Running Head: Performance and L-Menthol

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Spraying with 0.20% L-Menthol does not enhance 5k running performance in the heat in untrained runners

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Abstract

Aim: L-Menthol stimulates cutaneous thermoreceptors and induces cool sensations improving thermal comfort but has also been linked to heat storage responses. Therefore, L-Menthol application could lead to a conflict in behavioural and thermoregulatory drivers improving comfort but leading to a higher rate of deep body temperature rise; the present study examined this possibility. **Method:** Six untrained male participants **(**age 21[1] years; height 1.80 [0.07]m; mass 78.9 [6.9]kg; surface area 1.98 [0.13]m^2) took part. They completed three trials in hot conditions (34°C) where their clothing was sprayed (CONTROL-SPRAY or MENTHOL-SPRAY) or not sprayed (CONTROL) after a fixed intensity exercise period (15-minutes), which induced thermal discomfort, before completing a 5 km treadmill time trial (TT). Thermal perception (thermal sensation and comfort; TS, TC), thermal responses (aural temperature [T_{au}], skin temperature [T_{skin}]), perceived exertion (RPE), heart rate, pacing (1 km split time) and performance (TT completion time) were measured. **Results:** MENTHOL-SPRAY induced improvements in TS (up to 3 km of TT) and TC (up to 1 km) with T_{au} showing a tendency to be higher than CONTROL-SPRAY (+0.20 [0.29]°C) and CONTROL condition (0.30 [0.34]°C); this was not statistically significant and the rate of rise in T_{au} was linear. T_{au} was continuing to rise between the 4th and 5th kilometre of the TT. The other variables were unchanged. TT completion time and pace were not different: CONTROL 27.92 [1.65], CONTROL-SPRAY 28.10 [1.12], MENTHOL-SPRAY 27.53 [2.85] minutes. **Conclusions:** Spraying L-MENTHOL prior to exercise in the heat culminated in improved perception but not altered performance.

Key words: Thermal perception, ergogenic aid, exercise pacing, heat-illness

Introduction

The application of L-Menthol to the skin stimulates cool sensations mediated by specialized sensory neurons (1). These cells feature a highly sensitive receptor, TRPM8, which is activated either by temperatures ranging from 8 to 28 °C, or by chemical compounds such as L-Menthol (2,3). Gillis and colleagues (4) recently demonstrated that the application of L-Menthol to the torso (0.2 % or 0.05 % in 100mL solution) significantly improved thermal sensation (towards feeling cooler) in hot (31 °C), moist (70 %RH) ambient conditions during exercise. Gillis et al. (4) required their participants to exercise at two pre-set intensities (50 and 70% of a pre-trial [separate days] maximal power cycling test) in order to fix metabolic heat production which enabled them to examine whether there were any differences in thermoregulatory effector responses induced by menthol. Their data suggested that the effector response following the application of the 0.05 % L-Menthol solution was not different to that of a control solution (containing surfactants only) despite differences in thermal perception; this raised the novel possibility that application of L-Menthol at 0.05 % concentration could be used to separate thermal perception from thermal state when contrasted to a control spray (5). At the higher concentration examined, Gillis et al. (4) found that 0.20 % L-Menthol altered thermal sensation to a greater extent, but also triggered heat storage responses (possibly vasoconstriction) that culminated in higher rectal temperatures. However, the extent of the higher rectal temperature was never in excess of 0.2°C and mean skin and mean body temperature were unaffected. Similarly, Kounalakis et al. (6) described a higher rate of rectal temperature rise during fixed intensity exercise (60% VO_2max) when 4.6 g per 100 mL of menthol cream (~ 4.6 %) was applied over the entire body surface area. Consequently, participants reached 38°C deep body temperature an average of 7.8 minutes quicker during exercise. The onset of sweating was also delayed with the gain in the response

also affected. However, this study was performed in relatively temperate (24°C; 46 % RH) conditions.

The study design utilised by Gillis et al. (4) and Kounalakis et al. (6) did not attempt to determine whether there was any performance benefit associated with the improvement in thermal perception and this possibility remains. It has been suggested that thermal perception is a conscious and salient driver in changing exercise intensity (7) and in such circumstances alleviation of thermal discomfort may maintain or enhance performance in the heat thereby influencing behavioural thermoregulation (8). Recently we demonstrated that there was no significant alteration in performance or pacing strategy during 40 km time trial cycling exercise in the heat when 0.05 % L-Menthol (in solution) was applied to the torso prior to exercise in trained participants (5); this menthol concentration corresponded to the lowest menthol concentration used by Gillis et al. (4). However, the menthol was applied to the torso at a point when participants were not experiencing thermal discomfort. We speculated that this widened the range of thermal perceptual experiences during exercise in the heat but at a time when perception was not a primary driver of pacing and performance (*i.e* participants were not uncomfortably hot). A study where menthol is applied to the torso when participants were experiencing thermal discomfort may illuminate whether thermal perception is a meaningful initial driver of exercise pacing.

The data of Gillis et al. (4) raise another important consideration. Given that exercise performance in the heat could be limited by stored heat (9) and may be influenced by the feeling of thermal discomfort (8), a concentration of L-Menthol of 0.20 % has the potential to make persons exercising at an uncompensable intensity feel cooler (*i.e.* more comfortable) but store heat at a faster rate, if a higher work intensity is self-selected, leading to a conflict in

thermoregulatory and behavioural drivers. Moreover, commercially available 'performance enhancing' body sprays typically contain, in combination with alcohol, L-Menthol at a concentration of 0.20 % (i.e. Physicool, TM London, U.K) and recommend the application of 100 mL of solution. Such concentrations have yet to be shown, by an independent research study, to enhance exercise performance in the heat. It is apparent that at higher concentrations, L-Menthol is a potent vasoactive substance that can interfere with thermoregulation throughout a range of normothermic and hyperthermic body temperature states (6). Accordingly, the present study aimed to examine whether the application of 0.20 % L-Menthol to athletic clothing improves high intensity exercise performance in hot conditions and whether altered thermal sensation after thermal discomfort has been induced, is a primary driver of exercise behaviour. Based on the findings of Gillis et al (4) it was hypothesised that higher deep body temperatures would be achieved during a performance test after the application of L-Menthol (H_1) whereas the data of Barwood et al. (5) indicate that perception may be improved (H_2) but pacing and performance would remain unchanged (H_3).

Materials and Methods

The protocol was approved by the University of Portsmouth Research Ethics Committee and experiments conformed to the declaration of Helsinki. Participants provided written informed consent. Six recreationally active males (age 21 [1] years; height 1.80 [0.07] m; mass 78.9 [6.9]kg; surface area 1.98 [0.13]m^2; 10) volunteered to participate. An inclusion criteria of a 5 km completion time of 25 minutes or less was set for the study. They abstained from alcohol, caffeine consumption and strenuous exercise 24 hours prior to the test and were non-smokers.

Experimental Design

The study used a within participant, single-blind, repeated-measures design. Participants first completed a familiarisation trial followed by randomised completion of a CONTROL,

CONTROL-SPRAY and MENTHOL-SPRAY treatment condition. All trials took place on separate days at the same time of day (± 1 hour) with a minimum of 48 hours between tests.

Procedure

Following arrival at the laboratory the participant voided, and naked body mass was measured (Ohaus digital weighing scales, I-10, Canada). Prior to dressing, the participant was instrumented with a calibrated, insulated aural thermistor (T_{au}; Grant Instruments Ltd, Cambridge [Shepreth], U.K), skin thermistors at eight different sites (Grant Instruments Ltd, Cambridge [Shepreth], U.K) and a heart rate monitor; mean skin (T_{skin}) and mean body temperature (T_b) were subsequently calculated according to the equations of Olesen (11) using a weighted average of skin temperature at the bicep, chest, foot, hand, hamstring, quadriceps, scapular and shin and Colin et al. (12) respectively.

Participants then dressed and wore the same socks, running shoes and long sleeve t- shirt (the latter provided by the experimenters; 100 % polyester) on each occasion. Thereafter the participant entered an environmental chamber set to a dry bulb temperature of ~34°C and 55 % RH, which was measured by a WBGT weather station (1250 series, Squirrel Data Logger, Grant Instruments Ltd, Cambridge [Shepreth], U.K). They then sat on a chair situated on a calibrated, motorised treadmill (Powerjog GX220, Powerjog, London, UK) for a period of 5-minutes for the collection of pre-exercise resting data. Thereafter the chair was removed and the participant commenced exercise at a fixed intensity for 15-minutes (10 or 12 $km.h^{-1}$; fixed within participant between conditions based on thermal perception [achieving thermal discomfort] and achieving a corresponding sub-maximal heart rate response [this averaged 153 [5] $b.min^{-1}$] in the warm up of the familiarisation trial). At the start of the exercise period a fan positioned 1 metre from the participant (Lloytron 16” fan, Model FO59, Lloytron

Electronics Ltd) and pointed in the direction of the participant's torso, was switched on. The wind speed produced by the fan was verified at a fixed position on the treadmill before and after the experiment by an anemometer (Meterman Anemometer, Model TMA10, Meterman Tests Tools, China; this approximated 1 to 1.5 $m.s^{-1}$). Participants were permitted to consume water *ad libitum* throughout the trial (water temperature ~19 °C). Participants gave perceptual ratings of RPE (13) thermal comfort (TC; 14) and thermal sensation (14) every 5 minutes of the fixed intensity period (TS & TC using a 20 cm visual analogue scale).

At the end of the 15-minute period the treadmill was stopped and the participant rested for 5-minutes during which time the fan was switched off. In two of the experimental conditions (CONTROL-SPRAY and MENTHOL-SPRAY) the participant's t-shirt was sprayed evenly (performed by the same experimenter on each occasion) with 100 mL of solution; the spray volume was measured using calibrated weighing scales situated within the environmental chamber (Mettler PC 400, Mettler Instrumente AG, Greifensee, Zurich, Switzerland) and the spraying procedure took approximately 3-minutes. The clothing was sprayed rather than the skin to minimise losses to the solution dripping from the skin. Towards the end of the 5-minute period the participant stood up and was reminded that they were to exert a maximal effort and cover the 5 km distance as quickly as possible in the subsequent TT; they received feedback only of distance covered. At this point the participant commenced exercise and the fan was switched back on. The time taken to complete each 250 m split of the TT was noted throughout the exercise test and participants provided perceptual votes at each 1 km of the TT. The participant continued to exercise until 5 km was completed, they reached volitional exhaustion or achieved a withdrawal deep body temperature of >39.5°C. At the end of the TT the participant exited the environmental chamber, was de-instrumented and re-weighed naked

behind a privacy screen. Fluid intake and pre and post trial naked body weights (OHAUS I-10 digital scales, Canada) were used to estimate sweat production.

Description of Sprays

Sprays were produced by an independent chemical consultant (Chemical Associates, Rosemead, Frodsham, United Kingdom). The CONTROL SPRAY contained 3% surfactants mixed in water, while the MENTHOL SPRAY contained a concentration of 0.20 wt/wt L-Menthol in 3% surfactants plus water. Sprays were stored at room temperature (20°C) and transferred into the chamber ~3 h before testing. In order to minimise supplementary perceptual cooling associated with a spray temperature lower than exercising skin temperature and ambient temperature, the bottles containing the sprays were immersed in a temperature controlled water bath held at 34°C within the chamber (Tempette Junior TE 8J, Techne, Cambridge, U.K), 1-hour before the trial commenced.

Data Analyses

Mean (SD) were calculated for each condition for the final data point of the rest period (*i.e.* 5-minutes after the end of the fixed intensity period) and on completion of every kilometre of the TT for all variables (T_{au}, T_{au} rate of rise, T_{skin}, T_b, cardiac frequency derived from heart rate [*f*c] RPE, TS, TC) with the exception of RPE [not examined at rest] and performance data [no data point at rest].

Comparisons were made between each condition and across time using factorial ANOVA with repeated measures. Assumptions of sphericity were checked using Mauchley's test and adjusted where necessary (Greenhouse-Geisser). Statistically significant effects were determined *post-hoc* using pairwise comparisons. Comparisons were also made between

conditions for fluid consumption and sweat production using a one-way ANOVA. The coefficient of variation (CV) within participant across trials was also calculated. The stability of ambient conditions were examined using repeated measures analysis of variance (ANOVA). The alpha level for all statistical tests was set at 0.05. ANOVAs were calculated using PASW version 18 (SPSS Inc, Chicago, Illinois). Given the low sample size, and where appropriate, statistical power to interrogate null findings were performed to alpha level of 0.05 power of .80 using mean (SD) differences between conditions; this was conducted using Minitab version 15 (Minitab Inc. USA). CV was calculated using Microsoft Excel.

Results

Environmental Conditions

There were no significant differences in the environmental conditions between the trials. Mean (SD) across conditions for the dry bulb, wet bulb and calculated WBGT were 33.9 (.10)°C; $F_{(2,10)} = 1.715$, $p = .229$; 26.2 (.30)°C; $F_{(2,10)} = 2.217$, $p = .160$) and 28.5 (.20)°C; $F_{(2,10)} = 1.616$, $p = .246$) respectively.

Perceptual Responses

At the end of the fixed intensity exercise period (*i.e.* immediately before participants were sprayed) the TS and TC averaged 15.5 (1.4) cm and 9.8 (3.0) cm across conditions which corresponded to the descriptors *warm to hot* and *just uncomfortable*; there were no differences between conditions at this stage (TS: CONTROL *cf* CONTROL-SPRAY $p = .931$ and MENTHOL, $p = .909$, MENTHOL *cf* CONTROL-SPRAY $p = .823$; TC: CONTROL *cf* CONTROL-SPRAY $p = .123$ and MENTHOL, $p = .529$, MENTHOL *cf* CONTROL-SPRAY $p = .744$). Following spraying and during the TT the TS showed significant differences between conditions (main effect for condition: $F_{(1.632,\ 8.160)} = 15.953$, $p = .002$) and an

interaction effect between condition and time ($F_{(2.281, 11.407)}$ = 7.979, p = .006). The TS votes were significantly lower (*i.e.* towards feeling cooler) in the MENTHOL condition compared to the CONTROL (p = .007) and the CONTROL-SPRAY (p = .002). As the TT ensued, and differences in the MENTHOL-SPRAY condition was sustained up to 3 km but were no longer different at the 4 km and 5 km distance point; significant differences between conditions are summarised in Figure 1A. All participants completed the TT stage.

INSERT FIGURE 1 NEAR HERE

TC neared being different between conditions (condition effects: $F_{(1.380, 6.900)}$ = 3.899, p = .083) but did show an interaction effect between condition and time ($F_{(3.444, 17.222)}$ = 3.063, p = .050). The interaction effects were similar, but not as strong, as the TS responses with improved comfort evident in the first 1 km of the TT in the MENTHOL-SPRAY condition relative to other two conditions; significant differences between conditions are summarised in Figure 1B.

RPE data did not differ significantly between condition (condition effects: $F_{(2,10)}$ = .045, p = .956) or show any interaction effect ($F_{(3.505, 17.527)}$ = 3.505, p = .672). At the equidistant time points the RPE vote was always within 1 point between conditions on the rating scale and culminated in a vote of 19 (1) across conditions at the end of the TT. Towards the start of the TT (*i.e.* after 1 km), when the menthol spray was evidently active the RPE vote was 12 (2), 13 (2) and 13 (1) in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY conditions respectively.

Thermal and fc Responses

The MENTHOL and CONTROL-SPRAYS did not significantly influence T_{au}, T_{skin} and T_b at the start or during the TT at any stage (condition effects: T_{au}: $F_{(2,10)} = 2.393$, $p = .142$; T_{skin}; $F_{(1.042,5.210)} = 1.839$, $p = .209$; T_b: $F_{(2,10)} = .401$, $p = .680$). The T_{au} response in the MENTHOL condition on average numerically tracked 0.30 (.34)°C above the CONTROL and 0.21 (.29)°C above the CONTROL-SPRAY. The CONTROL and the CONTROL-SPRAY varied by 0.10 (.44)°C. Based on these data a power calculation estimates that a total of 22, 31 and 305 participants respectively would need to be tested to see statistical differences between conditions; to an alpha level of 0.05 and power of .80. The T_{au} was still rising uncompensably between the 4th and 5th kilometre of the TT. This linear response described only some of the variance in performance data in the TT in the CONTROL ($r^2 = 0.545$), CONTROL-SPRAY ($r^2 = 0.716$) and MENTHOL-SPRAY ($r^2 = 0.553$) conditions; see figure 2A. The mean (SD) rate of rise in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY conditions was 3.25 (0.9) °C.hr^{-1}, 3.70 (0.85) °C.hr^{-1}, 3.53 (1.01) °C.hr^{-1} respectively and was not different (condition effects: $F_{(2,10)} = .909$, $p = .434$).

The T_{skin} response reflected the fact that participants were sprayed showing a tendency to be numerically lower in the CONTROL SPRAY (-0.54 [.15]°C) and MENTHOL SPRAY (-0.46 [.15]°C) compared to the CONTROL condition. The spray conditions were more closely aligned (-0.10 [.20]°C); T_{skin} responses are shown in figure 2B. The T_{au} and T_{skin} data were balanced to the extent that they produced very similar calculated T_b; T_b data not shown. *f*c data were very similar in each condition and not were significantly different (condition effects: $F_{(2,10)} = .856$, $p = .454$). At the end of the TT the *f*c response averaged 191 (6), 191 (9) and 184 (11) b.min^{-1} in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY respectively. The mean (SD) *f*c response in each condition is displayed in figure 3.

INSERT FIGURE 2 NEAR HERE

INSERT FIGURE 3 NEAR HERE

Time Trial Performance

The 1 km split times and total TT completion times did not differ between condition at any stage (split time: $F_{(2,10)}$ = .180, p = .838; completion time: $F_{(2,10)}$ = .192, p = 828) and did not show any interaction effects (split time: $F_{(2.323,11.616)}$ = .712, p = .680; completion time: $F_{(1.422,7.110)}$ = .375, p = .928). A power calculation would suggest that (difference to detect of .18 [1.65], .39 [1.12] and .57 [2.85] minutes) a total of 938, 694, 408 participants would need to be tested to see differences between the CONTROL and CONTROL-SPRAY, CONTROL and MENTHOL-SPRAY and CONTROL-SPRAY and MENTHOL-SPRAY conditions respectively. However, the 1 km split time did show significant time effects **(**$F_{(1.874,9.371)}$ = .11.446, p = .003) which were indicative of an end spurt evidenced by a faster final 1 km split than the preceding 1 km splits; see Figure 4. The CV for completion time across trials across conditions was 5.2 (2.8) %.

INSERT FIGURE 4 NEAR HERE

Spray Volumes, Sweat Production and Fluid Consumed

The volume of spray applied to the body in the CONTROL-SPRAY (102.5 [1.1] mL) and MENTHOL-SPRAY (100.3 [1.0] mL) conditions were similar. Sweat production and the volume of fluid consumed was not different between the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY conditions; sweat production (condition effects: $F_{(2,10)}$ = .959, p =

.416): 1.22 (0.16), 1.16 (0.16), and 1.13 (0.28) L respectively; fluid consumed ($F_{(1.157,5.785)}$ = .249, p = .784): 0.55 (0.18), 0.56 (0.18) and 0.50 (0.21) L respectively.

Discussion

This study examined whether the application of 0.20 % L-Menthol to athletic clothing improves high intensity exercise performance in hot conditions and whether altered thermal perception, after thermal discomfort has been induced, is an initial driver of exercise performance. The study design, utilising a fixed intensity period of exercise in the heat followed by a 5 km TT, successfully induced feelings of thermal discomfort and *warm* to *hot* thermal sensations before L-Menthol was applied (see Figure 1), following which the 5 km TT commenced. At the start and during the earlier parts of the TT thermal perception was significantly improved (TS improved up to 3 km and TC improved up to 1 km) by L-Menthol; H_2 can be accepted. However, evidence of performance enhancement did not arise and pacing template, in conjunction with RPE, remained unchanged; H_3 is therefore rejected. Despite the evident stimulation of cutaneous thermoreceptors indicated by the improved thermal perception, this did not result in higher deep body temperature; H_1 is also therefore rejected.

The study also sought to examine whether the application of 0.20 % L-Menthol altered the thermoregulatory response to exercise in the heat when the work intensity was self-paced, as opposed to fixed as in previous studies (4). The statistical evidence would suggest that deep body temperature, as measured by an aural thermistor, was not significantly different (see figure 2A). Numerically, the data would suggest that the application of L-Menthol to the torso culminated in a tendency towards a raised aural temperature in contrast to the CONTROL-SPRAY (+0.20 [0.10]°C) and CONTROL condition (0.30 [0.10]°C). This small difference was apparent at the start of exercise and did not increase as the trial ensued; these

data are in accordance with Gillis et al. (4). A simple power calculation using the above data shows that participant numbers notably exceeding those of previous studies would be required to see differences in the absolute T_{au} response. Collectively, it appears that the deep body temperature and behavioural response to L-Menthol is less clear when exercise intensity is self-selected. Accordingly, it is prudent, based on statistical evidence, to reject this component of the experimental hypothesis.

Of greater interest is the finding that the rate of rise of aural temperature was not different between conditions. Indeed the slope of the lines describing the uncompensable deep body temperature response to self-paced exercise were similar in each condition which would suggest that high internal temperatures, themselves associated with termination of exercise performance in the heat (9), would not necessarily result at a faster rate if 0.20 % L-Menthol were applied during high-intensity exercise although pre-exercise deep body temperature may prove important. It is evident that higher concentrations of L-Menthol than those employed in the present study can result in thermoregulatory impairment (6) and it would appear unwise to exceed a menthol concentration 0.20 % if normal thermoregulation is to be maintained. Indeed, Kounalakis et al. (6) reported a delay in sweating response following menthol application which was noted to occur to a greater extent in a thermally desensitised (swimmers) group of participants compared to a normal (not cool water exposed) group. Gillis et al. (4) also reported that differences in mean skin and mean body temperature did not arise as a consequence of the application of menthol; our data are also in accordance with these observations. Indeed, it has been shown that thermal preferences are sensed and primarily driven by a mean weighting of skin and deep body temperature rather than deep body temperature alone (15). L-Menthol clearly interferes with this process. Therefore,

maladaptive thermoregulatory behaviours may result if an appropriate dose of L-Menthol is not selected.

The thermal perception data are not consistent with some of the preceding literature. Schlader et al. (7) and Taylor et al. (16) have demonstrated that thermal discomfort is a primary behavioural controller and, in the former case, a driver of exercise performance in the heat. However, Schlader and colleagues (7) induced far greater thermal discomfort than was evident in the present study and utilised an RPE clamp protocol, where perceived effort was set at an RPE vote of 16 throughout their exercise protocol and participants were free to vary their work output to maintain this. Prior to the commencement of exercise, Schlader et al. (7) used topical application of L-Menthol cream (8.0 % concentration) or Capsaicin (0.025%) to the face in order to induce sensations of non-thermal cooling and heating respectively. Although revealing, the clamp protocol is not representative of the way in which persons engage in exercise in the real-world setting. A sub-maximal warm up followed by a competition intensity effort, as in the present study, appear more likely preparatory steps. Moreover, if L-Menthol were to be applied when discomfort were greatest (*i.e.* towards the end of the exercise bout) and used in a competitive setting, an individual may have to balance a possible benefit against the logistical burden of carrying and deploying the menthol whilst exercising. Our data suggest that the perceptual effects of 0.20 % L-Menthol decline after 19 (~3 km split time) and 24 (~ 4 km split time) minutes; this is consistent with the observations of Gillis et al. (4) who suggest a period of 20 minutes of perceptual stimulation. Activities lasting longer than 24 minutes may require Menthol reapplication to induce any beneficial ergogenic effect.

The pacing and performance data represent an important and interesting component to this study with evidence that high internal temperatures towards the end of the 5 km TT exercise bout were overcome to produce an end spurt evidenced by a faster final 1 km of the TT; these data are in accordance with previous studies (5,17,18). This observation adds weight to the argument that it is not the rate of rise of temperature that appears important in dictating exercise performance in the heat but the absolute temperature that is reached towards the end of exercise although this may be dependent on training status (17,18). Our participants reached modest absolute mean aural temperatures in the context of trained participants (~38.5°C) but approached the termination point associated with early fatigue in the untrained (38.7°C; 19) we consider our participants to represent a relatively untrained population. Nevertheless, it seems likely that the participants in the present study did not reach sufficiently high internal temperatures to terminate their exercise bout before task completion.

This study was not without limitation. Indeed, it may appear premature to conclude a null finding for the ergogenic effect of Menthol on performance in the heat with a relatively low sample size. Previous studies have concluded significant changes in thermoregulation have occurred with L-Menthol application, albeit with higher Menthol concentrations, and thermal perception was changed (feeling cooler and more comfortable) but also that pacing and performance was unaltered, in participant cohorts exceeding that of the present study; 12 participants (7; menthol concentration ~ 8 %), 16 participants (6; 4.6 % menthol concentration), and 11 participants respectively (5; 0.05 % menthol concentration). Using the present study data, it seems that L-Menthol induced variable performance rather than consistent change with a power calculation suggesting participant numbers far exceeding those of previous studies would be required to see statistically significant differences. It may

be that some of the irritation responses (which vary between individuals) induced by the application of L-Menthol (4) may underpin this variation, although we did not assess this. Moreover, the training status of our participants may be a contributory factor as evidenced by a CV in participant performance across trials that was roughly double that noted in trained participants completing a similar study (2.3 %5 *cf* 5.2 % in the present study). Collectively, previous literature and consideration of the present data underpin the null finding for the performance effect in this study. Lastly, the use of the inner auditory canal as in index of deep body temperature probably underestimates the actual internal temperature of the participant (20). Indeed, within the temperature range noted here, it is possible that true deep body temperature may be as much as 0.8°C higher if estimated by an alternative means (rectal temperature; 20). However, we suggest that both rectal and aural temperature should only be regarded as only reasonable estimates of pulmonary artery or oesophageal temperature. Although the aural site may underestimate true internal temperature, we contend that it is more suitable than rectal in a dynamic exercise situation such as this and that it enabled the hypothesis with regard to the rate of rise of temperature to be examined appropriately. Even with an addition of 0.8°C to the terminal aural temperature reported in the present study, the participants did not reach a critically high internal temperature (i.e. >40°C; 9).

Conclusion

In summary, there was no clear ergogenic benefit to the application of L-Menthol prior to or on commencing high-intensity exercise in the heat. The perceptual alterations observed in the present study declined over time and were not sufficiently powerful to extend to running performance when thermal discomfort was at its greatest towards the end of exercise in the heat; this may be when the effects of L-Menthol would be most influential. It may be that the

timely application of L-Menthol at this point (*i.e.* when thermal discomfort is greatest) would prove to be ergogenic although this must be balanced against the logistics of this act within the confines of a competitive situation.

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Figure Captions

Figure 1. Mean (SD) TS (Panel A) and TC (Panel B) at the end of the fixed intensity period (15 minutes) and throughout the 5 km TT (n = 6); * indicates difference between CONTROL-SPRAY and CONTROL; ** indicates significant difference between MENTHOL-SPRAY and CONTROL (1^{st} p value) and CONTROL-SPRAY conditions (2^{nd} p value).

Figure 2. Mean (SD) T_{au} response against self-selected pace (Panel A); linear response in the CONTROL (smallest dotted line), CONTROL-SPRAY (medium dotted line) and MENTHOL-SPRAY (large dotted line) and T_{skin} (Panel B) response at the start and at 500 m intervals during the 5 km TT (n = 6).

Figure 3. Mean (SD) fc response during the 5 km TT across 500 m intervals (n = 6).

Figure 4. Mean (SD) 1 km split times within and between conditions; ** indicates significant difference between 4th to 5th km split time compared to all other 1 km split times (n = 6).

Running Head: Performance and L-Menthol

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Spraying with 0.20% L-Menthol does not enhance 5k running performance in the heat in untrained runners

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Abstract

Aim: L-Menthol stimulates cutaneous thermoreceptors and induces cool sensations improving thermal comfort but has also been linked to heat storage responses. Therefore, L-Menthol application could lead to a conflict in behavioural and thermoregulatory drivers improving comfort but leading to a higher rate of deep body temperature rise; the present study examined this possibility. **Method:** Six untrained male participants **(**age 21[1] years; height 1.80 [0.07]m; mass 78.9 [6.9]kg; surface area 1.98 [0.13]m^2) took part. They completed three trials in hot conditions (34°C) where their clothing was sprayed (CONTROL-SPRAY or MENTHOL-SPRAY) or not sprayed (CONTROL) after a fixed intensity exercise period (15-minutes), which induced thermal discomfort, before completing a 5 km treadmill time trial (TT). Thermal perception (thermal sensation and comfort; TS, TC), thermal responses (aural temperature [T_{au}], skin temperature [T_{skin}]), perceived exertion (RPE), heart rate, pacing (1 km split time) and performance (TT completion time) were measured. **Results:** MENTHOL-SPRAY induced improvements in TS (up to 3 km of TT) and TC (up to 1 km) with T_{au} showing a tendency to be higher than CONTROL-SPRAY (+0.20 [0.29]°C) and CONTROL condition (0.30 [0.34]°C); this was not statistically significant and the rate of rise in T_{au} was linear. T_{au} was continuing to rise between the 4th and 5th kilometre of the TT. The other variables were unchanged. TT completion time and pace were not different: CONTROL 27.92 [1.65], CONTROL-SPRAY 28.10 [1.12], MENTHOL-SPRAY 27.53 [2.85] minutes. **Conclusions:** Spraying L-MENTHOL prior to exercise in the heat culminated in improved perception but not altered performance.

Key words: Thermal perception, ergogenic aid, exercise pacing, heat-illness

Introduction

The application of L-Menthol to the skin stimulates cool sensations mediated by specialized sensory neurons (1). These cells feature a highly sensitive receptor, TRPM8, which is activated either by temperatures ranging from 8 to 28 °C, or by chemical compounds such as L-Menthol (2,3). Gillis and colleagues (4) recently demonstrated that the application of L-Menthol to the torso (0.2 % or 0.05 % in 100mL solution) significantly improved thermal sensation (towards feeling cooler) in hot (31 °C), moist (70 %RH) ambient conditions during exercise. Gillis et al. (4) required their participants to exercise at two pre-set intensities (50 and 70% of a pre-trial [separate days] maximal power cycling test) in order to fix metabolic heat production which enabled them to examine whether there were any differences in thermoregulatory effector responses induced by menthol. Their data suggested that the effector response following the application of the 0.05 % L-Menthol solution was not different to that of a control solution (containing surfactants only) despite differences in thermal perception; this raised the novel possibility that application of L-Menthol at 0.05 % concentration could be used to separate thermal perception from thermal state when contrasted to a control spray (5). At the higher concentration examined, Gillis et al. (4) found that 0.20 % L-Menthol altered thermal sensation to a greater extent, but also triggered heat storage responses (possibly vasoconstriction) that culminated in higher rectal temperatures. However, the extent of the higher rectal temperature was never in excess of 0.2°C and mean skin and mean body temperature were unaffected. Similarly, Kounalakis et al. (6) described a higher rate of rectal temperature rise during fixed intensity exercise (60% VO_2max) when 4.6 g per 100 mL of menthol cream (~ 4.6 %) was applied over the entire body surface area. Consequently, participants reached 38°C deep body temperature an average of 7.8 minutes quicker during exercise. The onset of sweating was also delayed with the gain in the response

also affected. However, this study was performed in relatively temperate (24°C; 46 % RH) conditions.

The study design utilised by Gillis et al. (4) and Kounalakis et al. (6) did not attempt to determine whether there was any performance benefit associated with the improvement in thermal perception and this possibility remains. It has been suggested that thermal perception is a conscious and salient driver in changing exercise intensity (7) and in such circumstances alleviation of thermal discomfort may maintain or enhance performance in the heat thereby influencing behavioural thermoregulation (8). Recently we demonstrated that there was no significant alteration in performance or pacing strategy during 40 km time trial cycling exercise in the heat when 0.05 % L-Menthol (in solution) was applied to the torso prior to exercise in trained participants (5); this menthol concentration corresponded to the lowest menthol concentration used by Gillis et al. (4). However, the menthol was applied to the torso at a point when participants were not experiencing thermal discomfort. We speculated that this widened the range of thermal perceptual experiences during exercise in the heat but at a time when perception was not a primary driver of pacing and performance (*i.e* participants were not uncomfortably hot). A study where menthol is applied to the torso when participants were experiencing thermal discomfort may illuminate whether thermal perception is a meaningful initial driver of exercise pacing.

The data of Gillis et al. (4) raise another important consideration. Given that exercise performance in the heat could be limited by stored heat (9) and may be influenced by the feeling of thermal discomfort (8), a concentration of L-Menthol of 0.20 % has the potential to make persons exercising at an uncompensable intensity feel cooler (*i.e.* more comfortable) but store heat at a faster rate, if a higher work intensity is self-selected, leading to a conflict in

thermoregulatory and behavioural drivers. Moreover, commercially available 'performance enhancing' body sprays typically contain, in combination with alcohol, L-Menthol at a concentration of 0.20 % (i.e. Physicool, TM London, U.K) and recommend the application of 100 mL of solution. Such concentrations have yet to be shown, by an independent research study, to enhance exercise performance in the heat. It is apparent that at higher concentrations, L-Menthol is a potent vasoactive substance that can interfere with thermoregulation throughout a range of normothermic and hyperthermic body temperature states (6). Accordingly, the present study aimed to examine whether the application of 0.20 % L-Menthol to athletic clothing improves high intensity exercise performance in hot conditions and whether altered thermal sensation after thermal discomfort has been induced, is a primary driver of exercise behaviour. Based on the findings of Gillis et al (4) it was hypothesised that higher deep body temperatures would be achieved during a performance test after the application of L-Menthol (H_1) whereas the data of Barwood et al. (5) indicate that perception may be improved (H_2) but pacing and performance would remain unchanged (H_3).

Materials and Methods

The protocol was approved by the University of Portsmouth Research Ethics Committee and experiments conformed to the declaration of Helsinki. Participants provided written informed consent. Six recreationally active males (age 21 [1] years; height 1.80 [0.07] m; mass 78.9 [6.9]kg; surface area 1.98 [0.13]m^2; 10) volunteered to participate. An inclusion criteria of a 5 km completion time of 25 minutes or less was set for the study. They abstained from alcohol, caffeine consumption and strenuous exercise 24 hours prior to the test and were non-smokers.

Experimental Design

The study used a within participant, single-blind, repeated-measures design. Participants first completed a familiarisation trial followed by randomised completion of a CONTROL,

CONTROL-SPRAY and MENTHOL-SPRAY treatment condition. All trials took place on separate days at the same time of day (± 1 hour) with a minimum of 48 hours between tests.

Procedure

Following arrival at the laboratory the participant voided, and naked body mass was measured (Ohaus digital weighing scales, I-10, Canada). Prior to dressing, the participant was instrumented with a calibrated, insulated aural thermistor (T_{au}; Grant Instruments Ltd, Cambridge [Shepreth], U.K), skin thermistors at eight different sites (Grant Instruments Ltd, Cambridge [Shepreth], U.K) and a heart rate monitor; mean skin (T_{skin}) and mean body temperature (T_b) were subsequently calculated according to the equations of Olesen (11) using a weighted average of skin temperature at the bicep, chest, foot, hand, hamstring, quadriceps, scapular and shin and Colin et al. (12) respectively.

Participants then dressed and wore the same socks, running shoes and long sleeve t- shirt (the latter provided by the experimenters; 100 % polyester) on each occasion. Thereafter the participant entered an environmental chamber set to a dry bulb temperature of ~34°C and 55 % RH, which was measured by a WBGT weather station (1250 series, Squirrel Data Logger, Grant Instruments Ltd, Cambridge [Shepreth], U.K). They then sat on a chair situated on a calibrated, motorised treadmill (Powerjog GX220, Powerjog, London, UK) for a period of 5-minutes for the collection of pre-exercise resting data. Thereafter the chair was removed and the participant commenced exercise at a fixed intensity for 15-minutes (10 or 12 $km.h^{-1}$; fixed within participant between conditions based on thermal perception [achieving thermal discomfort] and achieving a corresponding sub-maximal heart rate response [this averaged 153 [5] $b.min^{-1}$] in the warm up of the familiarisation trial). At the start of the exercise period a fan positioned 1 metre from the participant (Lloytron 16” fan, Model FO59, Lloytron

Electronics Ltd) and pointed in the direction of the participant's torso, was switched on. The wind speed produced by the fan was verified at a fixed position on the treadmill before and after the experiment by an anemometer (Meterman Anemometer, Model TMA10, Meterman Tests Tools, China; this approximated 1 to 1.5 $m.s^{-1}$). Participants were permitted to consume water *ad libitum* throughout the trial (water temperature ~19 °C). Participants gave perceptual ratings of RPE (13) thermal comfort (TC; 14) and thermal sensation (14) every 5 minutes of the fixed intensity period (TS & TC using a 20 cm visual analogue scale).

At the end of the 15-minute period the treadmill was stopped and the participant rested for 5-minutes during which time the fan was switched off. In two of the experimental conditions (CONTROL-SPRAY and MENTHOL-SPRAY) the participant's t-shirt was sprayed evenly (performed by the same experimenter on each occasion) with 100 mL of solution; the spray volume was measured using calibrated weighing scales situated within the environmental chamber (Mettler PC 400, Mettler Instrumente AG, Greifensee, Zurich, Switzerland) and the spraying procedure took approximately 3-minutes. The clothing was sprayed rather than the skin to minimise losses to the solution dripping from the skin. Towards the end of the 5-minute period the participant stood up and was reminded that they were to exert a maximal effort and cover the 5 km distance as quickly as possible in the subsequent TT; they received feedback only of distance covered. At this point the participant commenced exercise and the fan was switched back on. The time taken to complete each 250 m split of the TT was noted throughout the exercise test and participants provided perceptual votes at each 1 km of the TT. The participant continued to exercise until 5 km was completed, they reached volitional exhaustion or achieved a withdrawal deep body temperature of >39.5°C. At the end of the TT the participant exited the environmental chamber, was de-instrumented and re-weighed naked

behind a privacy screen. Fluid intake and pre and post trial naked body weights (OHAUS I-10 digital scales, Canada) were used to estimate sweat production.

Description of Sprays

Sprays were produced by an independent chemical consultant (Chemical Associates, Rosemead, Frodsham, United Kingdom). The CONTROL SPRAY contained 3% surfactants mixed in water, while the MENTHOL SPRAY contained a concentration of 0.20 wt/wt L-Menthol in 3% surfactants plus water. Sprays were stored at room temperature (20°C) and transferred into the chamber ~3 h before testing. In order to minimise supplementary perceptual cooling associated with a spray temperature lower than exercising skin temperature and ambient temperature, the bottles containing the sprays were immersed in a temperature controlled water bath held at 34°C within the chamber (Tempette Junior TE 8J, Techne, Cambridge, U.K), 1-hour before the trial commenced.

Data Analyses

Mean (SD) were calculated for each condition for the final data point of the rest period (*i.e.* 5-minutes after the end of the fixed intensity period) and on completion of every kilometre of the TT for all variables (T_{au}, T_{au} rate of rise, T_{skin}, T_b, cardiac frequency derived from heart rate [*f*c] RPE, TS, TC) with the exception of RPE [not examined at rest] and performance data [no data point at rest].

Comparisons were made between each condition and across time using factorial ANOVA with repeated measures. Assumptions of sphericity were checked using Mauchley's test and adjusted where necessary (Greenhouse-Geisser). Statistically significant effects were determined *post-hoc* using pairwise comparisons. Comparisons were also made between

conditions for fluid consumption and sweat production using a one-way ANOVA. The coefficient of variation (CV) within participant across trials was also calculated. The stability of ambient conditions were examined using repeated measures analysis of variance (ANOVA). The alpha level for all statistical tests was set at 0.05. ANOVAs were calculated using PASW version 18 (SPSS Inc, Chicago, Illinois). Given the low sample size, and where appropriate, statistical power to interrogate null findings were performed to alpha level of 0.05 power of .80 using mean (SD) differences between conditions; this was conducted using Minitab version 15 (Minitab Inc. USA). CV was calculated using Microsoft Excel.

Results

Environmental Conditions

There were no significant differences in the environmental conditions between the trials. Mean (SD) across conditions for the dry bulb, wet bulb and calculated WBGT were 33.9 (.10)°C; $F_{(2,10)} = 1.715$, $p = .229$; 26.2 (.30)°C; $F_{(2,10)} = 2.217$, $p = .160$) and 28.5 (.20)°C; $F_{(2,10)} = 1.616$, $p = .246$) respectively.

Perceptual Responses

At the end of the fixed intensity exercise period (*i.e.* immediately before participants were sprayed) the TS and TC averaged 15.5 (1.4) cm and 9.8 (3.0) cm across conditions which corresponded to the descriptors *warm to hot* and *just uncomfortable*; there were no differences between conditions at this stage (TS: CONTROL *cf* CONTROL-SPRAY $p = .931$ and MENTHOL, $p = .909$, MENTHOL *cf* CONTROL-SPRAY $p = .823$; TC: CONTROL *cf* CONTROL-SPRAY $p = .123$ and MENTHOL, $p = .529$, MENTHOL *cf* CONTROL-SPRAY $p = .744$). Following spraying and during the TT the TS showed significant differences between conditions (main effect for condition: $F_{(1.632,\ 8.160)} = 15.953$, $p = .002$) and an

interaction effect between condition and time ($F_{(2.281, 11.407)}$ = 7.979, p = .006). The TS votes were significantly lower (*i.e.* towards feeling cooler) in the MENTHOL condition compared to the CONTROL (p = .007) and the CONTROL-SPRAY (p = .002). As the TT ensued, and differences in the MENTHOL-SPRAY condition was sustained up to 3 km but were no longer different at the 4 km and 5 km distance point; significant differences between conditions are summarised in Figure 1A. All participants completed the TT stage.

INSERT FIGURE 1 NEAR HERE

TC neared being different between conditions (condition effects: $F_{(1.380, 6.900)}$ = 3.899, p = .083) but did show an interaction effect between condition and time ($F_{(3.444, 17.222)}$ = 3.063, p = .050). The interaction effects were similar, but not as strong, as the TS responses with improved comfort evident in the first 1 km of the TT in the MENTHOL-SPRAY condition relative to other two conditions; significant differences between conditions are summarised in Figure 1B.

RPE data did not differ significantly between condition (condition effects: $F_{(2,10)}$ = .045, p = .956) or show any interaction effect ($F_{(3.505, 17.527)}$ = 3.505, p = .672). At the equidistant time points the RPE vote was always within 1 point between conditions on the rating scale and culminated in a vote of 19 (1) across conditions at the end of the TT. Towards the start of the TT (*i.e.* after 1 km), when the menthol spray was evidently active the RPE vote was 12 (2), 13 (2) and 13 (1) in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY conditions respectively.

Thermal and fc Responses

The MENTHOL and CONTROL-SPRAYS did not significantly influence T_{au}, T_{skin} and T_b at the start or during the TT at any stage (condition effects: T_{au}: $F_{(2,10)} = 2.393$, $p = .142$; T_{skin}; $F_{(1.042,5.210)} = 1.839$, $p = .209$; T_b: $F_{(2,10)} = .401$, $p = .680$). The T_{au} response in the MENTHOL condition on average numerically tracked 0.30 (.34)°C above the CONTROL and 0.21 (.29)°C above the CONTROL-SPRAY. The CONTROL and the CONTROL-SPRAY varied by 0.10 (.44)°C. Based on these data a power calculation estimates that a total of 22, 31 and 305 participants respectively would need to be tested to see statistical differences between conditions; to an alpha level of 0.05 and power of .80. The T_{au} was still rising uncompensably between the 4th and 5th kilometre of the TT. This linear response described only some of the variance in performance data in the TT in the CONTROL ($r^2 = 0.545$), CONTROL-SPRAY ($r^2 = 0.716$) and MENTHOL-SPRAY ($r^2 = 0.553$) conditions; see figure 2A. The mean (SD) rate of rise in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY conditions was 3.25 (0.9) °C.hr^{-1}, 3.70 (0.85) °C.hr^{-1}, 3.53 (1.01) °C.hr^{-1} respectively and was not different (condition effects: $F_{(2,10)} = .909$, $p = .434$).

The T_{skin} response reflected the fact that participants were sprayed showing a tendency to be numerically lower in the CONTROL SPRAY (-0.54 [.15]°C) and MENTHOL SPRAY (-0.46 [.15]°C) compared to the CONTROL condition. The spray conditions were more closely aligned (-0.10 [.20]°C); T_{skin} responses are shown in figure 2B. The T_{au} and T_{skin} data were balanced to the extent that they produced very similar calculated T_b; T_b data not shown. *f*c data were very similar in each condition and not were significantly different (condition effects: $F_{(2,10)} = .856$, $p = .454$). At the end of the TT the *f*c response averaged 191 (6), 191 (9) and 184 (11) b.min^{-1} in the CONTROL, CONTROL-SPRAY and MENTHOL-SPRAY respectively. The mean (SD) *f*c response in each condition is displayed in figure 3.

INSERT FIGURE 2 NEAR HERE

INSERT FIGURE 3 NEAR HERE

Time Trial Performance

The 1 km split times and total TT completion times did not differ between condition at any stage (split time: $F_{(2,10)}$ = .180, p = .838; completion time: $F_{(2,10)}$ = .192, p = 828) and did not show any interaction effects (split time: $F_{(2.323,11.616)}$ = .712, p = .680; completion time: $F_{(1.422,7.110)}$ = .375, p = .928). A power calculation would suggest that (difference to detect of .18 [1.65], .39 [1.12] and .57 [2.85] minutes) a total of 938, 694, 408 participants would need to be tested to see differences between the CONTROL and CONTROL-SPRAY, CONTROL and MENTHOL-SPRAY and CONTROL-SPRAY and MENTHOL-SPRAY conditions respectively. However, the 1 km split time did show significant time effects **(**$F_{(1.874,9.371)}$ = .11.446, p = .003) which were indicative of an end spurt evidenced by a faster final 1 km split than the preceding 1 km splits; see Figure 4. The CV for completion time across trials across conditions was 5.2 (2.8) %.

INSERT FIGURE 4 NEAR HERE

Spray Volumes, Sweat Production and Fluid Consumed

The volume of spray applied to the body in the CONTROL-SPRAY (102.5 [1.1] mL) and MENTHOL-SPRAY (100.3 [1.0] mL) conditions were similar. Sweat production and the volume of fluid consumed was not different between the CONTROL, CONTROL-SPRAY

and MENTHOL-SPRAY conditions; sweat production (condition effects: $F_{(2,10)}$ = .959, p = .416): 1.22 (0.16), 1.16 (0.16), and 1.13 (0.28) L respectively; fluid consumed ($F_{(1.157,5.785)}$ = .249, p = .784): 0.55 (0.18), 0.56 (0.18) and 0.50 (0.21) L respectively.

Discussion

This study examined whether the application of 0.20 % L-Menthol to athletic clothing improves high intensity exercise performance in hot conditions and whether altered thermal perception, after thermal discomfort has been induced, is an initial driver of exercise performance. The study design, utilising a fixed intensity period of exercise in the heat followed by a 5 km TT, successfully induced feelings of thermal discomfort and *warm* to *hot* thermal sensations before L-Menthol was applied (see Figure 1), following which the 5 km TT commenced. At the start and during the earlier parts of the TT thermal perception was significantly improved (TS improved up to 3 km and TC improved up to 1 km) by L-Menthol; H_2 can be accepted. However, evidence of performance enhancement did not arise and pacing template, in conjunction with RPE, remained unchanged; H_3 is therefore rejected. Despite the evident stimulation of cutaneous thermoreceptors indicated by the improved thermal perception, this did not result in higher deep body temperature; H_1 is also therefore rejected.

The study also sought to examine whether the application of 0.20 % L-Menthol altered the thermoregulatory response to exercise in the heat when the work intensity was self-paced, as opposed to fixed as in previous studies (4). The statistical evidence would suggest that deep body temperature, as measured by an aural thermistor, was not significantly different (see figure 2A). Numerically, the data would suggest that the application of L-Menthol to the torso culminated in a tendency towards a raised aural temperature in contrast to the

CONTROL-SPRAY (+0.20 [0.10]°C) and CONTROL condition (0.30 [0.10]°C). This small difference was apparent at the start of exercise and did not increase as the trial ensued; these data are in accordance with Gillis et al. (4). A simple power calculation using the above data shows that participant numbers notably exceeding those of previous studies would be required to see differences in the absolute T_{au} response. Collectively, it appears that the deep body temperature and behavioural response to L-Menthol is less clear when exercise intensity is self-selected. Accordingly, it is prudent, based on statistical evidence, to reject this component of the experimental hypothesis.

Of greater interest is the finding that the rate of rise of aural temperature was not different between conditions. Indeed the slope of the lines describing the uncompensable deep body temperature response to self-paced exercise were similar in each condition which would suggest that high internal temperatures, themselves associated with termination of exercise performance in the heat (9), would not necessarily result at a faster rate if 0.20 % L-Menthol were applied during high-intensity exercise although pre-exercise deep body temperature may prove important. It is evident that higher concentrations of L-Menthol than those employed in the present study can result in thermoregulatory impairment (6) and it would appear unwise to exceed a menthol concentration 0.20 % if normal thermoregulation is to be maintained. Indeed, Kounalakis et al. (6) reported a delay in sweating response following menthol application which was noted to occur to a greater extent in a thermally desensitised (swimmers) group of participants compared to a normal (not cool water exposed) group. Gillis et al. (4) also reported that differences in mean skin and mean body temperature did not arise as a consequence of the application of menthol; our data are also in accordance with these observations. Indeed, it has been shown that thermal preferences are sensed and primarily driven by a mean weighting of skin and deep body temperature rather than deep

body temperature alone (15). L-Menthol clearly interferes with this process. Therefore, maladaptive thermoregulatory behaviours may result if an appropriate dose of L-Menthol is not selected.

The thermal perception data are not consistent with some of the preceding literature. Schlader et al. (7) and Taylor et al. (16) have demonstrated that thermal discomfort is a primary behavioural controller and, in the former case, a driver of exercise performance in the heat. However, Schlader and colleagues (7) induced far greater thermal discomfort than was evident in the present study and utilised an RPE clamp protocol, where perceived effort was set at an RPE vote of 16 throughout their exercise protocol and participants were free to vary their work output to maintain this. Prior to the commencement of exercise, Schlader et al. (7) used topical application of L-Menthol cream (8.0 % concentration) or Capsaicin (0.025%) to the face in order to induce sensations of non-thermal cooling and heating respectively. Although revealing, the clamp protocol is not representative of the way in which persons engage in exercise in the real-world setting. A sub-maximal warm up followed by a competition intensity effort, as in the present study, appear more likely preparatory steps. Moreover, if L-Menthol were to be applied when discomfort were greatest (*i.e.* towards the end of the exercise bout) and used in a competitive setting, an individual may have to balance a possible benefit against the logistical burden of carrying and deploying the menthol whilst exercising. Our data suggest that the perceptual effects of 0.20 % L-Menthol decline after 19 (~3 km split time) and 24 (~ 4 km split time) minutes; this is consistent with the observations of Gillis et al. (4) who suggest a period of 20 minutes of perceptual stimulation. Activities lasting longer than 24 minutes may require Menthol reapplication to induce any beneficial ergogenic effect.

The pacing and performance data represent an important and interesting component to this study with evidence that high internal temperatures towards the end of the 5 km TT exercise bout were overcome to produce an end spurt evidenced by a faster final 1 km of the TT; these data are in accordance with previous studies (5,17,18). This observation adds weight to the argument that it is not the rate of rise of temperature that appears important in dictating exercise performance in the heat but the absolute temperature that is reached towards the end of exercise although this may be dependent on training status (17,18). Our participants reached modest absolute mean aural temperatures in the context of trained participants (~38.5°C) but approached the termination point associated with early fatigue in the untrained (38.7°C; 19) we consider our participants to represent a relatively untrained population. Nevertheless, it seems likely that the participants in the present study did not reach sufficiently high internal temperatures to terminate their exercise bout before task completion.

This study was not without limitation. Indeed, it may appear premature to conclude a null finding for the ergogenic effect of Menthol on performance in the heat with a relatively low sample size. Previous studies have concluded significant changes in thermoregulation have occurred with L-Menthol application, albeit with higher Menthol concentrations, and thermal perception was changed (feeling cooler and more comfortable) but also that pacing and performance was unaltered, in participant cohorts exceeding that of the present study; 12 participants (7; menthol concentration ~ 8 %), 16 participants (6; 4.6 % menthol concentration), and 11 participants respectively (5; 0.05 % menthol concentration). Using the present study data, it seems that L-Menthol induced variable performance rather than consistent change with a power calculation suggesting participant numbers far exceeding those of previous studies would be required to see statistically significant differences. It may

be that some of the irritation responses (which vary between individuals) induced by the application of L-Menthol (4) may underpin this variation, although we did not assess this. Moreover, the training status of our participants may be a contributory factor as evidenced by a CV in participant performance across trials that was roughly double that noted in trained participants completing a similar study (2.3 %5 *cf* 5.2 % in the present study). Collectively, previous literature and consideration of the present data underpin the null finding for the performance effect in this study. Lastly, the use of the inner auditory canal as in index of deep body temperature probably underestimates the actual internal temperature of the participant (20). Indeed, within the temperature range noted here, it is possible that true deep body temperature may be as much as 0.8°C higher if estimated by an alternative means (rectal temperature; 20). However, we suggest that both rectal and aural temperature should only be regarded as only reasonable estimates of pulmonary artery or oesophageal temperature. Although the aural site may underestimate true internal temperature, we contend that it is more suitable than rectal in a dynamic exercise situation such as this and that it enabled the hypothesis with regard to the rate of rise of temperature to be examined appropriately. Even with an addition of 0.8°C to the terminal aural temperature reported in the present study, the participants did not reach a critically high internal temperature (i.e. >40°C; 9).

Conclusion

In summary, there was no clear ergogenic benefit to the application of L-Menthol prior to or on commencing high-intensity exercise in the heat. The perceptual alterations observed in the present study declined over time and were not sufficiently powerful to extend to running performance when thermal discomfort was at its greatest towards the end of exercise in the heat; this may be when the effects of L-Menthol would be most influential. It may be that the

timely application of L-Menthol at this point (*i.e.* when thermal discomfort is greatest) would prove to be ergogenic although this must be balanced against the logistics of this act within the confines of a competitive situation.

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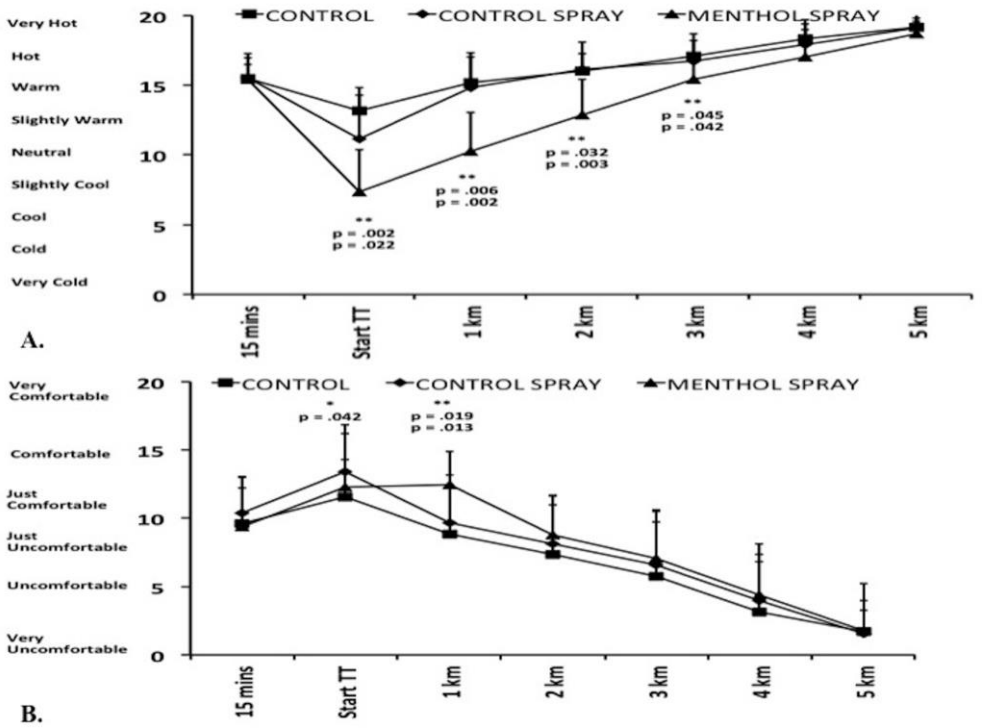
Figure Captions

Figure 1. Mean (SD) TS (Panel A) and TC (Panel B) at the end of the fixed intensity period (15 minutes) and throughout the 5 km TT (n = 6); * indicates difference between CONTROL-SPRAY and CONTROL; ** indicates significant difference between MENTHOL-SPRAY and CONTROL (1^{st} p value) and CONTROL-SPRAY conditions (2^{nd} p value).

Figure 2. Mean (SD) T_{au} response against self-selected pace (Panel A); linear response in the CONTROL (smallest dotted line), CONTROL-SPRAY (medium dotted line) and MENTHOL-SPRAY (large dotted line) and T_{skin} (Panel B) response at the start and at 500 m intervals during the 5 km TT (n = 6).

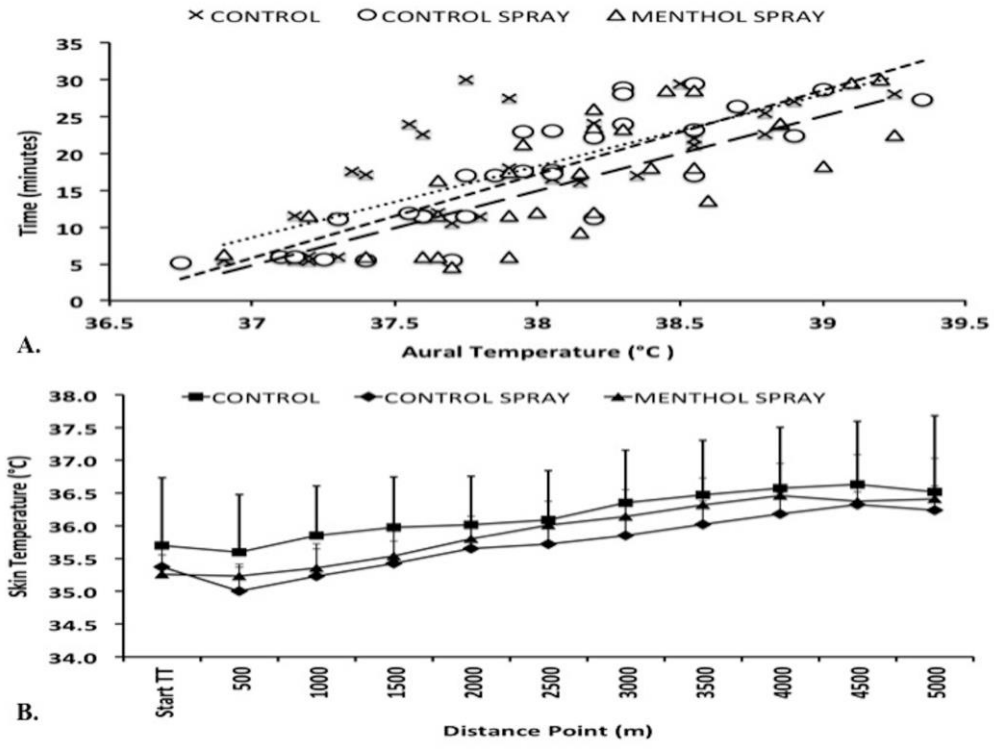
Figure 3. Mean (SD) fc response during the 5 km TT across 500 m intervals (n = 6).

Figure 4. Mean (SD) 1 km split times within and between conditions; ** indicates significant difference between 4th to 5th km split time compared to all other 1 km split times (n = 6).


CONTROL
CONTROL SPRAY
MENTHOL SPRAY
Very Hot
Hot
Warm
Slightly Warm
Neutral
Slightly Cool
Cool
Cold
Very Cold
20
15
10
5
0
** p = .002 p = .022
** p = .006 p = .002
** p = .032 p = .003
** p = .045 p = .042
15 mins
Start TT
1 km
2 km
3 km
4 km
5 km
A.
Very Comfortable
Comfortable
Just Comfortable
Just Uncomfortable
Uncomfortable
Very Uncomfortable
* p = .042
** p = .019 p = .013
B.

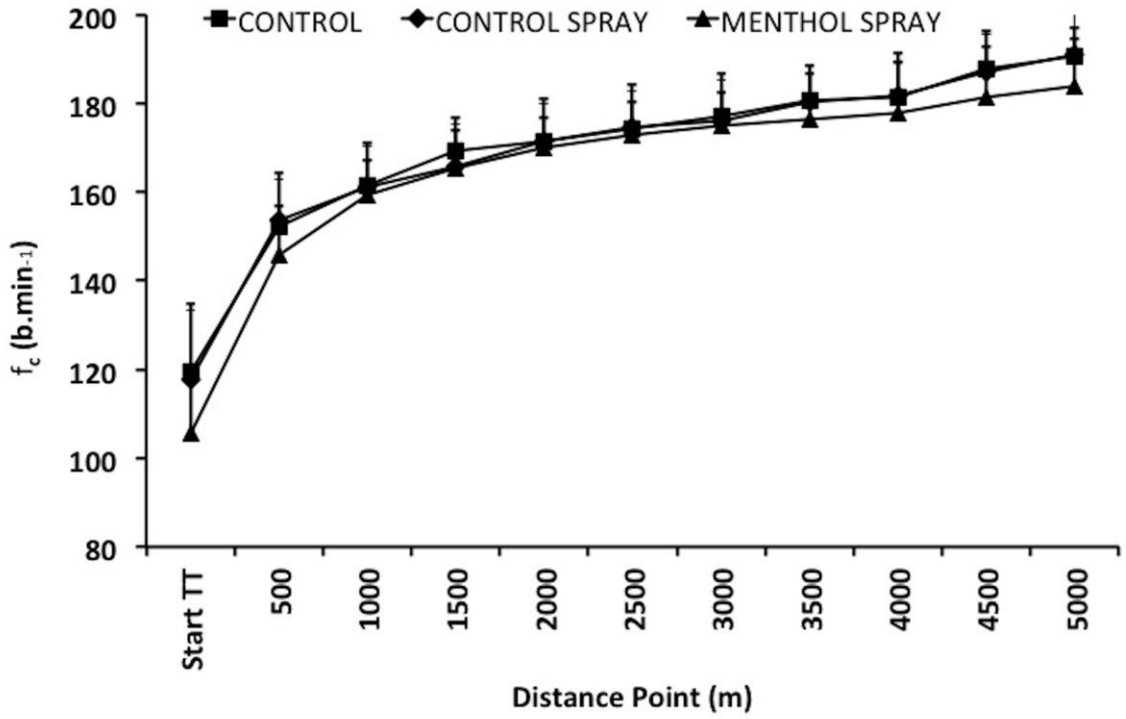
1

2

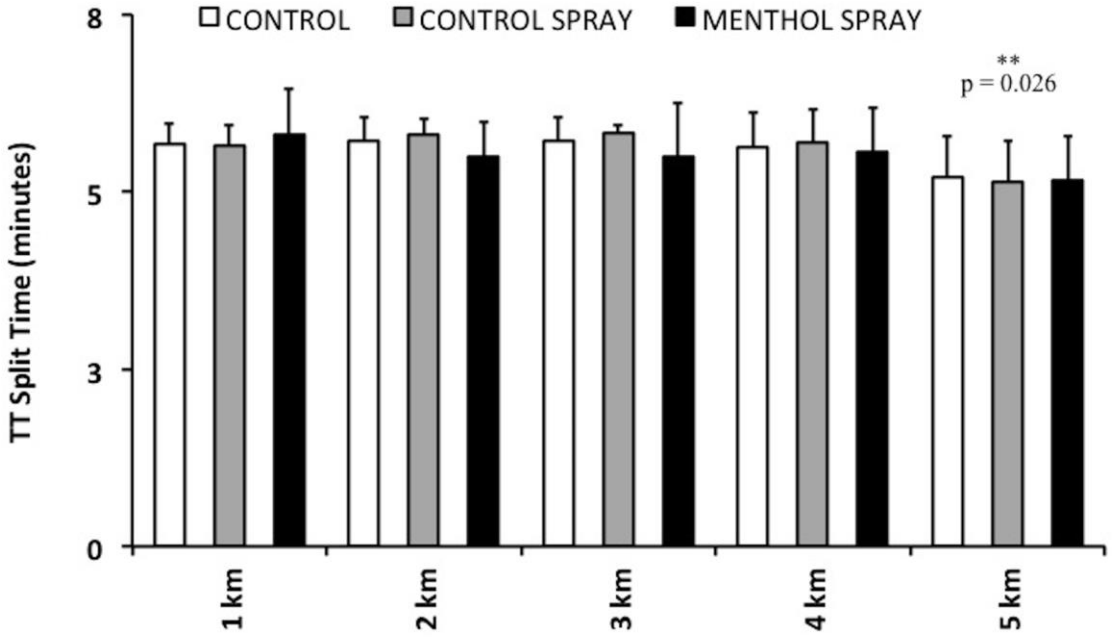

CONTROL
CONTROL SPRAY
MENTHOL SPRAY
Time (minutes)
35
30
25
20
15
10
5
0
36.5
37
37.5
38
38.5
39
39.5
Aural Temperature (°C)
A.
Skin Temperature (°C)
38.0
37.5
37.0
36.5
36.0
35.5
35.0
34.5
34.0
Start TT
500
1000
1500
2000
2500
3000
3500
4000
4500
5000
Distance Point (m)
B.

3

4


CONTROL
CONTROL SPRAY
MENTHOL SPRAY
200
180
160
140
120
100
80
f_c (b.min^{-1})
Start TT
500
1000
1500
2000
2500
3000
3500
4000
4500
5000
Distance Point (m)

1


CONTROL
CONTROL SPRAY
MENTHOL SPRAY
**
p = 0.026
8
5
3
0
TT Split Time (minutes)
1 km
2 km
3 km
4 km
5 km

2