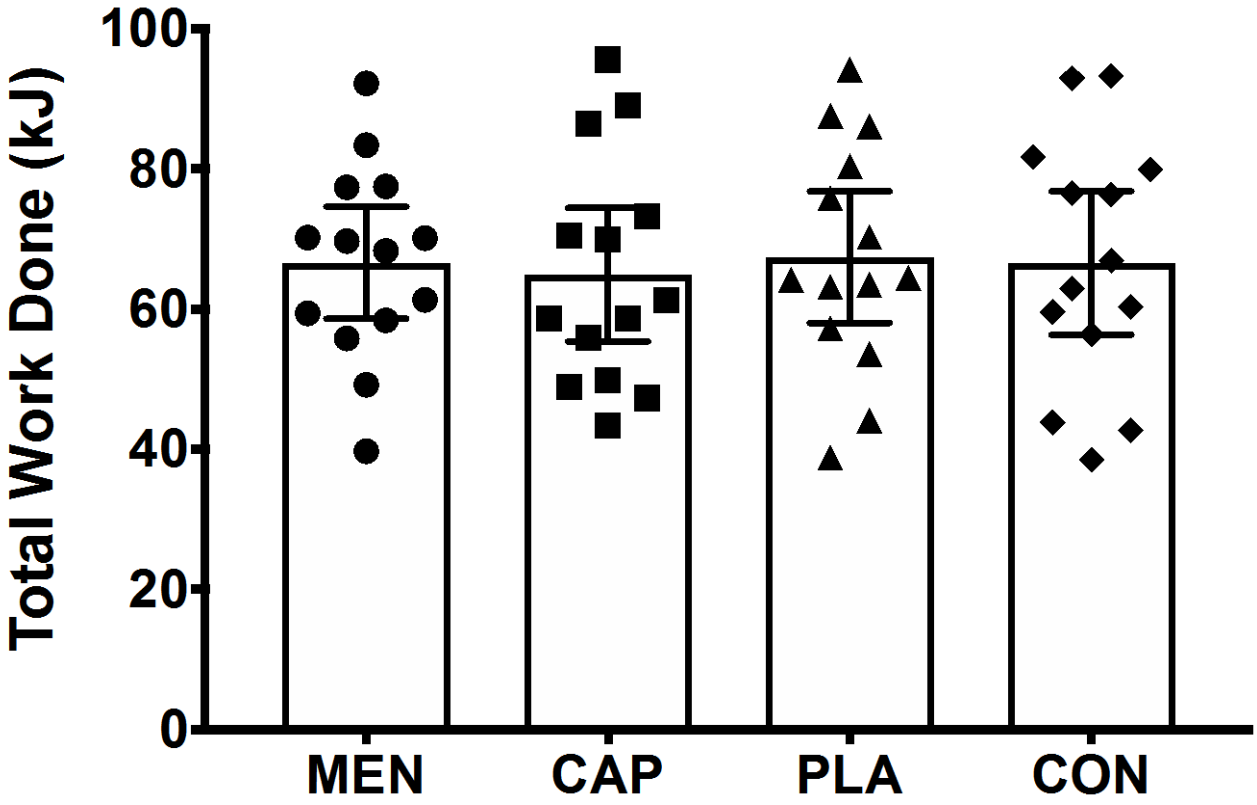

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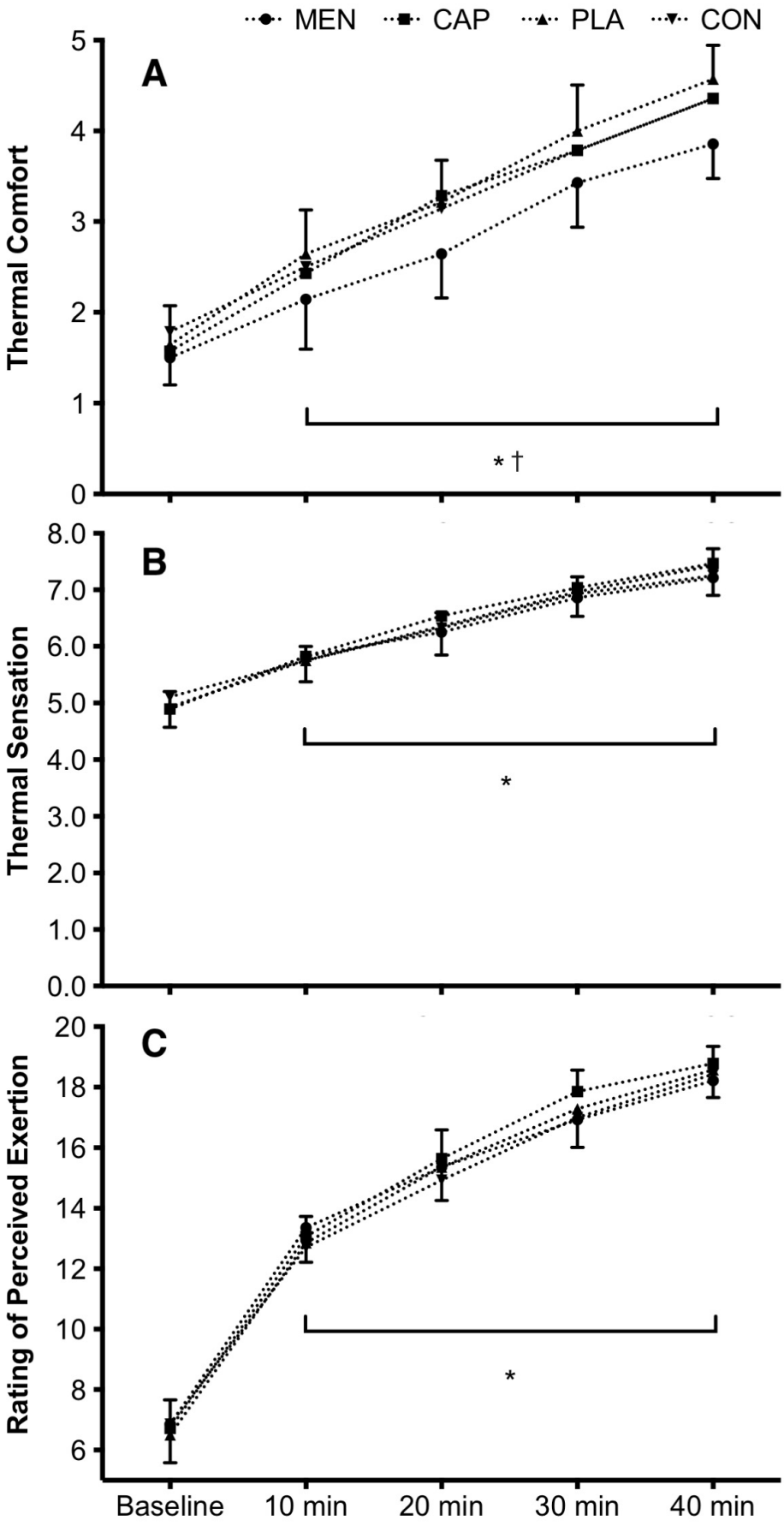
725 Figure 3



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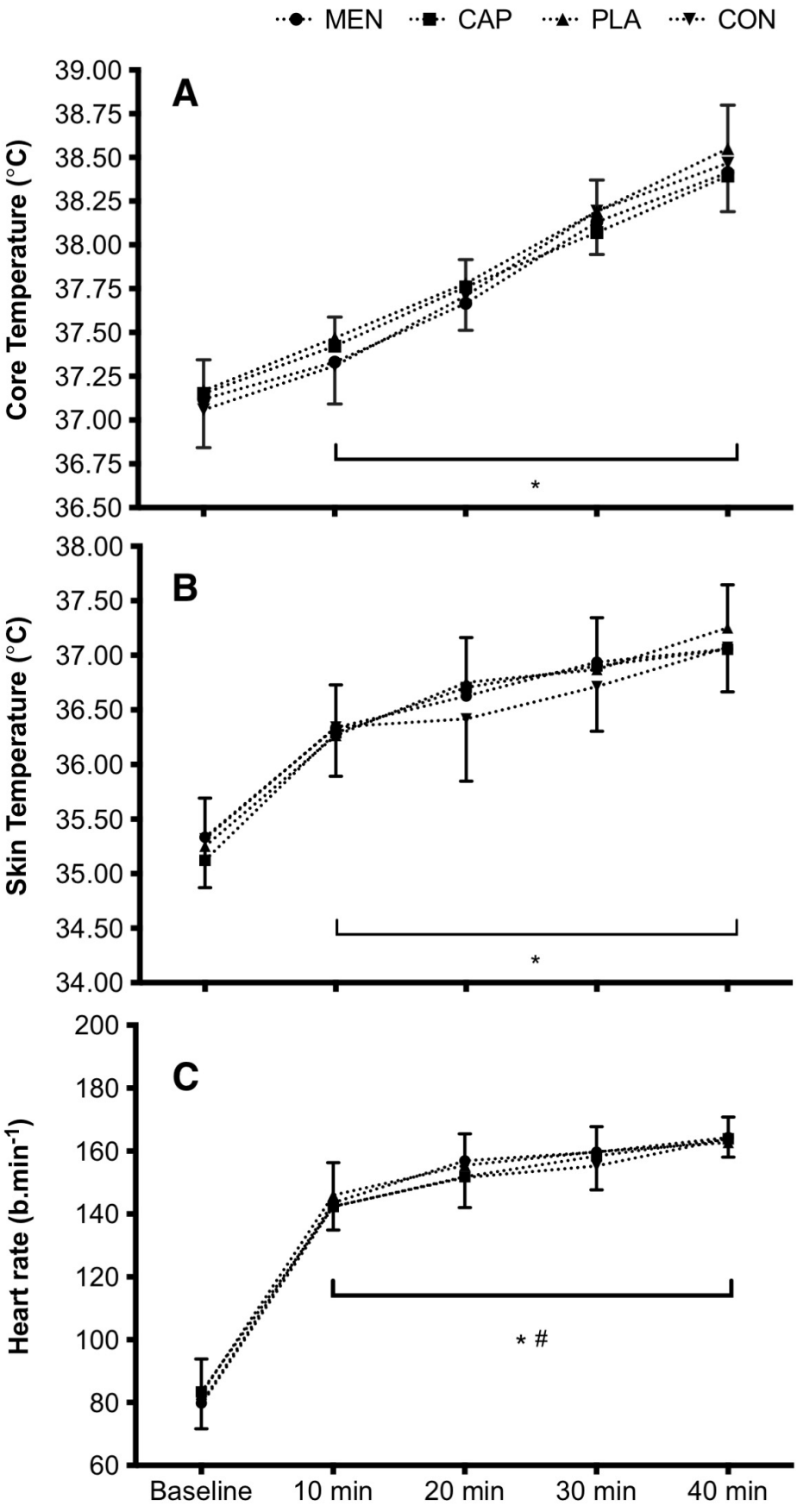
728 Figure 4



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731 Figure 5

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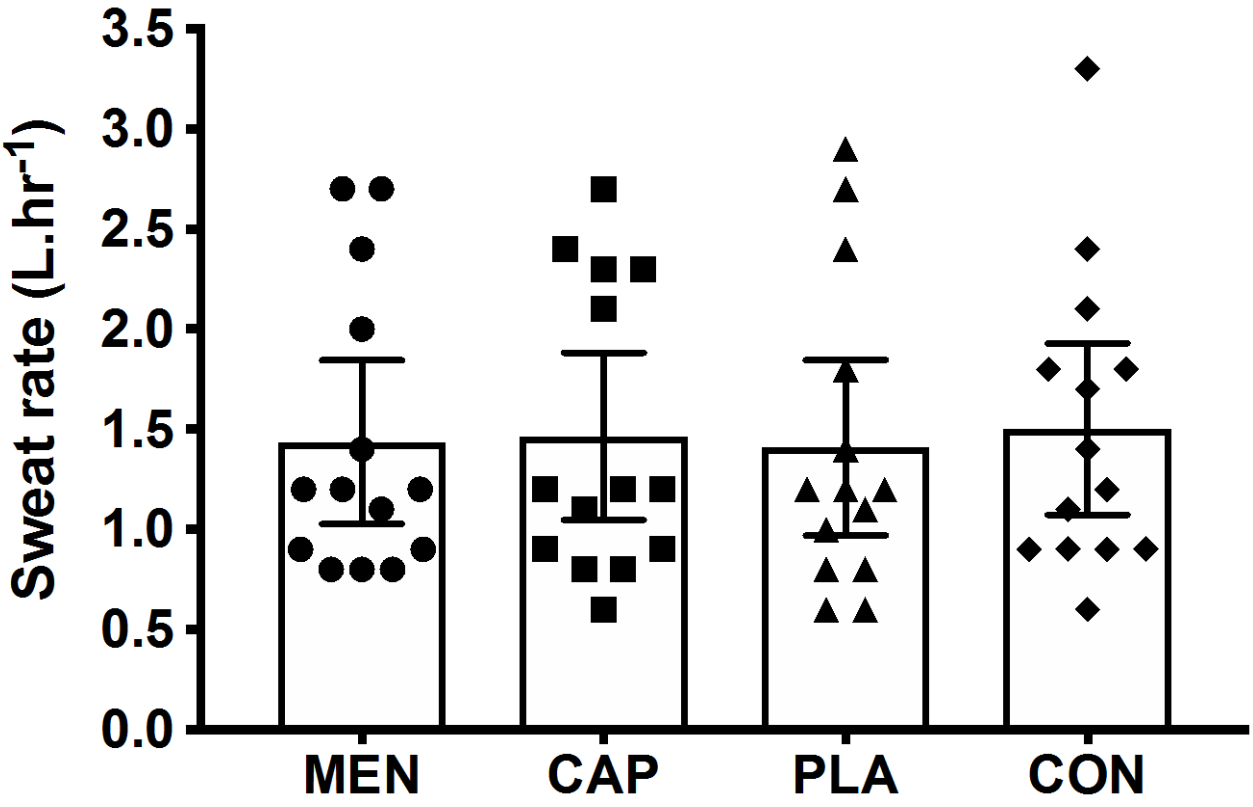

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