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Repeated Menthol spray application enhances exercise capacity in the heat

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Abstract

Purpose. Exercise performance is impaired in the heat and a contributing factor to this decrement is thermal discomfort. Menthol-spraying of skin is one means of alleviating thermal discomfort but has yet to be shown to be ergogenic using single spray applications. We examined whether repeated menthol-spraying could relieve thermal discomfort, reduce perception of exertion and improve exercise performance in hot (35ºC), dry (22% RH) conditions; we hypothesised it would. **Method.** Eight trained cyclists completed two separate conditions of fixed intensity (FI) cycling (50% P_{Max}) for 45-minutes before a test to exhaustion (TTE; 70% P_{Max}) with 100 mL of menthol-spray (0.20% menthol) or control-spray applied to the torso after 20 and 40-minutes. Perceptual (thermal sensation (TS), thermal comfort (TC), RPE) performance (TTE duration), thermal variables (skin temperature (T_{skin}), rectal temperature (T_{rec}), cardiac frequency (*f*c)) and sweating were measured. Data were compared using ANOVA to 0.05 alpha level. **Results.** Menthol-spray improved TS ('*cold*' sensation *cf* '*warm/hot*' after first spraying; p=.008) but only descriptively altered TC ('*comfortable*' *cf* '*uncomfortable*'; p=.173). Sweat production (994 (380) mL *cf* 1180 (380); p=.020) mL and rate (827 (327)$mL\cdot hr^{-1}$ *cf* 941 (319)$mL\cdot hr^{-1}$; p=.048) lowered. TTE performance improved (4.6 (1.74) *cf* 2.4 (1.55) minutes (p=.004). Menthol-spray effects diminished despite repeated applications indicating increased contribution of visceral thermoreceptors to thermal perception. **Conclusion.** Repeated menthol-spray improves exercise capacity but alters thermoregulation potentially conflicting behavioural and thermoregulatory drivers; care should be taken with its use. Carrying and deploying menthol-spray would impose a logistical burden which needs consideration against performance benefit.

Keywords. TRPM8 receptors, thermoregulation, sweating, thermal perception.

Introduction

Exercise performance is impaired in hot conditions with fatigue occurring prematurely compared to cool environments[1]. The aetiology of this fatigue is complex and multifaceted but is in part attributable to increased thermal sensations (i.e. feeling hot) and thermal discomfort[2]. Accordingly, any intervention that offsets these disturbances in thermal perception may prove to be ergogenic and influence exercise behaviour[3]. One such intervention with the potential to do so is the topical application of menthol to the skin. This has been found to change the action potential of the Transient Receptor Potential Melastatin 8 (TRPM8) subfamily of thermoreceptors thereby inducing cool sensations[4,5]. Although, menthol is also known to activate TRP vanilloid (TRPV) and ankyrin (TRPA) receptors[6] above temperatures of 37 °C thereby inducing warm sensations[7]. Accordingly in exercise and environmental scenarios where skin temperatures do not exceed 37 °C (i.e. the majority of scenarios) the chemical stimulation of the skin by menthol appears to be a viable means of improving thermal perception and potentially exercise performance.

Only one study to date has revealed an ergogenic benefit following the topical application of an 8% menthol gel applied to the face during self-paced exercise performed at a fixed perception of exertion[2]. Menthol application induced an approximate 18% increase in total work during the study where thermal stress was applied through a water-perfused suit[2]. Thermal perception was shown to be a relatively independent behavioural regulatory influence on exercise termination as shorter exercise duration was observed with the induction of hot sensations by capsaicin application to the skin[2]. However, in studies performed using ecologically valid laboratory protocols[3,8,9] an ergogenic effect has proved illusive leading to suggestions that menthol-spraying may only improve thermal perception but not performance[10]. Menthol applied to the skin at concentrations (0.05 to 0.20 % L-Menthol in solution), similar to that of commercially available products (Physicool™, London, U.K), has been reliably shown to induce improvements in thermal sensation and comfort, during fixed intensity[11] and self-paced exercise[3,8,9] in the heat. However, it has also been shown to induce heat gain responses (i.e. vasoconstriction[11]) and alter sweating responses[12]; in the latter case at higher concentrations (i.e. 4.6%[12]). Therefore, it is also plausible that menthol application could increase the risk of heat-illness and place behavioural and thermoregulatory drivers in conflict.

Nevertheless, there are iterations on the timing of menthol application that have not been explored experimentally which may mean concluding a lack of ergogenic effect is premature. To date, we have explored whether relieving thermal discomfort and improving thermal sensation is performance enhancing prior to and during the early minutes of a 40 km cycling time trial; it was not[8]. We have examined whether inducing hot and uncomfortable sensations using a heat pre-load followed by menthol application would result in improved performance of a shorter duration exercise of 5 km running but it did not[9]. Most recently we examined whether applying menthol towards the end of an exercise task (i.e. at 10 km of a 16.1 km cycling time trial; TT) would result in benefits to TT completion time[3]. Once again we saw no improvement although menthol-spray application did result in lowered RPE in addition to benefits to thermal perception. Each of these studies, and others where perceptual manipulation was the primary goal[11] involved *single* applications of menthol-spray. It has yet to be investigated whether *repeated* menthol application can act as an ergogenic aid.

Theoretically, in prior studies the acute bouts of thermal discomfort relief through menthol-spray application may have been insufficient to perturb the behavioural thermoregulatory drivers towards altering exercise performance. Whereas *repeated* application may provide a greater driver to change this. Moreover, the nature of the exercise task may also be important. Menthol is evidently more likely to influence an exercise task where tolerance is the critical factor[10] (e.g. test to exhaustion; TTE) rather than the spontaneous variation in power output (e.g. TT) which have consistently failed to be responsive to menthol in three of our previous studies[3,8,9]. Accordingly, the present study sought to examine this possibility.

We hypothesised that menthol application, applied every 20-minutes during exercise in the heat[11], would enhance exercise performance in a subsequent TTE where heat tolerance is the main limiting factor to performance (H_1). We also hypothesised that menthol-spray application would enhance thermal perception by inducing cool thermal sensations and relieving thermal discomfort which may result in reduced perception of exertion in contrast to a control-spray condition (H_2).

Method

Experimental Design

The local ethics committee approved the study which used a within participant, repeated measures design in which participants completed three exercise conditions. The first condition took place in a temperate environment and was to establish their maximal power output (P_{Max}) for use during the subsequent two conditions which took place in a hot environment. Conditions two and three were counter-balanced where the participants' t-shirt was repeatedly sprayed (i.e. every 20-minutes) with a menthol-spray or a control-spray. Tests took place at the same time of day (± 1 hour) with a minimum of 48 hours between tests.

Participants

Eight trained cyclists (mean ± SD: age 22 ± 2 yrs; height 1.84 ± 0.1 m; body surface area[13] 2.05 ± 0.1 m^2 P_{Max} 362.5 ± 35.4 W) volunteered and provided written informed consent. Participants were considered trained if they achieved a minimum P_{Max} of $\geq$350 watts[14]. Participants abstained from alcohol, caffeine consumption and strenuous exercise 24 hours prior to each test and were non-smokers.

Procedures

Condition One - P_{Max} Test: Participants arrived at the laboratory wearing cycle clothing. They were instrumented with a heart rate monitor (FT1, Polar Electro Oy, Kempele, Finland) and entered the environmental chamber held at a temperature of 16°C. Participants were made comfortable on the cycle ergometer (Velotron, Racermate, Seattle, USA); bike positioning was replicated for subsequent visits. Participants completed a standardised 5-minute warm up at 150 W and a cadence of 70 rev·min^{-1} followed by stretching. They remounted the ergometer and recommenced cycling at the same power output and cadence as the warm-up. The required power output was increased by 50 W every 2-minutes until volitional exhaustion or when the prescribed cadence could not be maintained for 15-seconds and having achieved a heart rate within 10 b.p.m^{-1} of age predicted maximum. Participants were instructed prior that they should make a maximal effort during the test.

Conditions Two and Three - Repeated Spray Applications: Participants arrived in a hydrated state; i.e. having consumed 500 mL of water the preceding night and 500 mL in the two hours prior to arrival at the laboratory. Participants were allowed to drink tepid tap water during the trials. Participants first voided and naked body mass was measured in private (Seca, Model 705 2321009, Vogel & Halke, Hamburg, Germany). They then donned their cycling shorts and were instrumented with a calibrated, insulated rectal thermistor (Grant Instruments Ltd, Cambridge [Shepreth], U.K) inserted (in private) 12-15 cm beyond the anal sphincter. They were also instrumented with skin thermistors (Grant Instruments Ltd, Cambridge [Shepreth], U.K) placed at eight different body sites[15] on the left side of the body secured by breathable tape (TransporeTM,1527-1, 3M Health Care, MN, USA). A heart rate monitor was also worn to measure cardiac frequency (*f*c). Rectal temperature (T_{rec}) and skin temperature (T_{skin}) were logged automatically every 5-seconds using a remote data logger (Squirrel 2020 series, Grant Instruments Ltd, Cambridge [Shepreth], U.K). Following instrumentation participants completed dressing by wearing socks, shoes and a close-fitting long sleeve t-shirt (100% polyester; Campri Sports Baselayer, Sportsdirect, Shirebrook, U.K). Identical clothing was worn in each condition that involved repeated spraying.

Participants then entered an environmental chamber set to 35°C and 20% relative humidity (RH). Environmental conditions were measured by a wet-bulb, globe, temperature (WBGT) station (1000 series, Squirrel Data Logger, Grant Instruments Ltd, Cambridge [Shepreth], U.K). One minute prior to the start of exercise, all data logging systems were activated and synchronised. Prior to the commencement in exercise participants provided a resting capillary sample of blood for measurement of blood lactate concentration (B_{lac}). Participants also reported their resting thermal comfort (TC[16]) and thermal sensation (TS[16]). Participants then mounted the cycle ergometer and completed the same standardised warm up as prior to the P_{Max} , and then commenced fixed intensity (FI) cycling at 50% P_{Max} for 45-minutes. Participants cycled in front of a fan positioned 80 cm from the velotron (Wahl, Model ZX220, Wahl, Sterling, IL, USA) and pointed at the participants' torso. The wind speed produced by the fan was verified at a fixed position by an anemometer (LM-8000 Anemometer, Digital Instruments, New York, USA; this approximated between 1.6 and 2.1 $m \cdot s^{-1}$).

Perceptual responses including RPE[17], TC and TS were obtained initially every 10-minutes of the FI period, until (i.e before) the first spray application at 20-minutes. They were recorded every 5-minutes thereafter; RPE was not collected at 30-minutes. After 20 and 40-minutes of exercise participants' jerseys were sprayed evenly with 100 mL of either the control-spray or the menthol-spray which was heated in a water bath to match environmental temperature[3]. Spray volume was measured on each occasion using calibrated, digital, weighing scales (Sartorius Mechatronics UK Ltd, TE6100, Surrey, U.K; 1 g resolution). Intervals between sprays were 20-minutes on the basis that the menthol-spray perceptual response has been shown to decay thereafter[11]. 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.

Upon completion of the FI period participants provided another capillary blood sample and immediately commenced a test to exhaustion (TTE) at 70 % P_{Max}. Participants received no feedback of exercise time elapsed or encouragement during the TTE. Upon TTE cessation (i.e. volitional exhaustion) the participant exited the chamber and were weighed naked and, in conjunction with measured fluid intake, sweat production and sweat rate were calculated. Performance times were not revealed until the post-experiment debrief.

Statistical Analysis

Mean (SD) were calculated for perceptual (TS, TC, RPE), performance, (B_{lac}, TTE duration), thermal (T_{skin}, T_{rec} and *f*c) spray variables (temperature and volume), environmental conditions and sweat production including rate. The normality of distribution was verified using a using Kolmogorov-Smirnov test. Data were compared using a repeated measures analysis of variance (ANOVA) at rest and fixed points during the FI period including TTE end point for the two hot trials (9 x 2 ANOVA) for perceptual (no RPE measure at rest and 30-minute point) and thermal variables. Sphericity was checked using Mauchley's test and, where necessary, a Greenhouse-Geisser adjustment was applied. The direction of statistically significant effects were determined using *Fisher's* (LSD) *post-hoc* pair-wise comparisons. Partial eta squared (ηp^2) are reported as estimates of effect size. Environmental conditions, spray temperature, volume, TTE duration, fluid consumed, sweat data and terminal B_{lac} were compared using paired samples t-test. The 95% confidence interval (CI) was calculated for the TTE data. Data are otherwise presented as mean (SD). An alpha level of 0.05 was used for all statistical tests which were conducted using SPSS (SPSS *v* 21, IBM, Chicago, Illinois, USA) and Prism (Graphpad, Prism *v* 6, San Diego, USA).

Results

Environmental Conditions

Ambient temperature averaged 35.0 (1.3) ºC and 34.6 (1.2) ºC in the control-spray and menthol-spray conditions respectively and did not differ (t = .846, p = .213). RH averaged 21.8 (0.90) % and 22.2 (1.0) % and did not differ (t = -1.06, p = .162).

Spray Volume and Temperature

Volume of spray applied was 200 (3) mL in the control-spray and 200 (2) mL in the menthol-spray conditions which were similar (t = 0.110, p = 0.460). The temperature of the control-spray averaged 37.4 (1.2) ºC and was 38.3 (1.6) ºC in the menthol-spray condition and were not different (t = 1.766, p = .097).

TTE Performance

TTE was 2.4 (1.55) minutes and 4.6 (1.74) minutes in the control-spray and menthol-spray conditions respectively and was significantly greater after menthol-spraying application (t = -3.63, p = 0.004; 95% CI 0.53 to 3.82 minutes).

Perceptual Responses

Participants' TS responses were similar in each condition before the first spray (i.e. at 20 minutes) and corresponded to the worded descriptor '*hot*'. At 25-minutes, 5-minutes after spraying, TS was significantly lower (main effect for condition: $F_{(1, 7)} = 13.139$, $p = 0.008$, $\eta p^2 = .652$ & interaction effect: $F_{(8, 56)} = 12.843$, $p = 0.001$, $\eta p^2 = .441$) in the menthol-spray condition (11.0 (2.4) cm) compared to the control-spray (15.7 (1.6) cm; p = 0.02). These ratings corresponded to the worded descriptors '*warm*' to '*hot*' in the control-spray and '*cold*' in the menthol-spray condition. The differences due to menthol-spraying remained until 40-minutes where TS was not different ($p = .255$). Following the second administration of menthol-spray TS once again declined (i.e. participants felt cooler) significantly ($p = .035$); see figure 1A.

Insert figure 1 near here

The differences in TS only resulted in numerical changes in TC after spray application (no condition effect: $F_{(1, 7)} = 2.297$, $p = .173$, $\eta p^2 = .247$; no interaction effect: $F_{(8, 56)} = 4.789$, $p = .270$, $\eta p^2 = .155$) probably because of larger variation in the TC response than TS. At 25-minutes, after first spray application, TC averaged 9.0 (3.9) cm and 11.8 (1.6) cm in the control-spray and in the menthol-spray conditions respectively corresponding to the worded descriptors '*uncomfortable*' and '*comfortable*'; see figure 1B.

RPE did not differ between conditions (condition effect: $F_{(1, 7)} = .057$, $p = .819$, $\eta p^2 = .008$) or show any interaction effect ($F_{(6, 42)} = .782$, $p = .620$, $\eta p^2 = .101$). RPE was always within one RPE rating between condition; see figure 1C.

Thermal Responses (Including fc)

One T_{rec} file was corrupted and consequently data from this participant were removed (T_{rec} data n = 7). T_{rec} increased steadily throughout FI exercise and the TTE, indicating that the exercise produced heat at a rate that was uncompensable (main effect for time: $F_{(7, 42)} = 49.490$, $p = .001$, $\eta p^2 = .892$); see figure 2A. There was no difference between condition ($F_{(1, 6)} = .017$, $p = .899$, $\eta p^2 = .003$) or interaction effect for T_{rec} ($F_{(7, 42)} = 2.097$, $p = .182$, $\eta p^2 = .259$). Terminal rectal temperature was 38.5 (0.26) and 38.4 (0.37) ºC in the control-spray and menthol-spray conditions respectively. The T_{skin} response was similar for the first 20-minutes of FI exercise before spray application. Despite the changes in TS, there was no evident condition effect for T_{skin} ($F_{(1, 7)} = .444$, $p = .527$, $\eta p^2 = .105$) or any interaction effect ($F_{(7, 49)} = .575$, $p = .389$, $\eta p^2 = .147$) although T_{skin} did change numerically in the same direction as the TS ratings. These data indicate an uncoupling of the T_{skin} and thermal perceptual response; see figure 2B. Following the first menthol-spray application the T_{skin} response had a tendency to be numerically lower until the commencement of the TTE; see figure 2B. *f*c was similar throughout each condition and averaged 171 (14) $b \cdot min^{-1}$ and 174 (7) $b \cdot min^{-1}$ in the control-spray and menthol-spray condition at test cessation. There was no difference between condition ($F_{(1, 7)} = .053$, $p = .825$, $\eta p^2 = .008$) or interaction ($F_{(5, 35)} = .108$, $p = .990$, $\eta p^2 = .015$).

Insert figure 2 near here

Fluid Consumed, Sweat Produced, Blood lactate and Cardiac Frequency

The volume of fluid consumed by each participant was relatively consistent between conditions and averaged 630 (169) mL and 545 (187) in the control-spray and menthol-spray conditions ($t = 1.12$, $p = .149$). These data combined with naked body mass measurements generated an estimated sweat production of 1180 (380) mL and 994 (380) mL in the control-spray and menthol-spray conditions with production being lower after menthol-spray ($t = 3.002$, $p = .020$). Due to the significantly longer exercise duration in the menthol-spray condition the estimated sweat rate (827 (327) $mL \cdot hr^{-1}$) was reduced ($t = 2.392$, $p = .048$) versus the control-spray condition (941 (319) $mL \cdot hr^{-1}$).

Terminal B_{lac} at the end of the FI period was 4.3 (2.1) mmol/L and 5.1 (3.1) mmol/L in the control-spray and menthol-spray conditions and was not different ($t = 1.189$, $p = 0.273$); further B_{lac} data not shown.

Discussion

The present study sought to examine whether *repeated* application on menthol-spray to the torso enhanced exercise performance in trained cyclists in an exercise task which was limited by tolerance rather than power output. Our data showed an improvement in TTE performance of 133 (104) seconds after menthol-spraying in contrast to a control-spray condition; H_1 is therefore accepted. We also suggested that *repeated* menthol-spray application would provide a greater benefit to thermal perception thereby driving behavioural thermoregulation. Our data suggest that only thermal sensation was significantly improved although thermal comfort did alter subjectively in the hypothesised direction. The performance change through perceptual mechanisms did not manifest itself through lowered perceived exertion; we therefore only provide partial support for H_2. An additional novel finding was the change observed in sweat production and sweat rate following repeated menthol-spray application which we have not seen previously with single application studies using this menthol concentration.

To our knowledge, this is the first study to investigate the possibility of an ergogenic effect of repeated menthol application using an ecologically valid protocol and a menthol concentration similar to those commercially available. The fact that repeated menthol application is required to produce an ergogenic effect provides a challenge to performers of sports where a weight bearing component may be limiting to their performance (e.g. running, tour cycling). A decision to carry and deploy menthol must be balanced against any performance decrement induced by bearing the additional weight. Moreover, our evidence that the perturbation in thermal perception was lesser after the second menthol spray application also suggests that repeated chemical stimulation of the skin may have limitations especially in a hot environment. Indeed, we speculate that repeated menthol application is likely to have a lesser effect because of acute habituation to the sensation[18] or because of an increased contribution of raised deep body temperature to thermoreception thereby reducing the contribution T_{skin} makes to thermal perception[19]. Even in the scenario of hot skin and a normothermic deep body temperature, menthol may evoke warm sensations if the mean T_{skin} is over 37 ºC which has been shown in isolated cells to activate warm sensitive thermoreceptors TRPA and TRPV[6]. In the present study, activation of these thermoreceptors by menthol may also contribute to the lessened perceptual effect with repeated application. Consequently, a combination of peripheral and visceral thermoreceptor stimulation may be a more viable target for performance enhancement rather than visceral or peripheral alone. There is good evidence that menthol ingestion is performance enhancing[10] and we show here it is premature to conclude that topical application is not. It is now also plausible that topical menthol application could be ergogenic in other activities (e.g. strength and power-based activities) which could be limited by hot environments or the perceptual mechanisms we describe here and elsewhere in relation to RPE[3].

The fact that repeated menthol-spray also altered sweating response by reducing it is also a novel finding although others have reported delayed sweating and reduced sweat production occurs after 4.6% menthol sediment application[12]. The extent of the reduction we see in the present study, albeit using different protocols and menthol concentrations (i.e. 0.20% *cf* 4.6%), was far lower (i.e. 12% *cf* 63% of sweat response seen in the control condition) than reported elsewhere[12] indicating a dose response relationship for menthol application to the skin. Others have also reported that menthol application activates different heat gain responses including vasoconstriction with resultant increases in rectal temperature[11,12]. Although we did not see the latter, we also saw evidence that T_{skin} was lowered after menthol-spray application (see figure 2B) indicating possible vasoconstriction. Any change in T_{skin} was also less substantial on secondary application supporting the idea that visceral thermoreceptors are applying a greater predominance of thermoregulatory input as deep body temperature increases[20]. Collectively across our study and those of others, we must be cautious when titrating the concentration and frequency of menthol application during exercise to avoid inducing heat gain responses which may increase heat illness risk, especially during high intensity efforts where heat load would be high or when performing in high ambient temperatures. This is especially prudent since an uncoupling of thermal state from thermal perception is plausible with menthol application thereby placing biophysical and behavioural thermoregulatory drivers in conflict. Using a menthol-spray of lower concentration which still induces perceptual benefits but does not alter thermoregulatory response (e.g. .05% concentration) may be a safer option to safeguard health [8,11,12]. Moreover, the addition of ethanol to the spray mix, which was deliberately excluded in the present and previous studies to maximise perceptual cooling through chemical stimulation and minimise physiological cooling through evaporation, may ensure the perceptual and thermoregulatory responses converge[20].

Practical Applications

Menthol-spray application triggers heat gain responses which could increase risk of heat illness in some circumstances and care should be taken with the concentration and frequency of application. The performance benefit of menthol-spray could be extended to other population groups (i.e untrained persons) and activities where perceptions are partially limiting. However, this must be balanced against the logistical burden to carrying and deploying the spray.

Conclusion

Repeated menthol-spray application is ergogenic in trained participants during cycling in hot conditions. The perceptual benefits of repeated menthol spraying are likely to be dependent on thermal profile with a diminishing effect when there is an increasing contribution of visceral thermoreceptors to thermoreception; i.e. when deep body temperature is raised.

References

1. Tucker R, Marle T, Lambert EV, Noakes TD. The rate of heat storage mediates an anticipatory reduction in exercise intensity during cycling at a fixed rate of perceived exertion. *J Physiol*, 2006, 574, 905–915.
2. Schlader ZJ, Simmons SE, Stannard SR, Mundel T. The independent roles of temperature and thermal perception in the control of human thermoregulatory behaviour. *Physiol Behav*, 2011, 103: 217-24.
3. Barwood MJ, Corbett J, Thomas K, Twentyman P. Relieving thermal discomfort: effects of sprayed L-Menthol on perception, performance and time trial cycling in the heat. *Scand J Med Sci in Sport*, 2015, S211-S218.
4. McKemy DD, Neuhausser WM, Julius D. Identification of a cold receptor reveals a general role for TRP channels in thermosensation. *Nature,* 2002, 416, 52-58.
5. Peier AM, Moqrich A, Hergarden AC, Reeve AJ, Andersson DA, Story GM *et al*. A TRP channel that senses cold stimuli and menthol. *Cell*, 2002, 108, 705-15.
6. Macpherson LJ, Hwang SW, Miyamoto T, Dublin AE, Patapoutian A, Story GM. More than cool: promiscuous relationships of menthol and other sensory compounds. *Mol Cell Neurosci*, 2006, 32, 335-343.
7. Green BG. Menthol modulates oral sensations of warmth and cold. *Physiol Behav*, 1985, 35, 427-434.
8. Barwood MJ, Corbett J, James J, White D. Early change in thermal perception is not a driver of anticipatory exercise pacing in the heat. *Br J Sports Med*, 2012, 46(13), 936-942.
9. Barwood MJ, Corbett J, White D. Spraying with 0.20% L-Menthol does not enhance 5k running performance in the heat in untrained runners. *J Sports Med Phys Fitness*, 2014, 54(5), 595-604.
10. Stevens CJ, Best R. Menthol: a fresh ergogenic aid for athletic performance. *Sports Med*, 2018, 47(6), 1035-1042.
11. Gillis DJ, House JR, Tipton MJ. The influence of menthol on thermoregulation and perception during exercise in warm, humid conditions. *Eur J Appl Physiol*, 2010, 110, 609-618.
12. Kounalakis SN, Botonis PG, Koskolou MD, Geladas ND. The effect of menthol application to the skin on sweating rate response during exercise in swimmers and controls. *Eur J Appl Physiol*, 2010, 109, 183-189.
13. DuBois D, DuBois EF (1915) The measurement of the surface area of man. *Int Arch Med*, 15, 868-81.
14. Bentley D J, McNaughton LR, Thompson D, Vleck VE Batterham AM. Peak power output, the lactate threshold, and time trial performance in cyclists. *Med Sci Sport Exerc*, 2001, 33(12), 2077-2081.
15. Olesen BW. How many sites are necessary to estimate a mean skin temperature? In: Hales JRS, ed. *Thermal Physiology*. New York: Raven Press; 1980, 33-38.
16. Zhang H. *Human thermal sensation and comfort in transient and non-uniform thermal environments (published dissertation)*. Berkley: CA; University of California; 2003.
17. Borg GAV. Psychological bases of perceived exertion. *Med Sci Sport Exerc* 1982, 14, 377–381.

18. Gillis DJ, Weston N, House JR, Tipton MJ. Influence of daily menthol exposure on human temperature regulation and perception. *Physiol Behav*, 2015, 139, 511-518.
19. Attia M, Engel P. Thermal pleasantness sensation: an indicator of thermal stress. *Eur J Appl Physiol Occup Physiol*, 1982, 50(1), 55-70.
20. Nadel ER, Horvath SM, Dawson CA, Tucker A. Sensitivity to peripheral and central thermal stimulation in man. *J Appl Physiol*, 1970, 29(5), 603-609.
21. Gillis DJ, Barwood MJ, Newton PS, House JR, Tipton MJ. The influence of a menthol and ethanol soaked garment on human temperature regulation and perception during exercise and rest in warm, humid conditions. *J Therm Biol*, 2016, 58, 99-105.

Figure Legends

Figure 1. Mean (SD) TS (panel A), TC (panel B) and RPE (panel C) response at rest, during FI exercise and at TTE end in the control-spray (circles) and menthol-spray (squares) conditions; *indicates significant difference between conditions at a given time point; --- indicates application of spray.

Figure 2. Mean (SD) T_{rec} (panel A) and T_{skin} (panel B) response at rest, during FI exercise and at TTE end in the control-spray (circles) and menthol-spray (squares) conditions; *indicates significant difference between conditions at a given time point; --- indicates application of spray.