THE EFFECTS OF A COOLING COLLAR ON RUNNING PERFORMANCE AND CAPACITY IN A HOT ENVIRONMENT

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Copyright statement

Abstract

Exercise performance and capacity are both impaired in hot compared to moderate environments although the mechanisms behind the premature termination of exercise are not fully understood. It is clear that the development of hyperthermia plays an integral role and so a variety of cooling interventions have been investigated in an attempt to attenuate the reduction in exercise performance and capacity observed. Cooling the torso has received substantial attention however, based upon data from animal, human and modelling studies, cooling the neck has been proposed as a more effective site to cool. The current thesis investigated the effect of a cooling collar on running performance and capacity in hot conditions (30 – 32°C; 53% rh). The thesis also examined the effect of the cooling collar on the physiological, perceptual and neuroendocrinological responses to the exercise bout. Chapter 4 reported that 15 min time-trial running performance was impaired by 9.8% in hot (30°C), compared to moderate (14°C), conditions. The application of the cooling-collar improved 15 min time-trial performance in a hot environment (30°C) in subsequent studies by ~6% and ~7% (Chapters 5, 6 and 8); attenuating much of the impairment in performance observed in Chapter 4. The improvements were observed without alterations in the physiological or neuroendocrinological response to the exercise bout. The rating of perceived exertion was also unaffected, although the collar positively altered the perception of thermal sensation. Cooling the neck also improved exercise capacity in a hot environment (32°) (by ~14%) due to a dampening of the perceived level of thermal and cardiovascular strain. Chapter 7 reported that cooling the neck had no performance benefit when the body was under insufficient thermal strain while Chapter 8 reported that there is a limit to the performance improvements that can be achieved. The data presented within the current thesis demonstrates that cooling the neck via a practical neck collar can improve running performance and capacity in a hot environment; this improvement has many potential advantages for athletes and coaches.

Acknowledgements

I would like to acknowledge the support and guidance that my director of studies, Caroline Sunderland, has offered me recently while I have been working on this thesis and previously as my placement supervisor while I was an undergraduate student at the University of Bath. Her enthusiasm for research is infectious and her supervisory style has allowed me to develop a wide-range of skills and become the researcher, and person, that I am now. Thank you, Caz.

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Without the enthusiasm and continued efforts of the participating volunteers the research presented within this thesis would not be possible and therefore I will be eternally grateful for the integral part that they all played in my research. Thank you all.

Finally, I will never be able to thank my parents enough for their unconditional and continued love, support and belief in me. **Thank you, Mum. Thank you, Dad. This thesis is for you.**

Publications

The following publications have contained work reported in this Thesis:

Journal articles

Tyler, C. and Sunderland, C. (2008). The effect of ambient temperature on the reliability of a preloaded treadmill time-trial. *International Journal of Sports Medicine*, **29**, 812-816.

Conference abstracts, presentations and proceedings

Tyler, C. and Sunderland, C. (2008). Short duration neck cooling does not improve 15-minute treadmill time-trial performance in the heat. *Proceedings of the European College of Sport Sciences*, Estoril, Portugal.

Tyler, C. and Sunderland, C. (2008). Neck cooling during exercise in the heat improves subsequent treadmill time-trial performance. *Medicine and Science in Sports and Exercise*, **40(5)**, S368.

Tyler, C. and Sunderland, C. (2007). The reliability of a 90-minute preloaded treadmill time-trial in hot and moderate ambient temperatures. *Medicine and Science in Sports and Exercise*, **39(5)**, S309.

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List of Abbreviations

%	Percentage
°C	Degrees Celsius
ANOVA	Analysis of variance
b·min^{-1}	Beats per minute
BM	Body mass
BV	Blood volume
C	Non-cold Collar trial
CC	Cold collar trial
CCrep	Cold collar replaced trial
cm	Centimetre
CV	Coefficient of variation
ELISA	Enzyme-linked immunosorbent assay analysis
FAM	Familiarisation/habituation trial
g	Gram
h	Hour
Hb	Haemoglobin
Hct	Haematocrit
HOT	Hot trial
HR	Heart rate
kg	Kilogram
km	Kilometre
L	Litre
m	Meter
min	Minute
ml	Millilitre
mm	Millimetre
mmol	Millimolar
MOD	Moderate trial
NC	No collar trial
ng	Nanogram
$\dot{V}\,\mathbf{O_{2max}}$	Maximal oxygen uptake
PV	Plasma volume
r	Pearson's product-moment correlation coefficient
rh	Relative humidity
RPE	Rating of perceived exertion

s	Second
SD	Standard deviation
t	Time
$\mathbf{T_{amb}}$	Ambient temperature
$\mathbf{T_{brain}}$	Brain temperature
$\mathbf{T_{exh}}$	Test to exhaustion
$\mathbf{T_{neck}}$	Neck temperature
$\mathbf{T_{rectal}}$	Rectal temperature
TS	Thermal sensation
$\mathbf{TS_{neck}}$	Thermal sensation of the neck region
TT	Time-trial
$\mathbf{TT_{pre}}$	Preloaded time-trial
µg	Microgram

Chapter 1: General Introduction

It has been suggested that a moderate elevation in body temperature may be advantageous for exercise because it has been demonstrated that the speed of the mechanical and metabolic processes increases by a factor of approximately two for every 10°C increase in muscle temperature. This is referred to as the Q_{10} temperature quotient or Van't Hoff-Arrhenius law (Belehradek, 1957). In contrast to the benefits associated with moderate increases in body temperature, excessive elevations have regularly been shown to impair both prolonged submaximal exercise (Gonzalez-Alonso *et al.*, 1999; Tucker *et al.*, 2004; Watson *et al.*, 2005c) and short-duration higher-intensity bouts (Gonzalez-Alonso & Calbet, 2003; Nybo & Nielsen, 2001a; Saltin *et al.*, 1970).

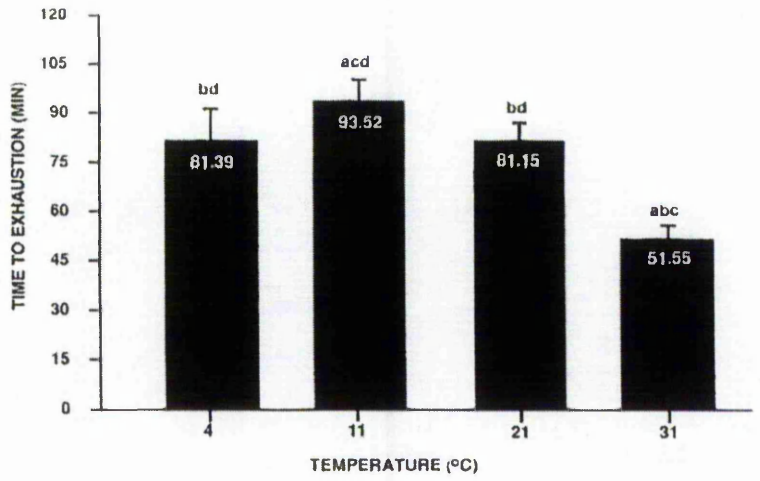

Figure 2.1. The effect of ambient temperature on exercise capacity [reproduced from Galloway and Maughan (1997), with permission].

The detrimental effect of hot environmental conditions on exercise performance is well documented (Galloway & Maughan, 1997; Marino *et al.*, 2004). Galloway and Maughan (1997) demonstrated the adverse effect of elevated ambient temperatures on exercise capacity (Figure 2.1.). They investigated the effect of different ambient temperatures on the ability to sustain steady-state (70% $\dot{V}O_{2max}$) cycle ergometer exercise and reported that participants managed to sustain the exercise bout for the longest period of time at temperatures of 11°C with an impairment in capacity observed in the trials conducted in temperatures in excess of this (21 and 31°C). Participants cycled for 93.5 ± 6 min at 11°C; however, exercise capacity was reduced to 81.2 ± 6 min when the temperature was increased to 21°C and was further reduced to 51.6 ± 4 min when the ambient temperatures were at their highest (31°C). The reduced ability to sustain exercise in hot environmental conditions is not contained to laboratory investigations. The fastest times for races of distances between 5000m and 100km are rarely recorded in temperatures in excess of 14°C (Marino, 2004), while the percentage of athletes completing the Olympic marathons

occurring between 1896 and 2000 drops from ~79% to ~54% when the ambient temperature exceeds 25°C (Marino, 2004; Martin & Gynn, 2000).

Although the exercise-limiting effect of high ambient and internal temperatures is well-documented, the mechanisms involved are not fully understood. A number of theories have been proposed and these are reviewed in Chapter 2 of this thesis. A common theme to all of the proposed mechanisms is the development of hyperthermia. Hyperthermia is defined as an increase in the temperature of the body above the normal range specified for a normal active state and is therefore classified as an elevation of core temperature above ~38°C during moderate-intensity exercise (IUPS Thermal Commission, 1987). Due to the impairment in exercise performance and capacity being heavily linked to an increase in body temperature a variety of cooling methods have been investigated in an attempt to attenuate the reduction in the ability to exercise observed in a hot environment (Marino, 2002). Many studies have examined cooling the body prior to exercise (pre-cooling) and the majority have reported an improvement in performance in the subsequent exercise bout (Gonzalez-Alonso *et al.*, 1999; Lee & Haymes, 1995; Marino, 2002). The improved performance observed following pre-cooling is often attributed to a reduction in initial core temperature, and an attenuated rate at which it increases, widening the range of the internal temperature which can be tolerated (Gonzalez-Alonso *et al.*, 1999; Lee & Haymes, 1995). Despite this proposal performance benefits have been observed following pre-cooling in the absence of core temperature alterations and the positive effect of pre-cooling on thermal comfort and the disruption of thermal feedback channels has also been proposed as an explanation for the ergogenic effect (Hessemer *et al.*, 1984; Kay *et al.*, 1999). The exact mechanisms by which pre-cooling improves performance in hot environments remain unclear. The majority of pre-cooling interventions lack practical application in a competitive sporting environment but despite this there is a lack of research investigating the effect of practical cooling devices. Cooling vests have been investigated, with mixed results (Arngrimsson *et al.*, 2004; Duffield *et al.*, 2003). The mixed results appear to be largely due to the provision of an insufficient magnitude of cooling which is a common problem with practical cooling devices. A potential solution to this is cooling the neck region as it has been demonstrated that cooling this region is more effective in improving thermal tolerance than cooling the same surface area of the torso (Shvartz, 1976) and so perhaps a reduced magnitude of cooling is required if cooling there. Under conditions of severe thermal strain, cooling the head and neck can elicit beneficial thermoregulatory and cardiovascular changes (Nunneley *et al.*, 1971; Shvartz, 1970; Simmons *et al.*, 2008) although improvements in perceptual variables are more common (*e.g.* Armada-da-Silva *et*

al., 2004; Mundel *et al.*, 2005; Simmons *et al.*, 2008). The head and neck region is a site of high sudomotor and alliesthesial thermosensitivity (Cotter & Taylor, 2005) and so improvements in perceptual responses following cooling this region is unsurprising.

Despite the data from Shvartz (1976) and Cotter and Taylor (2005), which suggests that the neck region might be an optimal site to cool, there is a lack of research investigating the effects of cooling the neck during exercise performed in a hot environment. The aim of this thesis was to examine the effects of cooling the neck via a practical neck cooling device on running performance and capacity in a hot environment and to investigate the physiological, perceptual and neuroendocrinological responses to the cooling intervention.

The thesis is presented in nine main Chapters (Chapter 2 - 10); the contents of which are outlined below:

- Chapter 2 presents a review of literature examining the mechanisms proposed to explain the reduced ability to exercise in hot environmental conditions, the cooling strategies investigated in an attempt to attenuate this reduction and the rationale for cooling the neck region.
- In Chapter 3 the general procedures, measurements, equipment and methods of analysis used within the experimental Chapters are presented.
- Chapter 4 investigates the effect of ambient temperature on the reliability of a preloaded treadmill time-trial.
- Chapter 5 investigates the effect that a practical neck cooling collar has on preloaded time-trial running performance in a hot environment.
- Chapter 6 presents data collected during the experiment reported in Chapter 5 and investigates the effect of the neck cooling collar on a marker of blood-brain barrier and neuronal damage.
- The effect of the cooling collar on short-duration time-trial performance in a hot environment is investigated in Chapter 7.
- In Chapter 8 the effect of maintaining the neck at a reduced temperature, via the replacement of a neck cooling collar, on pre-loaded time-trial performance in a hot environment is investigated.
- In Chapter 9 the effect of the cooling collar on exercise capacity in a hot environment is examined.

- The final Chapter of thesis (Chapter 10) discusses and summarises the results from the six preceding experimental Chapters and addresses some of the questions that subsequent research should seek to resolve.

Chapter 2: Review of Literature

2.1. Overview

This Chapter collates data from the most pertinent research studies that have proposed differing theories to explain the premature termination of exercise in hot, compared to moderate, environments and from studies that have investigated a variety of methods to combat the reduced ability to exercise. The review of literature is divided into three main sections. The first section (section 2.2) examines the proposed mechanisms for fatigue during exercise in a hot environment. Section 2.3 reviews the literature investigating the effects of different cooling interventions adopted both prior to, and during exercise. Section 2.3 is divided into three main sub-sections focussing on pre-cooling, cooling the torso and cooling the head, neck and face region. Section 2.4 examines the theoretical basis for cooling the neck region.

2.2. Proposed mechanisms of fatigue during exercise in hot environments

2.2.1. Introduction

Exercise capacity is limited in a hot environment (Galloway & Maughan, 1997) and it has been proposed that, from an evolutional perspective, there should be physiological safeguards in place to initiate the termination of physical activity (*e.g.* exercise or work) prior to the onset of catastrophic hyperthermia (MacDougall *et al.*, 1974; Nielsen *et al.*, 1993; Cheung, 2007). Two major paradigms have been proposed in an attempt to explain the premature termination of exercise in a hot environment. One theory suggests that fatigue occurs upon the attainment of a critical internal temperature (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993; Walters *et al.*, 2000), while the alternative perspective proposes that the body reduces its workload prior to the attainment of a dangerously high temperature due to a complex feed-forward and feedback system or a central governor (Marino, 2004; Noakes & St Clair, 2004). Often these two models are seen as conflicting and exclusive of each other (Marino, 2004); however, it has also been proposed that the two models are in fact complementary and that the existence of both protective systems would be advantageous to survival (Cheung, 2007). The critical core temperature hypothesis can only be examined during fixed intensity work while the anticipatory, central governor, model can only be assessed using performance tests with a variable work-load and this fundamental difference probably accounts for much of the proposed conflict. The purpose of this section is to review the literature that has focused on these

models of exercise fatigue in a hot environment and to explore the similarities and differences between the two.

2.2.2. The critical core temperature hypothesis

Observations that participants consistently terminate fixed-intensity exercise at core temperatures of ~40°C regardless of acclimation status (Nielsen *et al.*, 1993) or initial core temperature (Gonzalez-Alonso *et al.*, 1999) led to the hypothesis that there is a critical body temperature that limits exercise in the heat (Nielsen *et al.*, 1993). The mechanical efficiency of the human body is normally below ~25% and more than 75% of the total energy used is converted into heat (Astrand *et al.*, 2003). During exercise performed in environmental temperatures below ~28°C, the magnitude of elevation in core temperature is largely independent of the ambient temperature and is proportional to the metabolic rate (Gonzalez *et al.*, 1978); however, in higher temperatures the metabolic load acts in combination with the ambient conditions to increase the elevation of core temperature.

The recent interest in the possible existence of a critical core temperature stemmed largely from a study investigating the effect of heat acclimation on the human circulatory and thermoregulatory response to exercise in a hot environment (Nielsen *et al.*, 1993). Nielsen *et al.* (1993) reported that, although exercise capacity time almost doubled after a 9 – 12 day acclimation protocol, voluntary exhaustion occurred consistently at an internal (oesophageal) temperature of ~39.7°C regardless of acclimation status following submaximal (60% $\dot{V}O_{2max}$) cycle ergometer capacity tests in hot, dry conditions (40°C; 10% rh). At the point of exhaustion there were no reductions in cardiac output, muscle blood flow or skin blood flow and there was no lack of substrate or any accumulation of any substance traditionally thought to cause fatigue such as lactate or potassium ions and therefore the authors proposed the concept of exercise-limiting hyperthermia. A similar consistency of terminal core temperature was observed in moderately fit participants exposed to uncompensable heat stress while wearing nuclear, biological and chemical protective clothing (Cheung & McLellan, 1998) and in individuals commencing exercise at different starting temperatures (Gonzalez-Alonso *et al.*, 1999). Participants wearing the protective clothing were divided into two groups depending on fitness status and were restricted to a cut off core temperature of 39.3°C due to ethical constraints. In the high fitness group ($\dot{V}O_{2max}$ > 55 $ml \cdot kg^{-1} \cdot min^{-1}$), 24 trials were terminated due to reaching the ethically enforced cut-off core temperature with only 8 trials voluntarily terminated prior to the attainment of this temperature. In the lower fitness group ($\dot{V}O_{2max}$ 40 - 50 $ml \cdot kg^{-1} \cdot min^{-}$

[1]) all trials were voluntarily terminated below the 39.3°C ethically approved ceiling temperature and at consistent temperature of ~38.7°C. This data demonstrates that training status can affect thermal tolerance because individuals with higher levels of aerobic fitness were able to tolerate higher core temperatures. Further support for the idea of a critical cut-off core temperature was provided by Gonzalez-Alonso and colleagues (1999). Participants performed submaximal (60% $\dot{V}O_{2max}$) cycle ergometer exercise to volitional exhaustion on three occasions with manipulated starting core temperatures of 36, 37 and 38°C. Exercise capacity was inversely related to the starting core temperature. Participants completed 63 ± 3, 46 ± 3 and 28 ± 2 min in the cool, control and hot trials respectively. Despite the marked differences in capacity time, all trials were voluntarily terminated at remarkably consistent core temperatures- 40.1 ± 0.1, 40.2 ± 0.1 and 40.1 ± 0.1°C (Gonzalez-Alonso *et al.*, 1999) (Figure 2.2.).

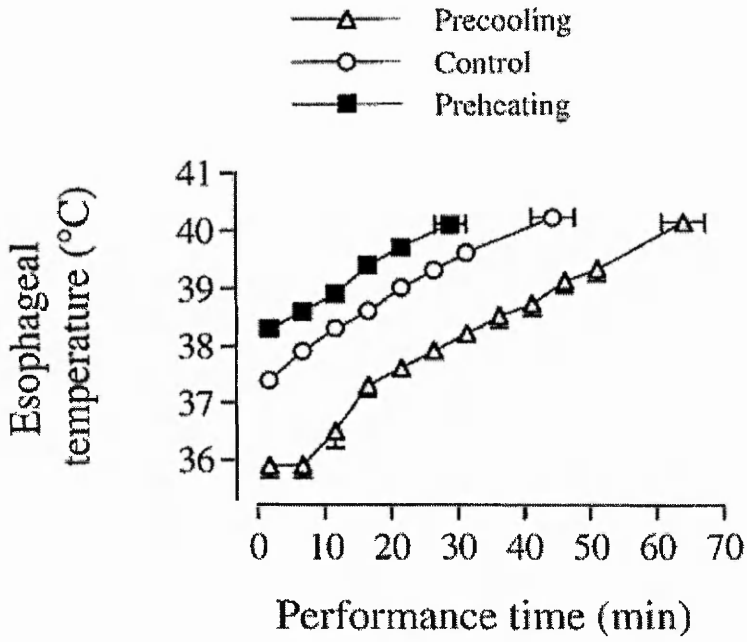

Figure 2.2. The consistency in core temperature observed at voluntary exercise termination [reproduced from Gonzalez-Alonso *et al.* (1999), with permission].

The data from Nielsen *et al.* (1993), Cheung and McLellan (1998) and Gonzalez-Alonso *et al.* (1999) demonstrates the consistency of temperatures at which human participants voluntarily terminate exercise. The idea that the attainment of a high internal temperature is a major factor limiting exercise performance, capacity and/or adherence had already been demonstrated in animal research (Caputa *et al.*, 1986). Caputa *et al.* (1986) investigated the relative contributions of elevated brain and trunk temperatures on the limitation of exercise performance in the heat (32°C). Toggenburg goats were trained daily for approximately one month walking on a treadmill at 3km·h^{-1}. There was a daily increase in the gradient of the treadmill until the largest workload that could be tolerated for 60 min for each animal was established. Once this workload had been established the goats

repeatedly walked on the treadmill for 60 min at 3km·hr^{-1} (16 - 20% gradient) in hot conditions (32°C). During the trials the temperature of the hypothalamus and trunk were manipulated via implanted thermoelements. Caputa *et al.* (1986) demonstrated that core and hypothalamic temperatures of up to 43.5°C and 42.6°C respectively could be tolerated and that the animals could complete 60 min of moderate exercise despite experiencing such high internal temperatures. These were the highest single levels of tolerance and both were greater than the highest combined thermal load tolerated for 60 min which was 43°C and 42°C for trunk and hypothalamic temperatures respectively. This data demonstrates that higher core than hypothalamic temperatures can be tolerated and indicates that a high hypothalamic temperature is the main factor that limits motor activity. More recent animal-model research has confirmed the existence of consistent hypothalamic temperatures at the point of fatigue and added support for the existence of a 'safety switch' (Fuller *et al.*, 1998; Walters *et al.*, 2000). Fuller *et al.* (1998) exercised male Sprague-Dewley rats to exhaustion in ambient temperatures of 23°C and 33°C and reported that although capacity time was significantly greater in the 23°C trials the abdominal temperatures and hypothalamic temperatures at fatigue were not significantly different between conditions. The rats fatigued at higher hypothalamic than abdominal temperatures (~40.1°C vs. 39.9°C) although the difference between the two sites was less pronounced than observed in the Toggenburg goats (Caputa *et al.*, 1986; Fuller *et al.*, 1998). Further support for the existence of a safety switch was provided by Walters *et al.* (2000) who rapidly heated Sprague-Dewley rats via exposure to microwaves to three different levels of thermal strain. As previously reported there was a significant negative correlation between capacity time and initial core temperature but in this study there was no significant difference between the rectal and hypothalamic temperatures at fatigue (41.9 - 42.2 vs. 42.2 - 42.5°C). Different exercise models have demonstrated that rats will run to heatstroke leading to death (Fruth & Gisolfi, 1983) however the rats in the study by Walters *et al.* (2000) fatigued prior to reaching a critical thermal load and all survived the exercise bout suggesting that exercise was voluntarily terminated by the animals before health and physiological function was compromised.

2.2.2.1. Hyperthermia and neuromuscular impairment

Due to the consensus amongst thermal physiologists that the premature termination of exercise in hot, compared to moderate, environments occurs due to the obtainment of a high internal temperature, the focus of research has begun to shift to investigations examining the mechanism(s) that trigger the termination of exercise.

It has been suggested that hyperthermia may have a direct effect upon the central nervous system (Nielsen *et al.*, 2001; Nybo & Nielsen, 2001b; Nybo & Nielsen, 2001a). More specifically, it has been suggested that hyperthermia may result in a reduction in the drive to the motorneuon pool (Gandevia, 2001) and there appears to be a centrally regulated reduction in neuromuscular activity when internal temperatures are elevated (Nybo & Nielsen, 2001a). Nybo and Nielsen (2001a) demonstrated that force production and the percentage of voluntary activation in exercised muscle groups (knee extensors) was reduced during sustained isometric maximal voluntary contraction following submaximal (60% $\dot{V}O_{2max}$) cycle ergometer exercise in hot (40°C) compared to temperate (18°C) conditions (Figure 2.3.). During the submaximal cycle preload in the hot conditions the participants reached core temperatures of ~40°C and prematurely terminated exercise after ~50 min while in the cooler conditions core temperature plateaued at ~38°C and the 1h of exercise was completed without exhaustion occurring. Despite reaching exhaustion in the hot trials superimposed electrical stimulation induced the same overall force in both conditions demonstrating that the capacity for the muscle to generate force was unaffected by the ambient conditions or the elevated core and muscle temperatures observed in the hot trials (Figure 2.3.).

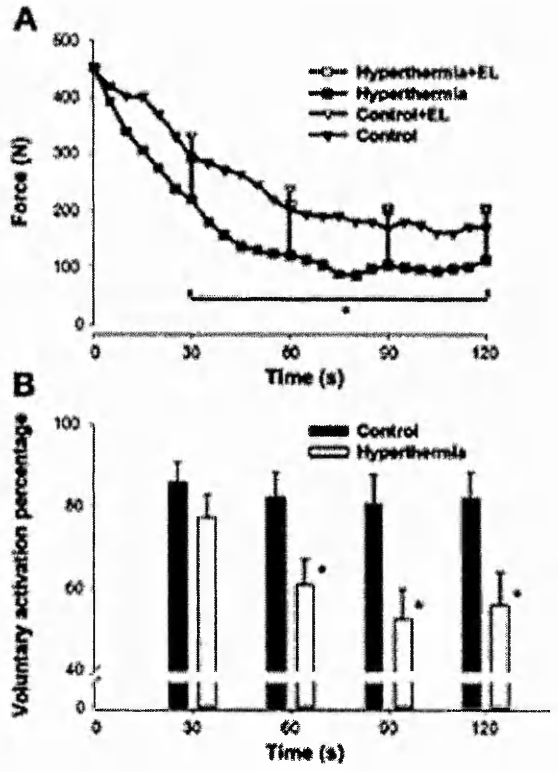

Figure 2.3. The changes in force (A) and voluntary activation (B) during 2 min of sustained maximal voluntary contraction with the knee extensors during hyperthermia and control. * = $P < 0.05$ [reproduced from Nybo and Nielsen, (2001a), with permission].

Nybo and Nielsen (2001a) also reported that the attenuated ability to generate force was not limited to the working muscles as a similar pattern was observed for non-exercised muscles during sustained hand-grip contraction. Although these results demonstrate a reduction in central activation as a result of hyperthermia, additional results from this study demonstrate that the central nervous system can quickly regain the ability to activate the skeletal muscles (Nybo & Nielsen, 2001a). In this adjoining study, Nybo and Nielsen (2001a) reported that although the ability to sustain an isometric contraction was reduced due to hyperthermia the elevated core temperature had no effect on maximal force development or central activation in 2 second repeated maximal voluntary contractions when they were interspersed with 3 second bouts of recovery. Although Nybo and Nielsen (2001a) demonstrated that hyperthermia reduced the ability to sustain an isometric contraction as a result of the study design, in which only pre and post measurements were taken, it was not possible to establish the nature of the decline in force generation. For example, the decline may have been progressive or may only have occurred upon the attainment of a high internal temperature. To allow for the collection of data as the individual becomes progressively hyperthermic, and to further explore the relationship between hyperthermia and the reduction in muscular contraction, passive-heating models have been used in an attempt to establish the nature of the decline (Cheung & Sleivert, 2004a; Morrison *et al.*, 2004).

Morrison *et al.* (2004) investigated the effect of manipulating core and skin temperature on the ability to produce a maximal voluntary isometric contraction of the quadriceps femoris muscle group. Maximal voluntary isometric contractions were performed at 0.5°C core temperature intervals increasing from ~37.5°C to ~39.5°C and then at the same intervals returning to ~37.5°C. Warming and cooling were both induced via a fluid-perfused garment circulated with either 52°C or 8°C fluid. The force produced during the maximal voluntary contractions and the percentage of voluntary activation decreased significantly from the start to the end of the passive warming bout. After the passive warming bout, rapid cooling of the skin (- 8°C) was initiated. Despite decreasing cardiovascular and psychophysical strain, the skin cooling intervention had no effect on core temperature (it remained stable at ~39.5°C), maximal voluntary contraction or voluntary activation (Figure 2.4). This data suggests that an elevated core temperature was the primary limiting factor. Further support for this proposal was provided by data which showed that the percentage voluntary activation and the maximal voluntary contraction returned to baseline levels at the end of the cooling bout when the core temperature had returned to ~37.5°C with a progressive recovery observed as core temperature decreased. Data from a subsequent

investigation from the same research group also reported a decline in voluntary isometric force production and muscle activation with progressive hyperthermia and established, via bilateral investigation (heating one calf while maintaining the other thermoneutral), that core rather than local muscle temperature is the key factor that causes this reduction (Thomas *et al.*, 2006). Isometric contractions are impaired following passive and active hyperthermia (Morrison *et al.*, 2004; Nybo & Nielsen, 2001a; Thomas *et al.*, 2006) and the impairment appears to be core temperature dependent (Morrison *et al.*, 2004). However, during isokinetic contractions, lowering skin temperature decreases the maximal voluntary contraction independent of core temperature (Cheung & Sleivert, 2004a). Cheung and Sleivert (2004a) passively warmed their participants from ~37.5°C to ~39.5°C via water immersion and then cooled as per Morrison *et al.* (2004) back down to a core temperature of ~38°C. Isokinetic maximal voluntary contractions were performed at speeds of 60, 120 and 240 $deg \cdot s^{-1}$ at 0.5°C core temperature intervals. The torque output of the isokinetic contractions at all three speeds was unaffected by the elevation in core temperature but was significantly reduced during the cooling-phase at all comparative core temperatures observed during the warming-phase. The data from these studies clearly demonstrate the effect that manipulating the thermal state of the body can have on the ability to generate force. The effect of the thermal manipulation and the magnitude of the impairment observed appear dependent on the length and type of contraction (dynamic v static), although the exact mechanisms behind this remain unclear (Cheung & Sleivert, 2004b; Nybo & Nielsen, 2001a).

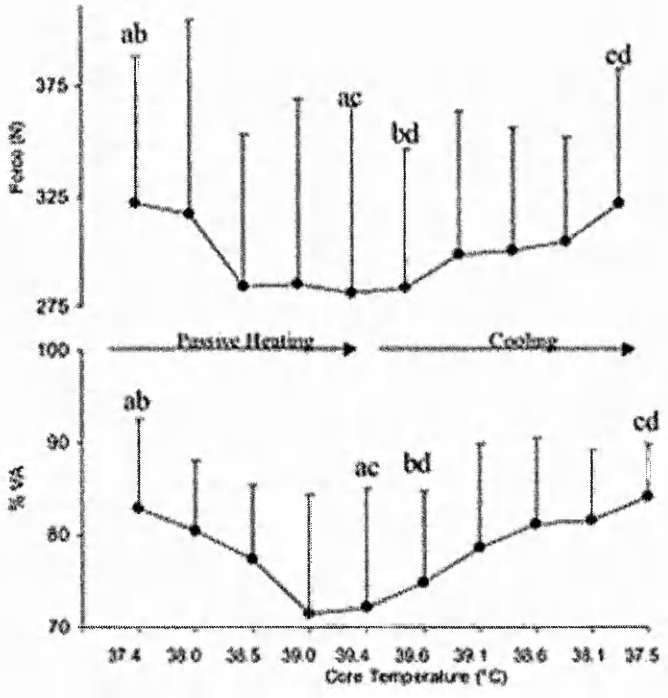

Figure 2.4. Knee extension maximal voluntary contraction (top) and voluntary activation (bottom) during passive heating and cooling. Matching letters indicate significant difference ($P < 0.001$) [reproduced from Morrison *et al.* (2004), with permission].

2.2.2.2. The critical core temperature hypothesis summary

During fixed-intensity exercise, voluntary termination consistently occurs at core temperatures of ~40°C in both humans and animals (Caputa *et al.*, 1986; Fuller *et al.*, 1998; Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993; Walters *et al.*, 2000). The temperature at which termination occurs is influenced by factors such as motivation and training status (Cheung & McLellan, 1998; Pugh *et al.*, 1967). The reason for the termination of exercise at such temperatures is not fully understood although voluntary force production is attenuated with hyperthermia (Cheung & Sleivert, 2004a; Morrison *et al.*, 2004; Nybo & Nielsen, 2001a; Thomas *et al.*, 2006) and therefore this down-regulation in force development may play a key role in the reduced ability to sustain exercise.

2.2.3. The central governor theory

The critical core temperature theory proposes that exercise is terminated due to the attainment of a high core temperature which results in a catastrophic physiological event; however, it has been shown that, during self-paced exercise, a down-regulation of running speed and power output occurs long before such temperatures are reached (Marino *et al.*, 2004; Tucker *et al.*, 2004). Tucker *et al.* (2004) reported that rectal temperatures were the same for the first 15km (75%) of a 20km cycling time-trial in hot and moderate conditions, but despite the similar temperatures, work rate and skeletal muscle recruitment were down-regulated in the hot conditions within the first 30% of the trial. Based upon these observations it has been suggested that pacing strategies are adopted during exercise which prevent the attainment of a critically high temperature. This theory, termed the central governor theory, suggests that exercise in the heat is not limited by a fatigue process at a critical physiological threshold but by an anticipatory process regulated by an internal feedforward and feedback system which is activated as soon as exercise is initiated and ensures that catastrophe is avoided and that the task can be completed within homeostatic limits (Lambert *et al.*, 2005; Marino *et al.*, 2004; Marino, 2004; Morrison *et al.*, 2004; Tucker *et al.*, 2004) (Figure 2.6).

An anticipatory down-regulation in exercise intensity can only be observed in self-paced investigations because in capacity tests the exercise intensity is externally controlled and the physiological responses occur proportionally to the intensity and metabolic rate (Marino, 2004). Tatterson *et al.* (2000) reported that 30 min cycling time-trial performance

was impaired in hot (32°C) compared to moderate (23°C) conditions but despite the differences in ambient conditions and total work done participants finished the time-trial at near identical core temperatures (39.2 ± 0.2 v 39.0 ± 0.1°C; $P > 0.05$). Tatterson *et al.* (2000) proposed that the cyclists adjusted their power output to ensure that the core temperature did not prematurely reach a 'critical' level. Marino *et al.* (2004) provided further support for the anticipatory mechanism when comparing Caucasian and African runners. Participants ran a 30 min preload at 70% $\dot{V}O_{2max}$ prior to a self-paced 8km performance test in both hot (35°C) and cool (15°C) conditions. The African runners outperformed the Caucasian runners in the hot (29.7 ± 2.3 v 33 ± 1.6 min) but not cool trials (27.4 ± 1.0 v 27.4 ± 0.4 min). In the hot trials, the rectal temperature at the end of the preload phase was almost identical for both groups (~38.4°C) and was similar at the end of the self-paced performance test (39.2 ± 0.2 vs. 39.5 ± 0.5°C) despite the Caucasians immediately starting the performance test at a significantly lower self-selected speed and adopting a lower mean speed than the African runners throughout (Figure 2.5). The selection of a lower speed took place immediately and therefore prior to the attainment of a limiting temperature.

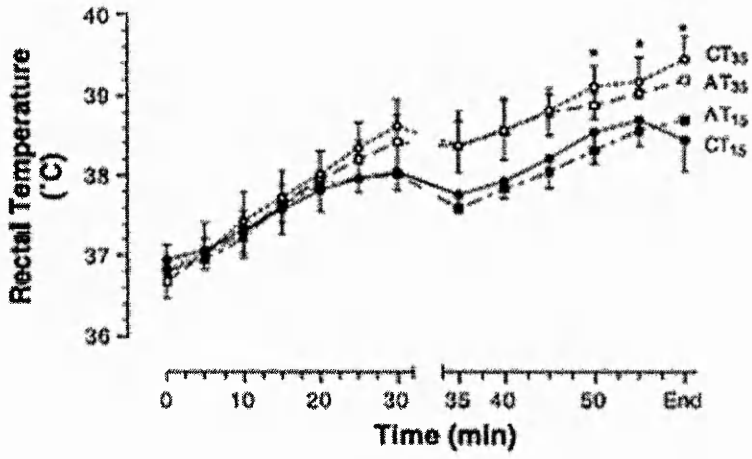

Figure 2.5. Rectal temperatures observed in Caucasian (C) and African (A) runners in hot (T_{35}) and temperate (T_{15}) conditions. * = $P <$ 0.05 [reproduced from Marino *et al.* (2004), with permission].

Marino *et al.* (2004) proposed that rather than being limited by the attainment of a critically high temperature, exercise performance is determined by the rate at which body temperature increases. In the study comparing Caucasian and African runners, the larger Caucasian runners (1.98 ± 0.2 vs. 1.66 ± 0.1m^2) reduced their pace early in the time-trial. Marino *et al.* (2004) postulated that this was due to the larger Caucasian runners having a reduced heat storage capacity and reducing their speed in anticipation of that. Participants terminated exercise at similar rectal temperatures in both the Caucasian and African groups and the magnitude of core temperature change was ~1.7°C in both groups. Further research

by Marino and colleagues (2004) reported that the provision of enough fluid to maintain body mass during exercise had no effect on the final rectal temperature recorded compared to a no fluid trial however it attenuated the rate at which rectal temperature increased which allowed participants to exercise for longer in the euhydration trial. Similar findings were reported by Fuller *et al.* (1998) who exercised rats in ambient conditions of 33°C and 38°C and manipulated their brain temperatures to be identical in each trial. Prior to the exercise bouts the rats were held in either cool (23°C) or hot (38°C) conditions. The rats that were pre-cooled in the 23°C conditions had a greater exercise capacity in both hot conditions (33°C and 38°C) which was attributed to the lower rate of core temperature rise (~5 and 6.4°C·h^{-1}) compared to the rate of rise observed in the rats held at 38°C (7.3°C·h^{-1}).

As eluded to earlier, in a study by Tucker *et al.* (2004), participants were asked to run two 20km time-trials at a self-selected pace, one in hot conditions (35°C) and one in cool (15°C). Tucker *et al.* (2004) reported that, although rectal temperatures were the same between trials for the first 75% of the performance test, participants reduced their self-selected speed in the hot conditions after only ~30% of the time-trial had been completed and well before the onset of a critically high core temperature. Marino *et al.* (2004) also identified that runners reduced their running speed in hyperthermic conditions well before the onset of an excessive core temperature (<38.5°C) while Castle *et al.* (2006) reported that peak power output during the last (20th) cycle sprint was elevated compared to the penultimate sprint showing that peripheral fatigue was not the cause of the reduction in exercise performance and suggesting that the participants "held something back" during the latter sprints. It is unclear what signal, or integrated signals, governs self-paced effort and establishing the signal(s) is troublesome as it is likely that it is a combination of physiological and psychological sources with added difficulty provided by individual mediators (*e.g.* training status) (Cheung, 2007). It would seem prudent to suggest that core temperature may provide cues for self-pacing as high core temperature is often linked to the termination of exercise (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993), although it appears that there are other cues too as many studies have reported improvements in power output in cooler conditions (*e.g.* 23°C v 32°C; (Tatterson *et al.*, 2000)) or following pre-cooling (Kay *et al.*, 1999) despite similar core temperatures.

Evidence for the role of the central nervous system in the down-regulation of skeletal muscle recruitment during exercise-induced hyperthermia has been provided by electrical stimulation studies. By superimposing an electrical stimulation on top of a voluntary contraction and calculating the ratio between the voluntary and evoked contractions the

level of central activation can be evaluated (Nybo & Nielsen, 2001a). As previously discussed, Morrison *et al.* (2004) reported a gradual reduction in voluntary activation percentage when participants were passively warmed from rectal temperatures of ~37.5°C to temperatures ~39.5°C. The activation percentage was restored when the internal temperatures returned to the initial values indicating a gradual reduction in central nervous system drive as hyperthermia develops. The central activation ratio was reduced in hyperthermic compared to normothermic conditions when the hyperthermia was induced by both active and passive means in exercised muscles (Nybo & Nielsen, 2001a). In addition, although central activation was not evaluated, hyperthermia also resulted in a reduction in the maximal voluntary contraction of non-exercised muscles (Nybo & Nielsen, 2001a). Saboisky *et al.* (2003) also reported that exercise-induced hyperthermia resulted in a reduction in force output and central activation in exercised muscle (leg extensors) but failed to observe similar findings in the non-exercise muscles (forearm flexors) investigated. The terminal temperature was only ~38.8°C- well below the temperatures often cited as limiting (~40°C) suggesting that rather than the obtainment of an extreme core temperature, hyperthermia *per se* may reduce the central drive to the skeletal muscle (Saboisky *et al.*, 2003). The discrepancy between the studies regarding the non-exercise muscle groups suggests that the hyperthermia-induced skeletal muscle down-regulation may be a selective, rather than consistent, outcome.

2.2.3.1. The central governor theory summary

The obtainment of a high core temperature results in the termination of exercise in tests of fixed intensity (Gonzalez-Alonso *et al.*, 1999; Walters *et al.*, 2000) whereas the central governor theory highlights that in self-paced activity work-rate is down-regulated well before a high core temperature is reached (Marino *et al.*, 2004; Tatterson *et al.*, 2000). The central governor theory proposes that this down-regulation is an evolutionary adaptation to prevent developing core temperatures that would lead to the onset of heat illness or injury. The central governor theory and critical core temperature models are often reported as being opposing viewpoints; however, they are in fact very similar in their main theme of preventing the development of a high core temperature and the onset of potentially fatal heat illness. The differences reported appear to exist largely as a result of the exercise model investigated.

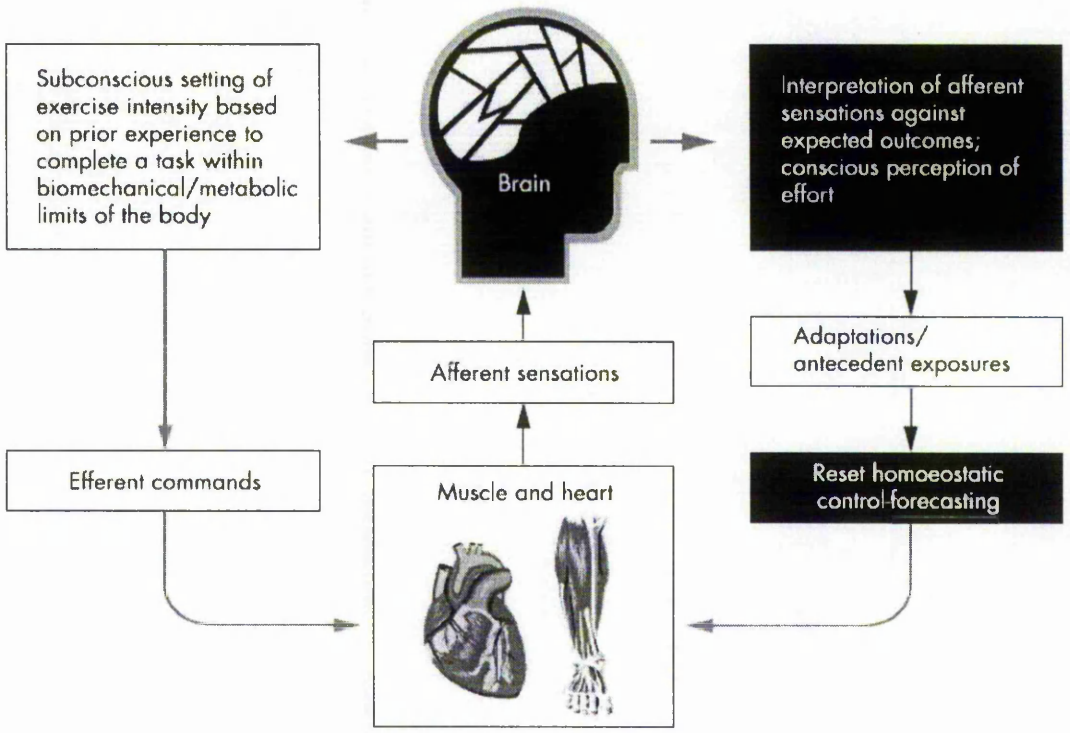

Figure 2.6. Diagrammatic representation of the central governor theory and the proposed teleoanticipatory control of exercise intensity [reproduced from Lambert *et al.* (2005), with permission].

2.2.4. The central fatigue theory

Exercise is regularly impaired in hot environments and although the exact mechanisms are yet to be established the development of hyperthermia appears to be a key factor in both constant and variable work-rate models (Gonzalez-Alonso *et al.*, 1999; Noakes & St Clair, 2004; Thomas *et al.*, 2006; Tucker *et al.*, 2004). Depending on the exercise test adopted the attainment of a high internal temperature results in either the cessation of exercise or the down-regulation of self-selected intensity and although the critical core temperature and central fatigue hypotheses provide models from which to gain a greater understanding of the reasons limiting performance, they do not describe how hyperthermia ultimately limits exercise.

Fatigue is a complex, multi-faceted phenomenon. Exercise is believed to start and end in the brain (Kayser, 2003) and therefore many researchers have suggested that the fatigue observed due to hyperthermia may occur due to alterations in cerebral neurotransmitter activity and concentrations (Meeusen *et al.*, 2006). The altered synthesis and metabolism

of central monoamines forms the basis of the central fatigue hypothesis and the monoamines that are particularly implicated are serotonin and dopamine (Meeusen *et al.*, 2006).

2.2.4.1. Serotonin and the central fatigue hypothesis

The neurotransmitter that has probably received the greatest level of interest is serotonin (Blomstrand, 2001; Davis *et al.*, 1993; Davis & Bailey, 1997; Newsholme & Blomstrand, 1995; Newsholme & Blomstrand, 2006). Serotonin (5-hydoxytryptamine) is a monoamine neurotransmitter linked to the augmentation of lethargy and the loss of drive and serotonin forms the basis of the original central fatigue hypothesis (Newsholme & Blomstrand, 1995). Serotonin is unable to cross the blood-brain barrier and therefore for cerebral concentrations to increase cerebral neurons are required to synthesise it from serotonin's precursor, the amino acid tryptophan. The increased synthesis observed during exercise is thought to occur as a result of an exercise-induced increase in serum concentrations of nonesterified fatty acids (NEFA) and the subsequent indirect elevation of the cerebral levels of free tryptophan. Tryptophan binds to albumin in the blood, as do NEFA, and the binding processes of tryptophan and the fatty acids to albumin is a competitive one (McMenamy *et al.*, 1957). Tryptophan hydroxylase is the enzyme that converts tryptophan to serotonin- under normal conditions it remains unsaturated and therefore the limiting factor for the synthesis of serotonin is the availability of free tryptophan (Fernstrom, 1983). At rest only about 10-20% of the total tryptophan found within the circulation is free, unbound tryptophan however during exercise substantially greater levels of free tryptophan are present as a result of the elevated NEFA concentrations. The central fatigue hypothesis proposes that during exercise the increase in NEFA concentrations observed during prolonged exercise displaces tryptophan molecules from the albumin causing an increase in the circulating free tryptophan concentrations (Curzon *et al.*, 1974) which causes a subsequent increase in tryptophan crossing the blood-brain barrier and an increase in the synthesis of serotonin in the brain.

Support for this theory was provided by Chaouloff *et al.* (1986) who demonstrated that forced treadmill exercise elevated concentrations of free peripheral and cerebral tryptophan but had no effect on the total tryptophan concentrations in rats. They interpreted these findings to indicate that exercise resulted in an increase in serotonin synthesis, turnover and release due to a NEFA-induced increase in free (but not total) tryptophan (Chaouloff *et al.*, 1986).

This model was accepted by most exercise research groups but this area had previously received extensive attention in the pharmacological communities and the general consensus appears to be that peripheral free tryptophan concentrations have very little influence on the cerebral uptake of the amino acid. Studies have demonstrated that although dietary manipulation of free tryptophan via the administration of a high fat diet resulted in a sustained (2h) two-fold increase in serum free tryptophan, comparable to the increases observed following exercise (Blomstrand *et al.*, 1989; Chaouloff *et al.*, 1985; Chaouloff *et al.*, 1986) it had no effect on the tryptophan levels within the cerebral cortex or hypothalamus (Fernstrom & Fernstrom, 1993). Similar results were reported by Chaouloff *et al.* (1985) who abolished the rise in plasma NEFA via the administration of nicotinic acid and showed that despite abolishing the rise in free tryptophan the concentrations within the brain still increased. These findings demonstrate that the exercise-induced elevations in cerebral tryptophan concentrations do not require an increase in free tryptophan and this discrepancy raises questions regarding the cause of the exercise-induced tryptophan elevations.

Additionally, according to the central fatigue theory the administration of a serotonin antagonist would be expected to impair exercise capacity while a serotonin agonist would have the opposite effect and improve time to exhaustion. The administration of a serotonin receptor antagonist had no effect on running capacity (Pannier *et al.*, 1995) or performance in hot conditions (35°C) (Strachan *et al.*, 2005) nor did it alter cycle ergometer capacity in laboratory temperatures (Meeusen *et al.*, 1997) and so although the rationale for the serotonin-centred central fatigue hypothesis is clear human experimental data is not convincing (Nybo, 2007).

2.2.4.2. Dopamine and the central fatigue hypothesis

Several neurotransmitter systems are activated during exercise and many of these are located within key thermoregulatory areas such as the hypothalamus and preoptic region (Meeusen *et al.*, 2006). Although the serotonin hypothesis is the most widely discussed, evidence in human studies is lacking and it would seem naïve to suggest that the multi-faceted phenomenon that is fatigue is under the control of one physiological or neuroendocrinological pathway (Nybo, 2007). One alternative suggestion involves the neurotransmitter dopamine. Where as serotonin is linked to feelings of lethargy and a lack

of motivation dopamine is linked with feelings of arousal and motivation (Chaouloff, 1989).

Dopamine elevations caused by the administration of amphetamines benefits exercise performance in rats (Gerald, 1978; Bailey *et al.*, 1993) and humans (Chandler & Blair, 1980). It is thought that the benefit may be due to a reduction in the level of serotonergic activity occurring within the brain as concentrations of the serotonin metabolite 5-HIAA are reduced following amphetamine administration (Chaouloff *et al.*, 1987). Despite the link between dopamine and exercise relatively few studies have investigated this area in humans. In one investigation the effect of L-3,4-dihydroxyphenylalanine (L-DOPA) was explored (Meeusen *et al.*, 1997). L-DOPA is an intermediate in the synthesis of the catecholamine and used in the management of Parkinson's disease (Meeusen *et al.*, 2006). Previous data has demonstrated that L-DOPA elevates extra-cellular dopamine concentrations in the rat striatum (Dethy *et al.*, 1995) however Meeusen *et al.* (1997) reported that ingestion of L-DOPA had no effect on the exercise capacity of endurance trained males cycling to exhaustion in temperate conditions. Two recent publications have replicated these results reporting that the acute administration of both bupropion, a dopamine/noradrenaline reuptake inhibitor, and Methylphenidate (Ritalin), a dopamine reuptake inhibitor, had no effect on pre-loaded time-trial performance in ambient conditions of 18°C, however significantly improved performance in hot (30°C) conditions (Roelands *et al.*, 2008a; Watson *et al.*, 2005c). These results suggest that the role of dopamine in the onset of fatigue may be important only in hyperthermic conditions.

2.2.4.3. Central fatigue and hyperthermia

Although both serotonin and dopamine have been implicated in the control of thermoregulation (Lipton & Clark, 1986; Strachan *et al.*, 2005) and despite the fact that the areas of fatigue in a hot environment and central fatigue have received a substantial amount of interest individually, relatively few investigations that explored the effect of pharmacological manipulation during exercise in a hot environment.

The role of serotonin in the onset of central fatigue is an often cited explanation for the reduction in exercise performance and/or capacity in hot conditions but few studies have investigated the effects of hyperthermia on cerebral serotonin levels and those that have appear to dispute this proposed mechanism (Hasegawa *et al.*, 2000; Ishiwata *et al.*, 2004). Manipulation of cerebral levels of serotonin via the administration of fluoxetine (a

selective serotonin reuptake inhibitor) and 8-OH-DPAT (a 5-HT_{1A} agonist) following hot (35°C) or cold (5°C) exposure demonstrated that core temperature was unaffected by alterations in PO/AH serotonin levels (Ishiwata *et al.*, 2004). This suggests that serotonin may not be the mediator of thermoregulatory adjustments and Hasegawa *et al.* (2000) proposed that dopamine, rather than serotonin, may be the key thermoregulatory neurotransmitter. Hasegawa *et al.* (2000) reported that PO/AH dopaminergic neural activity was increased by moderate exercise and the increased activity was associated with a reduction in the heat storage observed. While dopaminergic activity was elevated the extracellular concentrations of serotonin and its metabolite 5-HIAA were unaffected.

Acute administration of bupropion (a duel dopamine/noradrenaline reuptake inhibitor) has been shown to improve pre-loaded time-trial performance in hot (30°C) conditions by ~9% however it had no effect on the same performance test conducted in moderate (18°C) temperatures (Watson *et al.*, 2005c). Watson *et al.* (2005a) reported that seven of the nine participants in this study achieved rectal temperatures of $\geq$ 40°C in the bupropion trial compared to only 2 achieving such temperatures in the control trial. The enhanced performance in the warm trials was observed without an alteration in the perceptual measurements despite higher core temperatures. This suggests that the administration of the pharmacological agent may have overridden inhibitory signals allowing the participants to upregulate their work rate. Recent data has shown that although an acute dose of bupropion can improve time-trial performance in the heat (Watson *et al.*, 2005c) chronic bupropion supplementation (10-day programme) has no effect in identical conditions (Roelands *et al.*, 2008b) suggesting that chronic administration may result in an adaptation of the central neurotransmitter homeostasis and a subsequent alteration in the response to the drug. This highlights the difficulty in elucidating the role of the neurotransmitter systems on the onset of fatigue and in the regulation of body temperature. In an attempt to further explore the mechanisms two recent follow-up studies investigated the effects of administering a specific noradrenaline reuptake inhibitor (reboxetine) (Roelands *et al.*, 2008c) and a specific dopamine reuptake inhibitor (Ritalin) (Roelands *et al.*, 2008a) on exercise in moderate (18°C) and hot (30°C) environments (Figure 2.7). The same pre-loaded time-trial protocol adopted in the previous bupropion study was used. It was established that preloaded time-trial performance was improved by ~16% following the administration of Ritalin in warm conditions (30°C), although elevating dopamine concentrations had no effect on performance in moderate (18°C) trials (Roelands *et al.*, 2008a) and that performance was impaired in both conditions (by ~10% in moderate and by ~20% in hot) following reboxetine administration. The exercise performance benefit

observed following Ritalin dosage in a hot environment was greater than that observed following bupropion and this, in addition to the impaired time-trial performance observed following reboxetine suggests that dopamine plays a dominant role in thermoregulation and the enhancement of exercise in a hot environment.

2.2.4.4. Central fatigue during exercise in a hot environment summary

Although the rationale for the serotonin-centred central fatigue theory is clear, evidence from studies conducted in elevated conditions and on human participants is not convincing. In contrast, the role of dopamine in the onset of fatigue appears to be particularly important during exercise in hot environments. The manipulation of cerebral concentrations of dopamine via the acute administration of a dopamine/noradrenaline reuptake inhibitor and a selective dopamine reuptake inhibitor has been shown to improve time-trial performance in hot, but not moderate conditions (Roelands *et al.*, 2008a; Watson *et al.*, 2005c). Chronic administration of a dopamine/noradrenaline reuptake inhibitor had no effect on time-trial performance in a hot environment suggesting that central neurotransmitter homeostasis can adapt and demonstrating that there is still much that is unknown about this area (Roelands *et al.*, 2008b). Recent data from acute administration studies shows that in a hot environment the inhibition of noradrenaline reuptake impairs performance (Roelands *et al.*, 2008c) while the inhibition of dopamine reuptake improves performance (Roelands *et al.*, 2008a) and therefore dopamine appears to play a major role in the enhancement of time-trial performance in a hot environment (Figure 2.7).

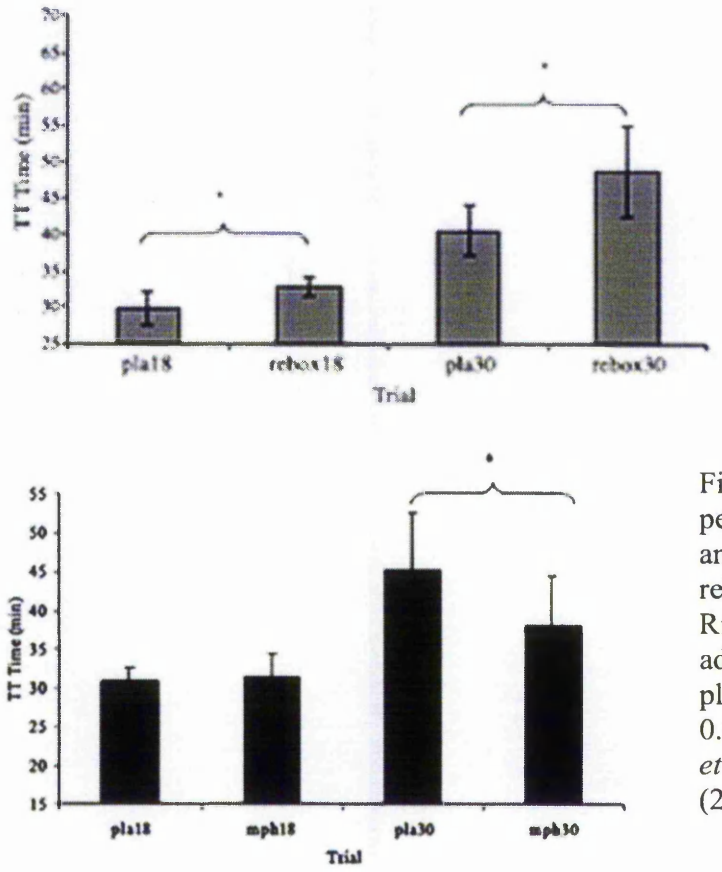

Figure 2.7. Time-trial performance in temperate (18) and hot (30) conditions with reboxetine (rebox) (top) and Ritalin (mph) (bottom) administration compared to placebo (pla) (A and B). * = P < 0.05 [reproduced from Roelands *et al.* (2008a) and Roelands *et al.* (2008c) with permission].

2.2.5. Summary of proposed mechanisms for fatigue in the heat

Exercise performance and capacity are regularly impaired in hot compared to moderate conditions however the reason for the reduced ability is not fully understood. It is clear that during fixed-intensity protocols exercise capacity is voluntarily terminated at a core temperature of ~40°C in both human and animal studies (Fuller *et al.*, 1998; Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993; Walters *et al.*, 2000), and that it is likely to be the obtainment of a high brain, rather than body, temperature that results in the termination (Caputa *et al.*, 1986). In performance studies self-selected workloads are reduced in hot conditions prior to the obtainment of high internal temperatures and it has been suggested that the anticipatory down-regulation allows for the task to be completed within homeostatic limits (Noakes & St Clair, 2004; Tucker *et al.*, 2004). Fatigue in the heat does not occur as a result of many peripherally-located causes of fatigue in temperate conditions (Nielsen *et al.*, 1993) and therefore the suggestion that it is a centrally regulated fatigue appears to be the most plausible proposal. Central levels of a variety of neurotransmitters have been implicated in the onset of fatigue during exercise and the data suggests that cerebral dopamine concentrations play a dominant role in the regulation of exercise

performance and adherence in hot environmental conditions (Roelands *et al.*, 2008a; Roelands *et al.*, 2008c).

2.3. Cooling interventions adopted prior to and during exercise

2.3.1. Introduction

Despite the mechanisms underpinning fatigue in the heat not being fully understood the detrimental effect of hyperthermia is well documented and therefore many applied researchers have investigated methods designed to confront the problem of hyperthermia in an attempt to offset the reduction in performance observed during exercise heat stress. These methods have included acclimation programmes (Sunderland *et al.*, 2008), hydration strategies (Watson *et al.*, 2008) and, the focus of this thesis, cooling interventions (Quod *et al.*, 2006). A variety of different cooling methods have been investigated differing in their times and sites of application. In this section pre-cooling interventions as well as those adopted during exercise and rest bouts will be reviewed.

2.3.2. Whole-body cooling prior to exercise (pre-cooling)

Many different methods of pre-cooling have been investigated and these include water-immersion up to the supra-illiac crest (Duffield & Marino, 2007; Wilson *et al.*, 2002) or up to the to suprasternal notch (Bolster *et al.*, 1999; Booth *et al.*, 1997; Hasegawa *et al.*, 2006; Kay *et al.*, 1999; Booth *et al.*, 2004; Marino & Booth, 1998); cool water showering (Drust *et al.*, 2000); the wearing of ice-cooling garments (Arngrimsson *et al.*, 2004; Cheung & Robinson, 2004; Duffield *et al.*, 2003; Duffield & Marino, 2007; Webster *et al.*, 2005); cold air exposure with air at 0°C (Hessemer *et al.*, 1984; Olschewski & Bruck, 1988; Schmidt & Bruck, 1981) or 5°C (Lee & Haymes, 1995) and a combination of cool-air exposure with the addition of fan-cooling and water-spraying (Ansley *et al.*, 2007; Mitchell *et al.*, 2003). The different effects of these pre-cooling interventions are reviewed in this section.

2.3.2.1: Thermoregulatory response

2.3.2.1.1. Core temperature

Tolerance to high ambient heat exposure is inversely related to the initial core temperature (Veghte & Webb, 1961) and the physiological response to exercise appears to be largely

dependent on the magnitude of change observed in core temperature during the bout (Marino *et al.*, 2004). As discussed previously, an elevated core temperature has been shown to limit endurance capacity and performance (Fuller *et al.*, 1998; Gonzalez-Alonso *et al.*, 1999) and it has been suggested that a major way in which pre-cooling could improve exercise performance is by decreasing the core temperature experienced during exercise at any given time-point thereby reducing the thermal strain, improving the heat storage capacity (Booth *et al.*, 1997; Lee & Haymes, 1995; Wilson *et al.*, 2002), and widening the core temperature range that can be tolerated before a high level of hyperthermia is reached (Marino, 2002).

Pre-cooling has often been shown to result in an initial increase in core temperature (Hessemer *et al.*, 1984; Lee & Haymes, 1995; Olschewski & Bruck, 1988) and it has been proposed that this initial elevation may be due to the rapid cooling-induced vasoconstriction of the peripheral blood vessels directing the warm blood to the core (Schmidt & Bruck, 1981) and/or shivering-induced heat production (Lee & Haymes, 1995). When aggressive cooling methods are adopted a lag-time between the commencement of pre-cooling and a decrease in core temperature is observed (Booth *et al.*, 1997). This phenomenon has been described as an 'afterdrop' in core temperature (Romet, 1988). Kay *et al.* (1999) reported that core temperature was unaffected by the pre-cooling intervention at the onset of an exercise bout following pre-cooling; however, 15 min into the bout of self-paced cycling core temperature was significantly reduced compared to the non-cooled trial. Other studies have reported that core temperature was significantly lower at the beginning of exercise compared to at the end of the pre-cooling intervention (Booth *et al.*, 2004; Drust *et al.*, 2000; Marino & Booth, 1998). It seems most likely that the 'afterdrop' occurs as a result of the cooled peripheral blood returning to the core and that the delay is due to the necessity to remove the extra heat transferred to the core as a result of the redirection of the peripheral blood caused by the initial vasoconstriction (Schmidt & Bruck, 1981). Less extreme magnitudes of cooling may prevent this 'afterdrop' in core temperature by providing an insufficient stimulus to promote vasoconstriction (Marino & Booth, 1998). Despite the initial increases in core temperature that are regularly observed following a pre-cooling intervention, pre-cooling has been shown to significantly lower the core temperature at the onset of exercise in most (Booth *et al.*, 1997; Castle *et al.*, 2006; Cheung & Robinson, 2004; Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Mitchell *et al.*, 2003; Wilson & Maughan, 1992; Arngrimsson *et al.*, 2004) but not all (Duffield *et al.*, 2003; Duffield & Marino, 2007) studies. Duffield and Marino (2007) investigated the effect of pre-cooling via cooling vests

and water immersion and found that core temperature was only significantly lowered via the water immersion protocol suggesting that the method and magnitude of the pre-cooling intervention is important. Arngrimsson *et al.* (2004) also investigated the effects of pre-cooling via a cooling vest. They reported that the vest significantly lowered core temperature for the first 3.6km of the 5km time-trial after which there was no difference between the control and the cooling groups. This demonstrates that cooling vests can reduce core temperature and have a practical application but also suggests that their effect is heavily magnitude dependent. Despite some studies showing no alterations in core temperature following pre-cooling the majority show that pre-cooling can reduce core temperature but that a sufficient lag time between the cessation of the pre-cooling and commencement of the exercise bout is required to allow for the 'afterdrop' phenomenon (Romet, 1988). In addition to increasing the core temperature range that can be tolerated, by lowering the initial core temperature, pre-cooling has also been shown to attenuate the rate at which core temperature rises during exercise (Hunter *et al.*, 2006; Webster *et al.*, 2005) and has been shown to result in a decreased core temperature for up to 90 min of exercise (Booth *et al.*, 1997; Cheung & Robinson, 2004; Drust *et al.*, 2000; Hasegawa *et al.*, 2006; Wilson *et al.*, 2002). The rate at which core temperature rises has been heavily implemented in the regulation of exercise performance and capacity (Marino *et al.*, 2004; Marino, 2004).

2.3.2.1.2. Skin temperature

Pre-cooling often results in a reduction in skin temperature, lasting up to 60 min, during exercise (Booth *et al.*, 1997; Castle *et al.*, 2006; Cheung & Robinson, 2004; Hasegawa *et al.*, 2006; Kay *et al.*, 1999; Lee & Haymes, 1995; Mitchell *et al.*, 2003; Arngrimsson *et al.*, 2004) and an attenuation of the rate at which skin temperature increases (Webster *et al.*, 2005) however Duffield *et al.* (2003) and Duffield and Marino (2007) found no reductions in skin temperature following pre-cooling via the application of a cooling vest. It is likely that the observations of Duffield *et al.* (2003) and Duffield and Marino (2007) are as a result of the cooling mechanism used (a cooling vest worn over the exercise attire), the short duration of the cooling intervention (5 to 15 min) and the fact that 3 of the 4 thermistors worn were in no direct contact with the vest because Arngrimsson *et al.* (2004) reported that skin temperature could be lowered at the onset of exercise via the application of a cooling vest during a 38 min active warm-up bout. As the underlying mechanism by which pre-cooling is thought to work relies on the constant cooling of the circulating peripheral blood, it is unsurprising that pre-cooling regularly reduces the temperature of

the skin and it is possible that the extent of the benefits gained from pre-cooling are directly related to the magnitude by which the intervention can cool the skin.

2.3.2.1.3. Muscle temperature

To the author's knowledge only three studies have measured muscle temperature following pre-cooling. Gonzalez-Alonso *et al.* (1999) cooled to a specific muscle temperature (~4°C reduction) via 30 min of cold (17°C) water immersion and so the reduction in muscle temperature was deliberately manipulated where as Booth *et al.* (2004) and Castle *et al.* (2006) monitored muscle temperature following pre-cooling interventions. Booth *et al.* (2004) reported that muscle temperature was significantly reduced by 6.3°C following 60 min of cool-water immersion prior to 35 min of submaximal (~60% $\dot{V}O_{2max}$) cycling. During the submaximal cycling bout the rate at which the temperature of the pre-cooled muscle increased was significantly greater than in the control group ($0.23 \pm 0.01°C \cdot min^{-1}$ v $0.10 \pm 0.01°C \cdot min^{-1}$) however the terminal muscle temperature remained lower following pre-cooling. Castle *et al.* (2006) investigated the effect of three different methods of pre-cooling (Cooling vest, cold water immersion (18°C) and cool pack application placed on the thigh- all applied for a 20 min bout) on peak power output and repeated 5-second sprint performance. While there was no reduction in the temperature of the vastus lateralis observed in the control or vest trials, muscle temperature was lowered at a rate of $0.06 \pm 0.002°C \cdot min^{-1}$ and $0.1 \pm 0.03°C \cdot min^{-1}$ with the cool water immersion and cool packs respectively. Although the muscle temperature was lower at the beginning of the exercise in both the cool water immersion and cool pack groups the difference was only significant in the cool pack group (lower by $1.0 \pm 2.5°C$, $P < 0.05$). Castle *et al.* (2006) reported that the cooling effect of the ice packs lasted until the eighth sprint (time = 16 min) while water immersion maintained the muscle at a significantly lower temperature than observed in the control throughout the protocol. In summary, pre-cooling can reduce muscle temperature and the extent to which it does so is dependent on the magnitude of the cooling (*i.e.* the water temperature) and the duration which the muscle is exposed to the cooling intervention.

2.3.2.2. Cardiovascular response

Mixed results have been reported for the heart rate response to pre-cooling. Some studies have shown heart rate is unaffected by pre-cooling (Drust *et al.*, 2000; Duffield *et al.*, 2003; Hessemer *et al.*, 1984; Hornery *et al.*, 2005; Kay *et al.*, 1999; Marsh & Sleivert,

1999; Olschewski & Bruck, 1988; Webster *et al.*, 2005) while others have reported cooling-induced bradycardia following such an intervention (Arngrimsson *et al.*, 2004; Bolster *et al.*, 1999; Castle *et al.*, 2006; Cheung & Robinson, 2004; Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Lee & Haymes, 1995; Mitchell *et al.*, 2003; Schmidt & Bruck, 1981; Sleivert *et al.*, 2001; Uckert & Joch, 2007; Wilson *et al.*, 2002). Castle *et al.* (2006) investigated three different cooling methods and found that heart rate was unaffected when cooling was induced by cooling packs or a cooling vest but it was lowered via cool water immersion for the first 16 min of the 40 min protocol. Similar results were reported by Duffield and Marino (2007), who also reported that pre-cooling via a cooling vest had no effect on the heart rate recorded while water immersion reduced the heart rate for the first 10 min of the protocol, and Arngrimsson *et al.* (2004), Booth *et al.* (1997) and Lee and Haymes (1995) who all reported that although heart rate can be reduced during the early stages of exercise by pre-cooling there are no prolonged effects. It has been suggested that the cooling-induced bradycardia may be due to the cooled skin resulting in decreased cutaneous blood flow and thus a reduced need to elevate heart rate to maintain cardiac output (Drust *et al.*, 2000). This may explain the lack of a prolonged response because as the cooling stimulus decreases and body temperatures increase over time, the skin will warm and blood will be redirected to the periphery compromising cardiac output unless heart rate is elevated.

2.3.2.3. Body fluid regulation

Pre-cooling appears to help maintain body fluid balance during exercise. In the absence of any differences in voluntary water intake (Kay *et al.*, 1999) pre-cooling has been shown to consistently decrease total body sweating during exercise in moderate conditions (Hessemer *et al.*, 1984; Lee & Haymes, 1995; Olschewski & Bruck, 1988) and decrease sweat loss in some (Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Kay *et al.*, 1999), but not all (Booth *et al.*, 1997; Drust *et al.*, 2000), trials conducted in elevated temperatures. Pre-cooling has also been shown to have an effect on sweat rate and has been shown to reduce it in most (Lee & Haymes, 1995; Webster *et al.*, 2005; Wilson *et al.*, 2002) but not all (Bolster *et al.*, 1999) studies.

It has been suggested that pre-cooling may reduce the thermoregulatory drive to dissipate heat due to the manipulation of body temperatures (Lee & Haymes, 1995) and this certainly appears to be the case with regards to sweat response regulation. The sweat response is believed to be initiated by a combination of core and skin temperature neural

inputs (Nadel *et al.*, 1971b; Nadel & Stolwijk, 1971; Nadel *et al.*, 1971a). The extent to which the ratio between core and peripheral inputs dictate the sweat response is not fully understood however Sawka and Wenger (Sawka & Wenger, 1988) suggested that a 1°C decrease in core temperature has a nine times greater thermoregulatory effector response than the same change in skin temperature.

To the author's knowledge no studies have directly looked at the effect of pre-cooling on the maintenance, or otherwise, of hydration status however the consistently reported decrease in sweat loss and sweat rate combined with unaltered voluntary water consumption (Kay *et al.*, 1999) suggests that pre-cooling may indirectly help prevent the onset of dehydration during exercise in a hot environment.

2.3.2.4. Rating of perceived exertion and thermal comfort

It has been suggested that fatigue is a conscious sensation and that it is the sensation of exertion that governs exercise adherence and/or termination (Noakes & St Clair, 2004). This conscious sensation is often assessed in an exercise setting using the rating of perceived exertion scale (Borg, 1982) on the assumption that the rating given is derived from integrated feedback from a variety of physiological systems (Crewe *et al.*, 2008). As documented, pre-cooling is highly effective in altering the physiological state of the human body however, perhaps surprisingly due to this, it does not reduce the rating of perceived exertion during subsequent exercise in most studies (Arngrimsson *et al.*, 2004; Bolster *et al.*, 1999; Booth *et al.*, 1997; Cheung & Robinson, 2004; Drust *et al.*, 2000; Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Kay *et al.*, 1999; Sleivert *et al.*, 2001; Wilson *et al.*, 2002; Arngrimsson *et al.*, 2004; Duffield & Marino, 2007) although reductions (Hornery *et al.*, 2005; Simmons *et al.*, 2008) have been reported. The studies in which a reduction in the rating of perceived exertion was reported both adopted cooling during exercise and therefore prior to a subsequent bout rather than a single bout. The reduction observed may, therefore, be due to the cooling being administered at a time which the participants were already feeling thermally and physically stressed. Thermal comfort is regularly improved with a pre-cooling intervention (Arngrimsson *et al.*, 2004; Bolster *et al.*, 1999; Booth *et al.*, 1997; Castle *et al.*, 2006; Cheung & Robinson, 2004; Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Mitchell *et al.*, 2003; Sleivert *et al.*, 2001; Wilson *et al.*, 2002) however the improvement is not always observed (Bolster *et al.*, 1999; Hornery *et al.*, 2005; Webster *et al.*, 2005). Of those studies that did not report an improvement Bolster *et al.* (1999) noted that there was a large variation between individual responses and suggested that

comparisons of such a subjective scale was extremely difficult while Hornery *et al.* (2005) and Webster *et al.* (2005) both cooled via a cooling vest which has been shown to be less effective than cooling via other methods (Lopez *et al.*, 2008). Castle *et al.* (2006) compared three types of pre-cooling and found that there was no effect on thermal comfort when individuals were cooled via a cooling vest for 20 min however thermal comfort was significantly improved following cool water immersion for the same duration ($P < 0.01$). This further supports the notion that the magnitude of the cooling dictates the benefits that can be achieved.

2.3.2.5. Exercise performance

One of the main problems with assessing the effectiveness of pre-cooling interventions on exercise performance is the method of assessment chosen by the investigators. Tests to exhaustion (also referred to as capacity/open-loop tests) are regularly used to investigate physiological mechanisms during steady-state exercise however they are not directly applicable to sporting settings and have been found to suffer from poor test-retest reliability and high levels of within-subject variation during cycling (Graham & McLellan, 1989; Jeukendrup *et al.*, 1996; Krebs & Powers, 1989; McLellan *et al.*, 1995), running (Billat *et al.*, 1994) and swimming (Alberty *et al.*, 2006). Most studies have used exercise capacity, rather than performance, tests and so the benefit of pre-cooling on performance can only be inferred in the majority of studies however there is a beneficial trend evident, particularly during prolonged exercise.

2.3.2.5.1. Short duration and high intensity exercise

The effectiveness of pre-cooling on short duration and high intensity exercise is an area of contention, even within research groups. Marsh and Sleivert (1999) reported that $\geq$ 30 min of pre-cooling improved 70s high intensity cycling performance by ~3.3% in warm, humid (29°C, 80% rh) conditions. The authors suggested that the improvement may have been due greater metabolite removal resulting from the pre-cooling induced peripheral vasoconstriction increasing central blood volume and elevating muscle blood flow (Marsh & Sleivert, 1999). This however relies in part upon exercise being limited by impaired muscle blood flow, something that has been shown to not be the case (Nielsen *et al.*, 1993). Hornery *et al.* (2005) investigated the effect of half-time cooling and found that there was a trend for a higher mean work output to be achieved during the 10 min performance section of the trial with 11 of the 14 participants producing a greater work output following

cooling while Webster *et al.* (2005) reported a significant improvement in exercise capacity at 95% $\dot{V}O_{2max}$ following 35 min of pre-cooling (20 min rest, 5 min stretching and 10 min warming-up at 50% $\dot{V}O_{2max}$) via the application of a cooling vest. Cooling the vastus lateralis area via cooling packs increased peak power output by ~4% however there was no difference in the peak power output of the trials in which cooling was induced by water immersion or vest compared to control (Castle *et al.*, 2006). In the same study pre-cooling via the cool packs and the cooling vest significantly improved the total work done in 20 repeated 5-second sprints compared to the control however the water immersion protocol had no effect. These findings highlight differences in the effectiveness of various pre-cooling strategies and methodological differences help explain why pre-cooling has been shown to have no significant effect on work done or power produced during repeated sprinting in other studies (Cheung & Robinson, 2004; Duffield *et al.*, 2003; Duffield & Marino, 2007; Sleivert *et al.*, 2001). In one study, similar to that of Webster *et al.* (2005), Mitchell *et al.* (2003) reported that 20 min of pre-cooling prior to a treadmill run to exhaustion at 100% $\dot{V}O_{2max}$ significantly impaired running performance despite significant reductions in core, skin and body temperatures. In contrast, Duffield and Marino (2007), who also compared cooling vests and water immersion, found no differences in mean 15m sprint time, total sprint time or percentage decline in sprint time between bouts for either cooling intervention compared to control. It has been suggested that high muscle temperatures may be a reason for the onset of fatigue (Drust *et al.*, 2005) and reductions in mean power output (Linnane *et al.*, 2004), however the temperature of the muscle in the study by Castle *et al.* (2006) was ~40°C at the end of the exercise in both the control and the vest group despite differences in performance measures and so muscle temperature alone cannot explain the reduction in performance observed in the control group compared to the vest.

2.3.2.5.2. Prolonged exercise

Although there is some controversy regarding the effectiveness of pre-cooling during intense exercise pre-cooling has regularly been shown to enhance exercise endurance in hot environmental conditions during closed (*i.e.* performance tests) (Booth *et al.*, 1997; Hessemer *et al.*, 1984; Kay *et al.*, 1999; Arngrimsson *et al.*, 2004; Castle *et al.*, 2006; Marsh & Sleivert, 1999) and open (*i.e.* capacity tests) (Gonzalez-Alonso *et al.*, 1999; Hessemer *et al.*, 1984; Lee & Haymes, 1995; Olschewski & Bruck, 1988; Hasegawa *et al.*, 2006; Uckert & Joch, 2007; Webster *et al.*, 2005) exercise tests.

In a classic study by Gonzalez-Alonso *et al.* (1999) it was demonstrated that exercise tolerance time was inversely related to the core temperature at the beginning of the test to exhaustion (Figure 2.2.). They reported that exercise capacity could be enhanced by ~37% by pre-cooling to an oesophageal temperature of 35.9 ± 0.2°C and reduced by ~39% by warming to a temperature of 38.2 ± 0.1°C. Despite the wide range of performance times between conditions participants fatigued at almost identical temperatures (~40.1°C) in all three conditions. Many of the studies that have shown pre-cooling to enhance exercise performance or capacity have attributed the improvement to a reduction in core temperature at any given time-point during the exercise bout. Booth *et al.* (1997) reported that 30 min time-trial running performance in elevated ambient conditions (32°C, 60% rh) was improved by 4% after pre-cooling via the immersion to the neck in cool water (28-29°C). Booth *et al.* (1997) used a progressive cooling method which resulted in a reduction in core temperature of ~0.7°C during the interval between cooling and exercise and no further 'afterdrop' as documented in other pre-cooling studies that used a faster pre-cooling intervention (Hessemer *et al.*, 1984; Schmidt & Bruck, 1981). The pre-cooling method adopted by Hessemer *et al.* (1984) consisted of two bouts of cool air exposure with an intermediate re-warming phase. The re-warming phase was used to try and restore thermal comfort, reduce shivering and minimise metabolic changes while still reducing core temperature. Hessemer *et al.* (1984) reported that participants achieved a 6.8% greater (172W v 161W) mean work rate following pre-cooling compared to the control trial in a 60 min cycle ergometer time-trial at 18°C despite no significant main effect between conditions for oesophageal temperature. The core temperatures recorded differed between conditions in their time course with pre-cooling initially lowering core temperature during the first 15 min by up to 0.4°C but resulting in higher core temperatures after 20 min of the protocol. Skin temperature was significantly lowered by the pre-cooling intervention (Hessemer *et al.*, 1984) and it has been shown that a reduction in skin temperature in the absence of a reduce core temperature can result in improved time-trial performance (Kay *et al.*, 1999). Kay *et al.* (1999) demonstrated that pre-cooling via water immersion improved 30 min self-paced cycling performance (14.9 ± 0.8km to 15.8 ± 0.7km) without observed reductions in core temperature while Arngrimsson *et al.* (2004) reported a 1.1% improvement in 5km time-trial performance following vest-induced pre-cooling despite the reductions in core and skin temperatures observed at the beginning of the bout disappearing for the final third (3.6km onwards). The authors noted that out of the 12 participants who improved with the vest 17.4% of the improvement occurred during the first 1.6km of the race when the effects of the cooling were most pronounced with 55.0% and 27.6% occurring in the second and third sections of the time-trial respectively. This

shows that participants were still able to increase their self-selected pace during the later stages of the time-trial when the physiological effects of the pre-cooling were either reduced or not evident supporting suggestions that perceptual cues regarding thermal state may have a pace-regulating influence.

2.3.2.6. Pre-cooling summary

Pre-cooling appears to elicit thermoregulatory and cardiovascular changes that benefit endurance performance. If the cooling stimulus is sufficient pre-cooling has been shown to regularly decrease the thermal strain experienced by lowering core temperature, attenuating its rate of rise and/or decreasing skin temperature (Arngrimsson *et al.*, 2004; Bolster *et al.*, 1999; Booth *et al.*, 1997; Booth *et al.*, 2004; Castle *et al.*, 2006; Cheung & Robinson, 2004; Drust *et al.*, 2000; Duffield *et al.*, 2003; Duffield & Marino, 2007; Gonzalez-Alonso *et al.*, 1999; Hasegawa *et al.*, 2005; Hasegawa *et al.*, 2006; Kay *et al.*, 1999; Lee & Haymes, 1995; Marino & Booth, 1998; Marino, 2002; Marsh & Sleivert, 1999; Mitchell *et al.*, 2003; Olschewski & Bruck, 1988; Palmer *et al.*, 2001; Schmidt & Bruck, 1981; Simmons *et al.*, 2008; Sleivert *et al.*, 2001; Uckert & Joch, 2007; Webster *et al.*, 2005; Wilson *et al.*, 2002). Decreasing core temperature prior to exercise and attenuating the rate at which it rises during exercise has been shown to increase the time taken to reach the high internal temperatures associated with the termination of exercise (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993) while reductions in skin temperature would be expected to improve perceptions of thermal comfort (Cotter & Taylor, 2005). The effects of pre-cooling on acute short-duration exercise is yet to be fully understood with methodological differences making comparisons between existing data sets difficult however such activities are unlikely to be compromised due to thermoregulatory strain and therefore the mechanisms by which pre-cooling may enhance short-duration exercise appear to differ markedly from those which may explain enhanced prolonged exercise performance. Prolonged exercise performance and capacity are far more likely to be compromised by thermoregulatory strain and therefore the thermoregulatory alterations that occur due to the cooling strategies adopted prior to exercise help explain the regular enhancement observed in subsequent prolonged exercise (Arngrimsson *et al.*, 2004; Booth *et al.*, 1997; Gonzalez-Alonso *et al.*, 1999; Hasegawa *et al.*, 2006; Hessemer *et al.*, 1984; Kay *et al.*, 1999; Lee & Haymes, 1995; Olschewski & Bruck, 1988; Uckert & Joch, 2007; Webster *et al.*, 2005). Pre-cooling can improve endurance performance in the absence of alterations in core temperature (Hessemer *et al.*, 1984; Kay *et al.*, 1999) and therefore it seems that it may be the enhancement of thermal comfort, due to the disruption of the

thermal feedback channels (regularly accompanied with beneficial reductions in actual thermoregulatory strain), that explains the benefits of pre-cooling.

2.3.3. Cooling the torso at rest and during exercise

2.3.3.1. Introduction

Often the pre-cooling treatments used are impractical for sporting settings due to the time required prior to exercise or the intervention used itself (e.g. water-immersion (Kay *et al.*, 1999)) and therefore a practical cooling intervention that could be adopted during exercise (during the warm-up or bout itself) and enhance performance would be of great interest to athletes, coaches and physiologists. It seems prudent to suggest that if pre-cooling the body prior to exercise can enhance performance due to the manipulation of physiological and perceptual parameters, sufficient cooling interventions applied during exercise may have similar, or indeed cumulative, effects if combined. Relatively few studies have looked at cooling the torso during exercise with one focusing on cooling the skin via skin-wetting (Bassett, Jr. *et al.*, 1987) and another five investigating the effect of wearing a cooling garment (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005; Shvartz, 1976; Webster *et al.*, 2005; Shvartz *et al.*, 1976). As with many of the pre-cooling interventions there are practical problems associated with the interventions investigated regarding torso cooling. The water-spray intervention used by Basset *et al.* (1987) and the water perfused heat-exchange cooling systems investigated by Shvartz and co-workers (1976a; 1976b) have obvious limitations that prevent their application in a sporting setting while wearing a cooling vest has been shown to increase the energy demands of running and result in some reports of participant dislike due to excess weight (~4.5 kg) and skin irritation (Arngrimsson *et al.*, 2004). The discomfort associated with such cooling devices may account for the lack of interest in this area. The effects of cooling the torso at rest and during exercise on the physiological and perceptual responses to exercise and on exercise performance and capacity are reviewed in this section. Investigations examining the effect of cooling the torso during exercise are summarised in Table 2.1.

2.3.3.2. Thermoregulatory response

2.3.3.2.1. Core temperature

As previously discussed pre-cooling the torso has been shown to lower core temperature by cooling the peripheral blood that travels to the core and cooling during exercise would

be expected to work via the same mechanism however mixed results have been reported. Cooling vests have been shown to reduce core temperature during active warm-ups lasting between 9 and 38 min (Arngrimsson *et al.*, 2004; Webster *et al.*, 2005) but not during a prolonged (~65 min) bout of submaximal exercise (Hasegawa *et al.*, 2005). Shvartz and colleagues (1976a; 1976b) used a water-perfused cooling heat exchanger to cool a small section of the chest or back and found that this reduced core temperature during bench-stepping in very hot conditions (39.5°C) however Bassett *et al.* (1987) failed to see similar results using a less intensive cooling technique, mist spray, during exercise in less sever conditions (29.5°C). These findings suggest that, as with cooling at rest, the effectiveness of the cooling intervention appears to be heavily magnitude and duration dependent. Arngrimsson *et al.* (2004) and Webster *et al.* (2005) both reported reductions in core temperature with the acute application of a cooling vest where as Hasegawa *et al.* (2005) did not see reductions in core temperature after a longer exposure. The two former studies both measured the temperature beneath the vest and found a significant reduction in skin temperature however Hasegawa *et al.* (2005) did not measure the direct cooling effect of the vest. Despite the lack of data regarding the direct effect of the cooling it would seem prudent to suggest that the effects of the cooling vest on the temperature of the area it was cooling was reduced over time. If, as assumed, the cooling vest began to warm-up during the 60 min protocol a point was probably reached during the trial in which the vest became an active insulator rather than a cooler and restricted the amount of heat that could be lost negating the earlier cooling benefits. When the vests were worn for shorter durations during the active warm-up phases of the studies by Arngrimsson *et al.* (2004) and Webster *et al.* (2005) the temperature immediately beneath the vest was maintained at a cooler temperature showing that although it may have warmed slightly in comparison to the temperature recorded at the time of application, the vest did not become an insulator during those trials.

2.3.3.2.2. Skin temperature

Unsurprisingly, due to the rationale of the cooling-intervention and the direct application of the cooling intervention, all of the interventions adopted to cool the torso during active warm-ups and exercise bouts in the heat are effective in significantly reducing skin temperatures during exercise (Arngrimsson *et al.*, 2004; Bassett, Jr. *et al.*, 1987; Hasegawa *et al.*, 2005; Shvartz, 1976; Shvartz *et al.*, 1976; Webster *et al.*, 2005). Mean skin temperature is usually expressed as a weighted skin temperature and therefore care needs to be taken when interpreting the data. Arngrimsson *et al.* (2004) and Hasegawa *et al.*

(2005) both reported a reduction in mean skin temperature using a skin weighting system biased towards the torso (0.5 and 0.3 respectively) and so may have overestimated the magnitude of changes due to direct cooling from the vests however the other investigators used formulae that were not torso-biased and all reported reductions in skin temperature (Bassett, Jr. *et al.*, 1987; Shvartz, 1976; Shvartz *et al.*, 1976; Webster *et al.*, 2005). Webster *et al.* (2005) recognised the potential to overestimate the cooling effect using conventional skin temperature assessment techniques and therefore reported the temperature measured at the abdomen and bicep to represent the temperatures of the skin in regions covered and uncovered by the vest. There was a marked decline in skin temperature measured at the abdomen and a lesser, but still significant, reduction in skin temperature measured at the bicep. This data, in addition to that reported in other investigations, shows that torso cooling via the application of a cooling vest can lower the skin temperature in areas uncovered as well as in those directly cooled.

2.3.3.3. Cardiovascular response

Despite consistently reducing skin temperature (Arngrimsson *et al.*, 2004; Bassett, Jr. *et al.*, 1987; Hasegawa *et al.*, 2005; Shvartz, 1976; Shvartz *et al.*, 1976; Webster *et al.*, 2005), and therefore presumably skin blood flow, the effects of torso cooling during exercise on the heart rate response to exercise are mixed with some studies reporting cooling-induced bradycardia (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005; Shvartz *et al.*, 1976) and some reporting no effect (Bassett, Jr. *et al.*, 1987; Shvartz, 1976; Webster *et al.*, 2005). The findings of Basset *et al.* (1987) and Shvartz (1976), who cooled via mist spaying and a small chest heat exchange device respectively, suggest that the effectiveness of cooling during exercise on reducing cardiovascular strain is once more magnitude-dependent, however Webster *et al.* (2005) also reported no effect of cooling despite investigating three highly effective cooling vests. They acknowledged that their findings differed from a number of published articles and attributed the difference to other studies using cooling during longer, submaximal bouts rather than during the shorter maximal oxygen consumptions test that they investigated. In the torso-cooling studies that showed a reduction in the heart rate response to exercise the cooling was administered for 38 to 120 min during submaximal exercise (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005; Shvartz *et al.*, 1976) supporting the suggestion of Webster *et al.* (2005). Drust *et al.* (2000) suggested that if cooling was sufficient to result in a decreased need to dissipate heat there would be an associated reduction in the need for cutaneous blood flow which in turn would reduce the need to elevate heart rate to maintain cardiac output. During exercise however,

cooling-induced reductions in heart rate do not appear to be dependent on reductions in core or skin temperature as Shvartz (1976) and Webster *et al.* (2005) both reported significant reductions in core and skin temperature without any change in the heart rate response to exercise. Although the literature is not conclusive it appears that cooling the torso during exercise can reduce the cardiovascular strain experienced by reducing the heart rate during exercise if the cooling intervention is worn during a prolonged period (> 38 min) of submaximal exercise.

2.3.3.4. Body fluid regulation

Arngrimsson *et al.* (2004), Hasegawa *et al.* (2005) and Webster *et al.* (2005) all reported that wearing a cooling vest during exercise reduced the sweat rate and total sweat loss observed however these findings were not replicated by Bassset *et al.* (1987) or Shvartz and colleagues (1976). This data suggests that, as with other physiological responses, the effectiveness of the cooling intervention used dictates the magnitude of the physiological response evoked. The reductions in the sweat response following the application of a cooling vest would be expected due to the cooling-induced disruption of heat loss mechanisms (Lee & Haymes, 1995) and are in line with the majority of pre-cooling studies (Duffield & Marino, 2007; Hasegawa *et al.*, 2006; Kay *et al.*, 1999; Lee & Haymes, 1995; Wilson *et al.*, 2002) supporting the idea that cooling interventions may be effective in the conservation of body water during exercise.

2.3.3.5. Rating of perceived exertion and thermal comfort

The subjective sensation of task difficulty as represented by the rating of perceived exertion can be reduced via the application of a cooling vest (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005) but not by mist spraying (Bassett, Jr. *et al.*, 1987) during exercise in a hot environment. It has been suggested that rating of perceived exertion is a key component of the regulatory system during exercise and plays a role in the termination of exercise (Crewe *et al.*, 2008). If this is true it would be prudent to suggest that disrupting the perception of task difficulty (e.g. via a cooling vest) and altering the rating of perceived exertion at any given point would have beneficial performance implications. This may also apply to alterations in thermal comfort or sensation and although cooling is ineffective when applied prior to exercise (Castle *et al.*, 2006), Arngrimsson *et al.* (2004), Hasegawa *et al.* (2005) and Webster *et al.* (2005) reported that wearing a cooling vest significantly improved the perception of thermal comfort during exercise. In the study by Castle *et al.*

(2006) participants wore a vest while at rest so it is likely that the exercise-generated heat counteracted the potentially noxious cooling stimulus and allowed the participants to gain a benefit from the vest during the exercise studies.

2.3.3.6. Exercise performance

To the author's knowledge Shvartz *et al.* (1976) is the only study to have looked at the effects of cooling the torso during exercise on performance although it has been shown to improve subsequent performance (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005; Webster *et al.*, 2005). Shvartz *et al.* (1976) investigated the effect of cooling ~10% of the total body surface using a heat exchanger placed on the back of the participants during submaximal bench stepping in very hot conditions (49.3°C). They reported that the mean tolerance time was 45 min in the no cooling trial but that the participants completed the entire 70 min protocol 'without any signs of exhaustion' in the cooling trial. The study only included three individuals rendering statistical interpretation redundant however this small study suggests that cooling during exercise can have positive effects upon exercise performance. The improvement in tolerance time was accompanied by a reduction in core and skin temperature and it seems prudent to suggest that if a cooling intervention worn during exercise can elicit a sufficient cooling stimulus, as with pre-cooling, and elicit similar beneficial thermoregulatory and cardiovascular changes then these findings could be replicated. This is an area that warrants further investigation because although wearing such garments during exercise may be impractical or in breach of the rules and regulations of the sporting event they may have a role to play in maximising the gain from training sessions or out-of-competition events.

2.3.3.7. Cooling the torso summary

The physiological, perceptual and performance data presented from studies which have investigated the effects of cooling the torso during exercise demonstrates that the effect that cooling the torso can elicit is heavily magnitude dependent. When the cooling is sufficient it can lead to reductions in the temperatures recorded at the core and the skin, attenuated heart rate and improvements in the perception of task difficulty and thermal comfort (Arngrimsson *et al.*, 2004; Hasegawa *et al.*, 2005; Shvartz, 1976; Webster *et al.*, 2005) however many cooling interventions have a mixed effect and appear to offer an insufficiently prolonged cooling effect (Bassett, Jr. *et al.*, 1987; Hasegawa *et al.*, 2005; Shvartz, 1976; Webster *et al.*, 2005). Although cooling vests are often touted as practical

cooling devices and they are used by many elite sporting teams (Martin *et al.*, 1998) data from the studies reviewed suggest that more work is required before cooling the torso during exercise can be recommended as an effective, practical intervention for the improvement of performance in a hot environment.

Table 2.1. Cooling the torso during exercise

Ref	Participants	Protocol	Cooling intervention	Area Cooled	Perf	Core temp	Heart rate	RPE	TS	Sweat rate	Skin temp
Arngrimsson *et al.*, (2004)	N = 17 (9 male; 8 female)	38-min active warm-up (prior to 5km TT without) T_{amb} = 32°C; 50% rh	Torso (ice jacket)	↓*	-	↓*	↓*	↓*	↓*	-	↓*
Bassett *et al.*, (1987)	N = 10 male	2-hr TR @ 60%VO_{2max} T_{amb} = 29.5°C; 33% rh	Mist spray [50ml H_2O.10 min^{-1}]	-	-	↔	↔	↔	-	↔	↓*
Bassett *et al.*, (1987)	N = 10 male	2-hr TR @ 60%VO_{2max} T_{amb} = 29.5°C; 66% rh	Mist spray [50ml H_2O.10 min^{-1}]	-	-	↔	↔	↔	-	↔	↓*
Hasegawa et *al.*, (2005)	N = 9 untrained males	60min CE @ 60% (prior to T_{exh} @ 80%VO_{2max} without) T_{amb} = 32°C; 75% rh	Torso (ice jacket)	-	-	↔	↓*	↔	↔	↓*	↓*
Shvartz (1976)	N = 6 males	Bench-stepping at 40W T_{amb} = 39.5; ~51% rh	Chest cooling via water perfused system [64.5 $l.hr^{-1}$; 8.3°C]	-	-	↓*	↔	-	-	↔	↓
Shvartz *et al.*, (1976)	N = 3 male	70-min bench-stepping T_{amb} = 49.3°C	Back cooling via water-perfused garment [11.2°C]	-	↑	↓	↓	-	-	↔	↓

↑ increase; ↓ decrease; ↔ no change; - = not measured; * P=<0.05; **P=<0.01; ***P<0.001; CE = cycle ergometer; TR = treadmill; TT = time-trial; T_{exh} = test to exhaustion; T_{amb} = ambient temperature; perf = performance; temp = temperature. N.B. Data from Arngrimsson *et al.*, (2004) and Webster *et al.*, (2005) refers to the effect of the cooling intervention during an active warm-up and not subsequent exercise.

Table 2.1 (continued). Cooling the torso during exercise

Ref	Participants	Protocol	Cooling intervention	Area Cooled	Perf	Core temp	Heart rate	RPE	TS	Sweat rate	Skin temp
Webster *et al.*, (2005)	N = 16 (8 males; 8 females)	14-min warm-up (stretching 4-min; TR @ 50% VO_{2max} 9-min) T_{amb} = 37°C ; 50% rh	Torso (ice vest A- long, light-weight sport-specific close fitting garment)	↓**	-	↓*	↔	-	↓**	-	↓*
Webster *et al.*, (2005)	N = 16 (8 males ; 8 females)	14-min warm-up (stretching 4-min; TR @ 50% VO_{2max} 9-min) T_{amb} = 37°C ; 50% rh	Torso (ice vest B- short, impermeable, sport-specific close fitting garment)	↓**	-	↓*	↔	-	↓**	-	↓*
Webster *et al.*, (2005)	N = 16 (8 males ; 8 females)	14-min warm-up (stretching 4-min; TR @ 50% VO_{2max} 9-min) T_{amb} = 37°C ; 50% rh	Torso (ice vest C- industrial cooling garment)	↓**	-	↓*	↔	-	↓**	-	↓*

↑ increase; ↓ decrease; ↔ no change; - = not measured; * P=<0.05; **P=<0.01; ***P<0.001; CE = cycle ergometer; TR = treadmill; TT = time-trial; T_{exh} = test to exhaustion; T_{amb} = ambient temperature; perf = performance; temp = temperature. N.B. Data from Arngrimsson *et al.*, (2004) and Webster *et al.*, (2005) refers to the effect of the cooling intervention during an active warm-up and not subsequent exercise.

2.3.4. Cooling the head/face/neck region

2.3.4.1. Introduction

It has been suggested that thermal sensation and/or comfort may be determined in part by regional variation in the thermosensitivity of the skin surface and that this comfort could be manipulated with local heating or cooling (Cheung, 2007). The head and face have been shown to be sites of high alliesthesial thermosensitivity (Cotter & Taylor, 2005) and it has been suggested that cooling the head may represent a greater thermoregulatory advantage compared to cooling other parts of the body (Kissen *et al.*, 1971; McCaffrey *et al.*, 1975; Nunneley *et al.*, 1971; Shvartz, 1970). Despite this, and the fact that pre-cooling studies can be impractical (Wilson *et al.*, 2002), studies that have investigated the effect of cooling the head and neck region during periods of rest and exercise over the last 40 years are scarce.

Despite the scarcity of studies a wide variety of cooling methods have been used to lower the temperature of the head, face and/or neck during periods of rest and exercise. The methods fall into three main categories; the application of water-perfused garments (Greenleaf *et al.*, 1980; Nunneley *et al.*, 1971; Palmer *et al.*, 2001; Shvartz, 1970; Shvartz, 1976; Shvartz *et al.*, 1976; Watanuki, 1993); the supply of convectional air currents (with or without water mist spraying) (Ansley *et al.*, 2007; Armada-da-Silva *et al.*, 2004; Brisson *et al.*, 1987; Brisson *et al.*, 1989; Brisson *et al.*, 1991; Desruelle & Candas, 2000; Hamada *et al.*, 2006; Kratzing & Cross, 1984; Mundel *et al.*, 2005; Mundel *et al.*, 2006; Mundel *et al.*, 2007; Nybo *et al.*, 2002a; Quirion *et al.*, 1989; Riggs, Jr. *et al.*, 1981; Riggs, Jr. *et al.*, 1983; Stroud, 1991; Williams & Kilgour, 1993) and the direct application of ice packs or a specific neck cooling device (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990; Hamada *et al.*, 2006; Simmons *et al.*, 2008). The effects of the various interventions are reviewed in this section and practical neck cooling investigations are summarised in table 2.2.

2.3.4.2. Thermoregulatory response

2.3.4.2.1. Core temperature

The effectiveness of cooling the head, face or neck region on altering core temperature appears to heavily depend on both the extent of the cooling stimulus provided and the thermal stress experienced. Cooling the head and neck region via facial fanning or cool air (2°C - 27°C) provides an insufficient cooling stimulus to alter deep body temperature at

rest (Brisson *et al.*, 1991; Desruelle & Candas, 2000; Kissen *et al.*, 1971; Nunneley *et al.*, 1971) or during exercise (Ansley *et al.*, 2007; Armada-da-Silva *et al.*, 2004; Brisson *et al.*, 1991; Desruelle & Candas, 2000; Hamada *et al.*, 2006; Mundel *et al.*, 2006; Riggs, Jr. *et al.*, 1981; Williams & Kilgour, 1993) in a wide range of ambient conditions (22°C – 66°C). The same findings are reported when cooling is achieved via water-perfused cooling garments at rest (Brown & Williams, 1982; Greenleaf *et al.*, 1980; Kissen *et al.*, 1971; Nunneley *et al.*, 1971) however during exercise cooling the head and neck region has been shown to reduce core temperature in most (Nunneley *et al.*, 1971; Palmer *et al.*, 1996; Shvartz, 1970; Shvartz, 1976; Shvartz *et al.*, 1976), but not all (Nunneley *et al.*, 1971), studies. Nunneley *et al.* (1971) investigated the effect of cooling the head via a water-perfused cap (in which progressively cooler water (22°C to 10°C) was circulated at a rate of 720 $ml \cdot min^{-1}$ during exercise for the 120 min protocol) on the physiological response to exercise at ~50% $\dot{V}O_{2max}$ in three different ambient temperatures, 20°C, 30°C and 40°C. They reported that cooling the head had no effect on core temperature in the trials conducted at 20°C and 30°C however it reduced core temperature in the 40°C trials when the thermal stress was greatest. In the other studies that reported a reduction in core temperature with the water-perfused head/neck cooling garment either the exercise intensity (60% $\dot{V}O_{2max}$ (Palmer *et al.*, 1996)) or ambient conditions (39.5°C (Shvartz, 1976); 50°C (Shvartz, 1970)) were of a greater magnitude than in the two conditions in the study by Nunneley *et al.* (1971) in which no effect was observed. This provides further support for the suggestion that cooling the head and neck region is only effective at reducing core temperature when the cooling stimulus is of a sufficient magnitude and in conditions during which the body is under sufficient thermal stress.

In the four studies investigating direct neck cooling via the application of ice-packs or specific cooling collar devices only Gordon *et al.* (1990) and Simmons *et al.* (2008) reported changes in core temperature as a result of the interventions investigated. Simmons *et al.* (2008) observed that the application of cooling packs, combined with water mist cooling, during a period of prolonged (~85 min) resting sauna exposure between two bouts of moderate intensity cycle ergometer exercise (~70% $\dot{V}O_{2max}$) increased the rate at which core temperature dropped and allowed the participants to commence the second bout of exercise with a significantly lower core temperature. Gordon *et al.* (1990) reported that core temperature and the rate at which it rose was attenuated with a neck cooling device during 45 min of submaximal track-running in moderate ambient conditions (21°C) however neither Bulbulian *et al.* (1999) nor Hamada *et al.* (2006) replicated these findings. Bulbulian *et al.* (1999) found no differences in the core temperature (measured at both the

rectum and oesophagus) response to 30 min of cycle ergometer exercise at 60% $\dot{V}O_{2max}$ in hot ambient conditions (30°C; 25% rh) with the application of a commercially available head-neck cooling device despite significantly lowering the temperature of the neck ($P < 0.01$). Hamada *et al.* (2006) cooled the neck to 23.59 ± 2.18°C via the application of ice packs placed superior to the bilateral carotid arteries during the last 20 min of a 40 min bout of submaximal (60% $\dot{V}O_{2max}$) cycle ergometer. The ice packs were replaced at 10 min intervals in attempt to maximise the cooling stimulus achieved however despite maintaining a strong cooling stimulus they found that their intervention had no effect on core temperature.

Most of the literature suggests that in contrast to whole-body cooling, cooling the head and neck region has no effect on core temperature, particularly in temperatures below those that are approaching a "very hot" classification (>35°C). This is particularly true for the more practical, non-water pump based, methods of achieving the cooling. The proposed mechanism by which whole-body cooling is thought to work relies on the cooling of the blood in the periphery and the subsequent circulation of this cooler blood to the core (Schmidt & Bruck, 1981) and therefore the ineffectiveness of cooling the neck in altering core temperature is unsurprising. When cooling larger areas of the body there is a greater potential for the cooling of large volumes of peripheral blood resulting in a larger amount of cooled blood travelling to the centre of the body. The neck region only accounts for approximately ~12% of the total body surface area and thus the potential for cooling a sufficient volume of the blood that penetrates the body's core is greatly reduced.

2.3.4.2.2. Skin temperature

As discussed in most cases cooling the neck fails to alter the core temperature response to exercise-induced or passive hyperthermia due to the cooling of an insufficient surface area of the body. Due to this and the multi-site techniques used to measure skin temperature cooling the neck would be unlikely to alter skin temperature. The majority of studies have shown that cooling the head and neck region has no effect on skin temperature at rest or during exercise regardless of whether the cooling was achieved via water-perfused garments, cool air or neck cooling devices (Armada-da-Silva *et al.*, 2004; Brown & Williams, 1982; Bulbulian *et al.*, 1999; Desruelle & Candas, 2000; Hamada *et al.*, 2006; Mundel *et al.*, 2006; Nunneley *et al.*, 1971; Shvartz, 1976; Simmons *et al.*, 2008) however some studies have reported reductions in skin temperature (Ansley *et al.*, 2007; Hamada *et al.*, 2006; Kissen *et al.*, 1971; Shvartz, 1970). Kissen *et al.* (1971) reported that head and

neck cooling via a liquid-cooled helmet reduced skin temperature while individuals rested in very high temperatures (66°C) and in similarly hot conditions (50°C) Shvartz (1976) reported that cooling the head and neck via a water-perfused hood reduced skin temperature during exercise-induced hyperthermia created by prolonged (120 min) walking. It is worth noting that the studies by Kissen *et al.* (1971) and Shvartz (1976) only involved six participants each and that no statistical analysis was conducted by the authors. Visual representation of the data shows that there was a tendency for the cooling interventions used to reduce skin temperature however with the lack of statistical analysis full, accurate interpretation regarding the extent of the reduction is difficult to quantify. Hamada *et al.* (2006) compared three different methods of cooling and reported that although cooling the neck via the application of a cooling collar had no effect on skin temperature cooling the head via facial fanning significantly reduced skin temperature during 40 min of submaximal cycling (60% $\dot{V}O_{2max}$) at 30°C. Ansley *et al.* (2007) also reported that facial cooling lowered skin temperature during cycle ergometer exercise to exhaustion, in ambient conditions of ~30°C. Hamada *et al.* (2006) reported that the reduction in skin temperature observed coincided with a cooling-induced reduction in skin blood flow. The reduction in skin blood flow may help account for the reduction in skin temperature because the internal heat accumulating within the body due to the elevations in body temperature (~1°C in the study by Hamada *et al.* (2006)) would usually be directed, via the circulating blood, to the skin to be dissipated. The cooling intervention may have altered the thermoregulatory effector responses due to the presentation of a "false", misleading, thermoregulatory input, reducing blood flow to the skin and reducing the amount of heat brought to the periphery. It is also possible that the differences in findings could be attributed to an increase in convectional cooling via inadvertent cooling of other areas via the fanning protocols. Ansley *et al.* (2007) stated that the fans used were directed at the face to "only cool the face" however within the confined space of an environmental chamber it seems likely that the introduction of moving air would increase the total amount of air current within the chamber which could help explain some of the decreases in skin temperatures observed in the studies by Ansley *et al.* (2007) and Hamada *et al.* (2006).

2.3.4.3. Cardiovascular response

It appears that cooling the head and neck region has a similar effect on heart rate as it does on skin temperature. Despite a variety of cooling methods being implemented in a variety of different hot environmental conditions the majority of studies have shown that cooling the head and neck has no effect on heart rate during exercise (Ansley *et al.*, 2007;

Bulbulian *et al.*, 1999; Gordon *et al.*, 1990; Hamada *et al.*, 2006; Kratzing & Cross, 1984; Nunneley *et al.*, 1971; Palmer *et al.*, 1996; Shvartz *et al.*, 1976; Simmons *et al.*, 2008). However, as observed with the skin temperature response to cooling, cooling of a sufficient magnitude can attenuate the elevation in heart rate observed in hot conditions. Shvartz (1970), Hamada *et al*, (2006) and Mundel *et al.* (2006) all reported that cooling the head, neck and face induced bradycardia during submaximal exercise in hot conditions (30-50°C). Heistad *et al.* (1973) and Yamazaki and Sone (2000) have both demonstrated that during hyperthermia heart rate is controlled at a central level by an interaction between the thermoreceptor and baroreceptor reflexes as demonstrated by the independent nature of core and skin temperatures in heart rate regulation. Yamazaki and Sone (2000) proposed that a decrease in heart rate was due to an increase in vagal activity occurring due to the location of the vagus nerves and their proximity to the cooling stimulus. The cardiac parasympathetic fibres originate in the medulla oblongata and the efferent vagal nerves pass inferiorly through the neck lying adjacent to the carotid arteries and therefore it is has been proposed that cooling the neck regions could lower heart rate by direct vagal stimulation (Yamazaki & Sone, 2000).

2.3.4.4. Body fluid regulation

Cooling the head and neck region has been shown to decrease the sweat rate observed during exercise in a hot environment in some (Desruelle & Candas, 2000; Gordon *et al.*, 1990; Hamada *et al.*, 2006; Nunneley *et al.*, 1971; Shvartz, 1970; Shvartz, 1976) but not all (Bulbulian *et al.*, 1999; Desruelle & Candas, 2000; Mundel *et al.*, 2006; Palmer *et al.*, 1996) studies. Desruelle and Candas (2000) investigated three variations of cool air breathing and facial fanning and reported that only a combination of both- and therefore the greatest cooling stimulus investigated- attenuated the sweat rate observed. Further evidence to suggest that the effect of a cooling intervention of the sweat response is dependent on the magnitude of the cooling stimulus and thermal stress is provided by the observations that at rest the sweat response is unaltered by head, neck or face cooling (Brown & Williams, 1982; Desruelle & Candas, 2000) unless the thermal stress is of a severe magnitude (ambient temperature of 66°C (Kissen *et al.*, 1971)). Cooling the head neck and throat via cooling hood has also been shown to significantly reduce voluntary water intake during 2h of walking in very high ambient conditions (~50°C) (Shvartz, 1970) and so it appears that cooling this region may disrupt the feedback systems responsible for sweat response initiation and the thirst response when the thermal stress is severe. The sweat response is initiated due to signals from both peripheral and internal thermal

receptors with brain temperature being the main regulating factor (Smiles *et al.*, 1976). It is possible that any 'false' signal, detected via the thermoregulatory centre within the brain, provided about the thermal status of the body as a result of a the cooling intervention, either directly via the cooling of carotid blood or indirectly via neural signal, may cause the drive to dissipate heat to be reduced.

2.3.4.5. Rating of perceived exertion and thermal comfort

Participants who have their head, neck and/or face region cooled often report an enhanced level of thermal comfort (Brown & Williams, 1982; Hamada *et al.*, 2006; Kissen *et al.*, 1971; Mundel *et al.*, 2006; Palmer *et al.*, 1996; Simmons *et al.*, 2008; Armada-da-Silva *et al.*, 2004) however two studies have shown that cooling this region had no effect on thermal sensation (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). Bulbulian *et al.* (1999) and Gordon *et al.* (1990) both investigated commercially available cooling collars and it is possible that the ineffectiveness of the collars on the perception of thermal comfort is due to the provision of an inadequate level of cooling. To gain the maximal benefit from a cooling intervention it seems clear that the cooling stimulus must offer a prolonged bout of sustained cooling however Bulbulian *et al.* (1999) reported that their cooling collar only remained cool for ~10 min of their 30 min submaximal (60% $\dot{V}O_{2max}$) bout of cycle ergometer exercise in elevated temperatures (30°C; 25% rh). Gordon *et al.* (1990) did not report the localised effect of the collar tested however as demonstrated by Bulbulian *et al.* (1999) and the in-house collar design work (see general methods section) commercially available collars tend to provide a limited cooling stimulus.

Despite the majority of studies reporting an improvement in the perception of thermal comfort, cooling the head, neck and face region has been shown to have a mixed effect on the rating of perceived exertion. Cooling has been shown to both, reduce (Ansley *et al.*, 2007; Mundel *et al.*, 2006; Simmons *et al.*, 2008; Armada-da-Silva *et al.*, 2004) and have no effect (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990) on the rating of perceived exertion reported during exercise. Simmons *et al.* (2008) reported that cooling the head via a combination of ice-pack application and water spraying during a prolonged (~68 min) bout of sauna exposure sandwiched between two 12 min bouts of moderate exercise performed in a hot environment reduced the rating of perceived exertion during the second bout of exercise. They also showed that the cooling intervention attenuated the rate at which core temperature rose and reported a significant correlation ($r = 0.82$; $P < 0.001$) between the rating of perceived exertion and core temperature providing further support for the

suggestion that the increase in the rating of perceived exertion during exercise in hot conditions is related to the increase in core temperature (Crewe *et al.*, 2008). Ansley *et al.* (2007) and Armada-da-Silva *et al.* (2004) both suggested that the rating of perceived exertion is heavily influenced by the subjective level of thermal comfort and the review of the literature reveals that when thermal sensation is enhanced the rating of perceived exertion reacts similarly. In contrast when the cooling intervention is ineffective at improving thermal comfort the rating of perceived exertion is unaffected (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). As with the physiological response to cooling the effect of cooling on the subjective perception of exertion appears to be magnitude dependent. If sufficient cooling can be provided to alter the perception of thermal comfort then it seems that the rating of perceived exertion can be reduced which would be expected to delay the termination of exercise (Crewe *et al.*, 2008).

2.3.4.6. Exercise performance

To the author's knowledge only two studies have investigated the effect of cooling the head and neck region on exercise capacity or performance. Ansley *et al.* (2007) investigated the effects of head cooling via three fans placed 50cm away from the cyclist's face in combination with a fine mist of water administered at 30-second intervals. They reported that such a cooling intervention resulted in median cycling capacity being enhanced by 51% (21-65% inter-quartile range). The improvement was achieved without alterations in core temperature or heart rate but with reductions in skin temperature and ratings of perceived exertion. The authors postulated that the improvements may be due to improvements in perceived exertion and a thermoregulatory mediated suppression in the prolactin response to exercise. Palmer *et al.* (2001) looked at the effect of cooling the head via a water-perfused garment inducing the cooling via the circulation of 1°C water at a rate of 1.1L.min^{-1}. The participants rested for 60 min before completing 30 min of submaximal (60% $\dot{V}O_{2max}$) treadmill exercise prior to a 15 min time-trial performed in an environmental chamber regulated at 33°C. On separate visits to the lab the cooling garment was either worn during the combination of rest and exercise, just at rest or just during the bout of exercise. Full-time cooling (rest and exercise) improved the distance covered in the 15 min compared to no cooling and pre-cooling by 3.3 ± 3.4% and 2.3 ± 2.9% respectively. The cooling intervention at rest reduced core temperature and heart rate prior to exercise and when continued, or added, during exercise maintained a reduction in core temperature during the bout and improved thermal comfort. This limited data demonstrates

that water-perfused cooling and facial fanning can both enhance the ability to perform exercise in hot environments.

2.3.4.7. Cooling the head/neck/face region summary

Many of the thermoregulatory and cardiovascular benefits associated with pre-cooling and torso cooling during exercise occur due to the large surface area cooled which in turns increases the magnitude of the response that it can invoke. This helps to explain why the benefits of cooling the head, neck and face region are heavily dependent on the magnitude of the cooling intervention and the severity of the thermal strain that the body experiences. It has been shown that if the magnitude of cooling provided is sufficient and/or the thermal strain is severe enough cooling the head, neck and face region can elicit beneficial thermoregulatory and cardiovascular adjustments (Nunneley *et al.*, 1971; Shvartz, 1970; Simmons *et al.*, 2008) however practical cooling mechanisms rarely provide sufficient cooling (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). Cooling the body at any site appears to alter the perception of thermal comfort and this is the case when cooling the neck however the benefits of cooling on thermal comfort (Armada-da-Silva *et al.*, 2004; Brown & Williams, 1982; Hamada *et al.*, 2006; Kissen *et al.*, 1971; Mundel *et al.*, 2005; Palmer *et al.*, 2001; Simmons *et al.*, 2008) are not always matched with alterations in the participants' perception of the task difficulty as expressed via the rating of perceived exertion scale (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). The effects of practically cooling the head and neck on exercise performance are unclear as few studies have investigated this and mixed results have been presented. Based upon the data from other cooling studies in circumstances where the cooling stimulus and the thermal stress are of a (as yet un-known) sufficient magnitude the beneficial physiological changes would be expected to have an ergogenic effect however this is an area that requires further investigation.

Table 2.2. Cooling the neck region during exercise via neck cooling collar/device

Ref	Participants	Protocol	Cooling intervention	Area Cooled	Perf	Core temp	Heart rate	RPE	TS	Sweat rate	Skin Temp
Bulbulian *et al.*, (1999)	N = 20 (10 males; 10 females)	30-min CE @ 60%VO_{2max} T_{amb} = 30°C; 25% rh	Head and neck via cooling collar	↓**	-	↔	↔	↔	↔	↔	↔
Gordon *et al.*, (1990)	N = 10 male	45-min submaximal run T_{amb} = 21°C	Neck via cooling collar	-	-	↓* end	↔	↔	↔	↓*	-
Hamada *et al.*, (2006)	N = 7 male	40-min rest followed by ~40-min CE @ 60%VO_{2max} T_{amb} = 30°C	Neck cooling via ice pack application for last 20-min of exercise	↓*	-	↔	↔	-	↓*	↓*	↔

↑ increase; ↓ decrease; ↔ no change; NS = non-significant; * P=<0.05; **P=<0.01; ***P<0.001; CE = cycle ergometer; Perf= performance; temp = temperature; RPE = rating of perceived exertion; TS = thermal sensation; T_{amb} = ambient temperature; CE = cycle ergometer

2.4. Theoretical basis for the application of a cooling collar

2.4.1. Introduction

Hyperthermia impairs exercise capacity and performance (e.g. Galloway and Maughan, 1997) and, as reviewed in previous sections, a variety of cooling interventions have been investigated in an attempt to attenuate the decline in exercise ability observed. Pre-cooling and cooling the torso during exercise have both been shown to elicit physiological and perceptual adjustments which benefit exercise performance in a hot environment (e.g. Arngrimsson *et al.*, 2004). Cooling the neck region has received substantially less interest however when there is a sufficient gradient between the cooling and the environmental conditions cooling this area can evoke similar physiological and perceptual adjustments (e.g. Nunneley *et al.*, 1971) and improve performance (Palmer *et al.*, 2001). Researchers that have investigated cooling this region have suggested that cooling the neck offers many theoretical advantages over cooling elsewhere and these theories are reviewed in the following section.

2.4.2. Selective brain cooling in animals

The brain is one of the most metabolically active organs and is thus heavily dependent on the constant removal of the excess metabolically-produced heat to maintain thermal balance (Caputa, 2004). This is particularly important as it has been shown that neuronal tissue is particularly susceptible to overheating. Protein synthesis within the brain has been shown to be inhibited in rats when cerebral temperatures reach 41°C due to the disaggregation of polyribosomes to monosomes however the reduction in synthesis is not observed in other tissues (e.g. kidney, liver, spleen and testes) subjected to similar thermal conditions (Millan *et al.*, 1979). Due to the vulnerability of the cerebral tissue it would seem prudent for there to be a protective mechanism in place to protect the brain from the threat of hyperthermia and in many species such a mechanism exists.

It has been demonstrated that the temperatures of the cerebral arteries and brain can fall below central arterial blood temperature when upper respiratory heat loss is enhanced in a wide-range of species including cats (Baker, 1972), goats (Baker & Nijland, 1993), sheep (Baker & Hayward, 1968), dogs (Baker *et al.*, 1974), kestrels (Bernstein *et al.*, 1979) and horses (McConaghy *et al.*, 1995). The magnitude of the cooling response is proportional to the level of hyperthermia experienced and thus the response elicited is greater during

exercise than at rest (Bernstein *et al.*, 1979). In normothermic animals during walking it occurs weakly and intermittently whereas selective brain cooling occurs continuously and at a far greater intensity during hyperthermia (Caputa, 2004). To demonstrate this it has been shown that the core-brain temperature difference increases from 0.7°C at rest to 1.2°C during exercise in the American Kestrel (Bernstein *et al.*, 1979) and that during exercise the magnitude of the selective brain cooling is enhanced proportionally in goats (Baker & Nijland, 1993) (Figure 2.8). The lowering of the temperature of the brain below that of the trunk has been termed 'selective brain cooling'. The existence of selective brain cooling in a number of animal species is now widely accepted (Baker, 1979) and it has been shown to be a method employed by many species which pant and possess a carotid rete (Baker, 1982).

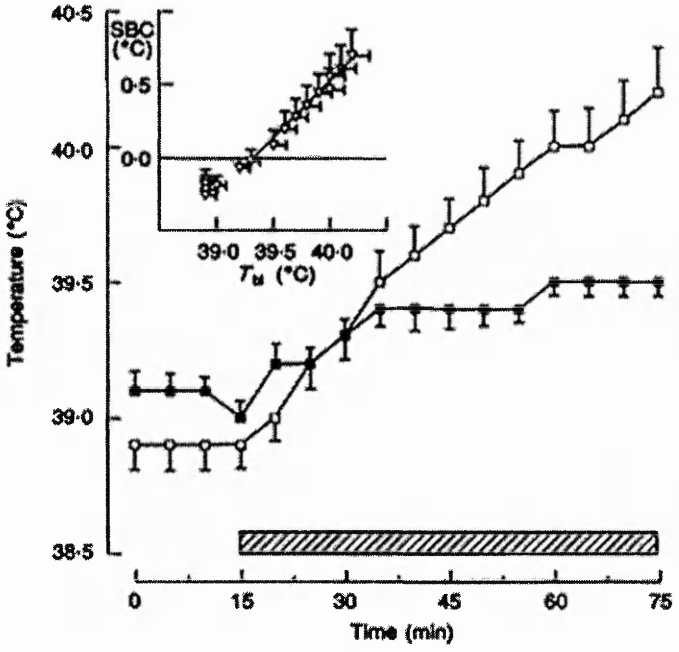

Figure 2.8. Central blood temperature (open squares) and brain temperature (filled squares) of 6 goats at rest and during exercise. Insert, selective brain cooling as a function of central blood temperature [reproduced from Baker and Nijland, (1993) with permission]

Selective brain cooling during hyperthermia has been shown to occur in a variety of species including dogs, antelopes, sheep, reindeer, gazelles, rabbits and pigeons (Baker, 1982; Johnsen *et al.*, 1987). The existence of the selective brain cooling mechanism allows these species to tolerate extremely high temperatures which would otherwise be lethal. Grant's gazelles deprived of water and in hyperthermic conditions are able to tolerate body temperatures of 46.5°C for durations of up to 6h due to the maintenance of a brain temperature below the temperature of the torso due to a highly efficient selective brain cooling mechanism (Taylor, 1970). The selective brain cooling mechanism that exists in most species involves the lowering of the temperature of the venous blood via the cool air

introduced by the act of panting (Johnsen *et al.*, 1987) and the movement of this cooled blood to the intracranium from the surface of the nasal cavity which cools the incoming arterial blood. It is thought that most of this cooling occurs within the carotid rete. The carotid rete is a network of medium-sized arteries which lay in a lake of venous blood in either the cavernous sinus or in the pterygoid plexus. Due to the position of the carotid rete network and the fact that the arterial and venous blood are only separated by the relatively thin arterial and venous walls there is a large potential for counter-current heat exchange (also called arteriovenous heat transfer (Nunneley & Nelson, 1994)) to occur. It has been proposed that this counter-current heat exchange mechanism can enhance the thermal tolerance of many species by reducing the temperature of the brain (Baker, 1982; Hayward & Baker, 1969). The temperature of the mammalian brain is essentially determined by three main factors; the rate of heat production of the brain; the blood flow through the brain and the temperature of blood supplying the brain (Baker, 1982; Kiyatkin, 2007). Kiyatkin *et al.* (2007) reported that the changes in the brain temperature of rats following stimulation (in this case social interaction and tail pinching) were more rapidly occurring and pronounced than the changes observed in the temperature of arterial blood suggesting that the temperature of the brain may also be compromised by intra-brain heat production as well as the heat delivered via the circulatory system.

Despite the contribution of intra-heat production it has consistently been shown that temperature changes in arterial blood result in almost immediate changes in temperature throughout the brain and that the temperature of this blood is the most variable and influential factor concerning the changes in brain temperature (Baker, 1972; Baker *et al.*, 1974; Baker, 1982; Hayward, 1968; Hayward & Baker, 1969). This data forms the premise of selective brain cooling and has led to the proposal that lowering the temperature of the blood that reaches the human brain would offer the same benefits as observed in animal species (Cabanac & Caputa, 1979).

2.4.3. Selective brain cooling in humans?

Humans do not possess a carotid rete (Cabanac, 1986) and therefore the potential for cooling the blood on route to the brain is reduced however it has been shown that the presence of a carotid rete is not a prerequisite for intracranial heat exchange as it is a feature absent in the rabbit, a species that has been shown to utilise selective brain cooling as a method of lowering the temperature of the brain (Cabanac, 1986). Blood is supplied to the human brain via the right and left common carotid arteries which split into internal and

external carotid arteries at the upper level of the larynx (Figure 2.9) (Gray, 1918) while the right and left internal jugular veins receive the blood drained from the face, neck and the sinuses such as the superior and inferior sagittal sinuses, straight sinus and transverse sinus (Zhu, 2000). The carotid arteries and jugular veins lie adjacent to each other and because this anatomical arrangement may facilitate heat exchange between the carotid and jugular blood flow (Cabanac, 1986) it has been suggested that a form of selective brain cooling may occur in humans.

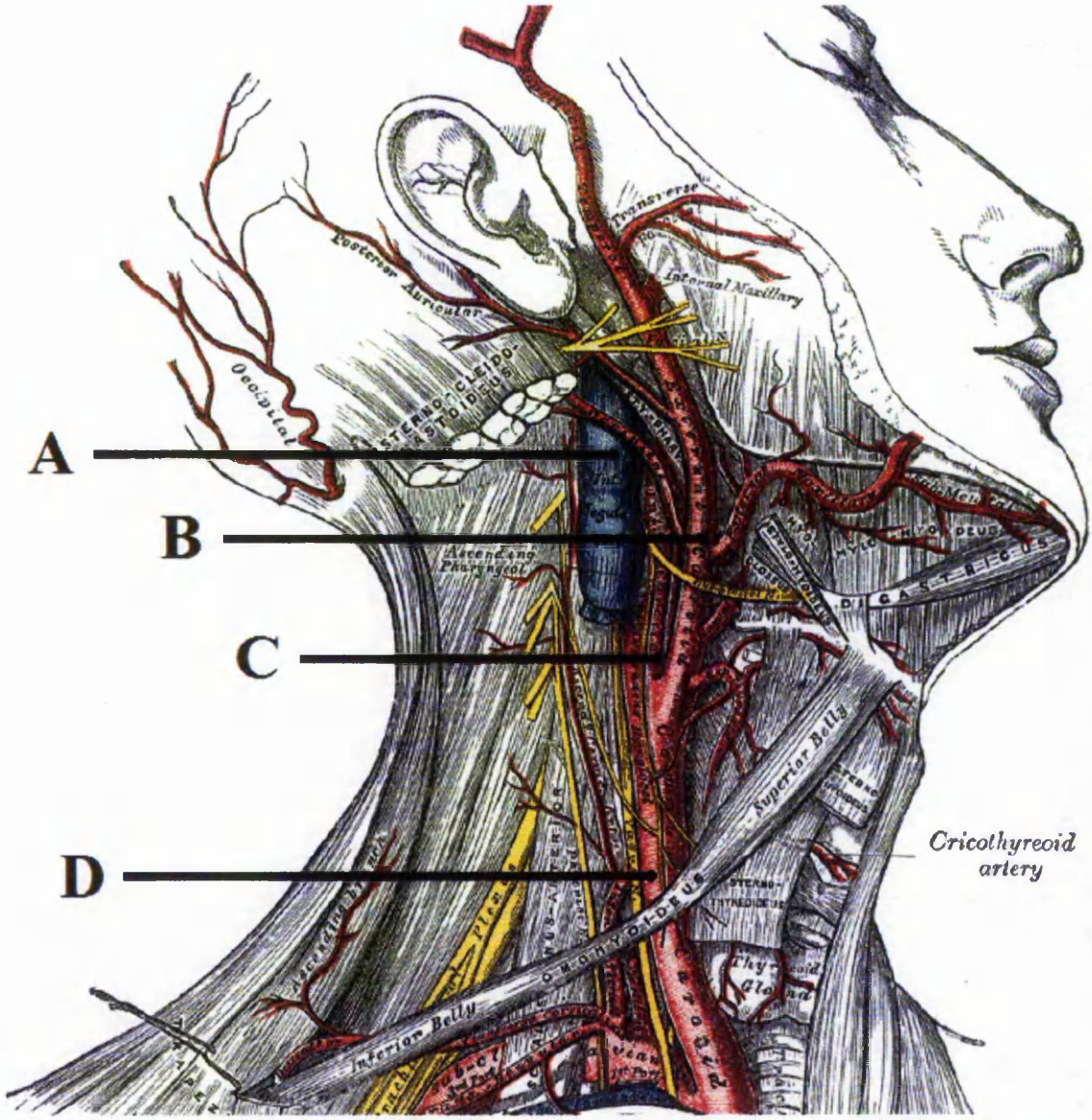

Figure 2.9. The major blood vessels of the human neck. A = internal jugular vein; B = external carotid artery; C = internal carotid artery; D = common carotid artery (modified from Grays, 1918)

It has been proposed that because an excessive heat load could severely damage the brain, a method of cooling is a survival necessity (Cabanac & Brinnel, 1985). During exercise-

induced hyperthermia the heat introduced to the cerebral blood flow poses a threat to brain homeothermia (Rasch *et al.*, 1991). The thermal stability of the brain is heavily dependent on the continuous removal of excess heat at rest and during exercise (Caputa, 2004) and the only way that excess cerebral heat can be removed is via the circulation of blood cooler than the tissue it perfuses (Caputa, 2004). Cabanac and Caputa (1979) proposed that one mechanism by which brain temperature could be reduced is via the pre-cooling of the arterial blood supply to the brain as observed with selective brain cooling in many animal species.

The idea that selective brain cooling occurs in humans has often been suggested by researchers who have observed tympanic temperatures below those recorded in the core of the body in hyperthermic individuals at rest and during exercise (Rasch *et al.*, 1991) however there are anatomical limitations to this suggestion.

The temperature of the arterial blood destined for the human brain is the main determinant of deep brain temperature (Hayward & Baker, 1969) and it has been suggested because the tympanic membrane is located 'close' to the stem of the internal carotid artery and to the hypothalamus (Benzinger, 1969) tympanic membrane temperature can be used as a surrogate for brain temperature and evidence of selective brain cooling. Although this practical measurement is an attractive option, strong evidence suggests that its usage as a surrogate of brain temperature is incorrect (Brengelmann, 1993; Shiraki *et al.*, 1988). In contrast to being 'close' the tympanic membrane and carotid artery are separated by ~1cm in an adult human, with the space being occupied by effective insulators such as bone while the membrane's blood supply comes from the external, not internal, carotid artery (Brengelmann, 1993). Further opposition to the use of tympanic temperature was provided by Nielsen (1988) and Shiraki *et al.* (1988). Nielsen (1988) reported that tympanic temperature was 1°C lower than the temperature measured at the oesophagus while participants cycled outdoors with a wind velocity of 5-7m·sec^{-1} however there was no difference when they cycled indoors in still-air conditions. Due to wax-plugging and insulation of the ear conductional heat loss was unlikely to explain the difference while computation by Nielsen (1988) suggested that only ~0.2°C of the reduction could be explained by convectional heat exchange as a result of the cooler face (~8°C) in the outdoor trials. The external carotid artery provides ~80% of the blood flow to the tympanic membrane and Nielsen (1988) concluded that the difference observed between the tympanic and oesophageal temperatures was explained by local cooling of the regions supplied by the external carotid in the trials conducted outdoors. Shiraki *et al.* (1988)

measured the brain, oesophageal and tympanic temperatures of an anesthetised patient while looking at the effect of facial fanning under various conditions. Facial fanning decreased tympanic temperature, as reported elsewhere (Brown & Williams, 1982),;however ,it had no effect on the temperature of the brain demonstrating directly the independent nature of brain and tympanic temperature. Despite reviews to the contrary (Cabanac, 1993) the evidence from Brenglemenn (1993), Nielsen (1988) and Shiraki *et al.* (1988) highlights the problems with using tympanic temperature as evidence for selective brain cooling in humans and as such the investigations which have used this measurement to highlight the existence of selective brain cooling in humans should be treated with appropriate caution. It appears that direct, rather than surrogate, measurements are required to establish the existence, or non-existence, of a human selective brain cooling mechanism however direct assessment is only possible in exceptional circumstances.

2.4.3.1. Mathematical modelling

Assessment of brain temperature in humans has many physiological and medical applications however, as previously discussed, it can not be measured, except during rare cases (Shiraki *et al.*, 1988), due to the intrusiveness and risk of the procedure. This has led to interest in other ways to indirectly measure or predict the temperature of the brain and factors that may influence brain temperature in a variety of settings. Nunneley and Nelson (1994) suggested that due to the relative simplicity of the anatomy of the neck and the well known thermal properties of the various tissues mathematical modelling of arteriovenous heat transfer can accurately be performed for humans.

A mathematical modelling paper by Nelson and Nunneley (1998) reported that surface cooling of the head was an ineffective method of cooling the human brain because, due to the dense capillarisation of the human brain, the cerebral temperature is largely dictated by the temperature of arterial blood supply and independent of cranial surface temperature. Further computation calculated that brain temperature is typically close to the temperature of the blood in the internal carotid artery before it reaches the circle of Willis (Zhu & Diao, 2001) and that arteriovenous heat transfer is ultimately limited in humans due to the lack of a sufficient temperature difference between the arterial and venous blood (Nunneley & Nelson, 1994). This would suggest that cooling the blood destined for the brain and thus increasing the temperature gradient between the arterial and venous blood may be able to lower the temperature of the brain and Zhu (2000) proposed that theoretically external cooling could lower the temperature of arterial blood on route to the brain.

Sukstanskii and Yablonskiy (2007) attempted to compute the effects of an external cooling device on the temperature of the brain using a different mathematical approach. They suggested that brain temperature is higher than that of the carotid blood as a result of metabolic heat production within the brain- a term they called 'metabolic temperature shift'. It has been shown that significant decreases in brain temperature can only be observed near to the brain surface- within a 'shielding length' inversely proportional to the square root of cerebral blood flow (Sukstanskii & Yablonskiy, 2004; Zhu *et al.*, 2006). The typical human cerebral blood flow is ~50 $ml \cdot 100g^{-1} \cdot min^{-1}$ which results in a shielding length of ~3-4mm, far shallower than the diameter of the human brain which is ~14cm (Sukstanskii & Yablonskiy, 2007). The consequence of this is that in normal conditions the temperature of the human brain remains essentially constant except for minor variation within a 3-4mm superficial shell. They computed that for deep cooling to occur (defined as a drop of 2-3°C at a depth of ~2-3cm) cerebral blood flow would have to fall to less than 10 $ml \cdot 100g^{-1} \cdot min^{-1}$. This cerebral blood flow computed by Sukstanskii and Yablonskiy (2007) has been reported in pre-term and full-term infants (Altman *et al.*, 1988) however it is well below the values observed by Nybo *et al.* (2001) during prolonged hyperthermic cycle ergometer exercise an adults (42.9 ± 4.3 $ml \cdot 100g^{-1} \cdot min^{-1}$). This suggests that although deep brain cooling may be theoretically possible, particularly in neonates, via the application of an external cooling device, during exercise a cooling device can only influence the extremities of the brain at best and have no influence on the temperature of the deep cerebral tissue. It is worth noting that due to the narrow temperature window in which life exists a drop of 2-3cm may be far in excess of what is actually required to gain a cooling benefit.

Although the deep brain temperature is practically homogenous it has been proposed that the application of an external cooling device may be able to alter the temperature of the blood on route to the thermoregulatory centre located within the hypothalamus (Zhu, 2000). The hypothalamus is located at the base of the brain, below the level of the thalamus. The blood is supplied to the hypothalamus via the terminal branches of the circle of Willis; the interior carotid; the anterior and posterior cerebral; the anterior and posterior communicating; and the basiliar arteries (Daniel, 1966). Extensive study of the human hypothalamus has established that the arterial blood supply can be separated into anterior, intermediate and posterior groups and that it is split between the three rostro-caudal regions of the hypothalamus (Haymaker, 1969). The preoptic and anterior hypothalmus regions which make up the thermoregulatory centre are primarily supplied by the anterior

cerebral and anterior communicating arteries (Haymaker, 1969). The anterior communicating arteries branch off from the anterior cerebral arteries which themselves branch off from the internal carotid artery. This relationship demonstrates the relatively direct route that the arterial blood takes from the carotid artery to the thermoregulatory centre and offers further support for theoretically being able to directly influence hypothalamic temperature via the manipulation of the temperature of the blood that supplies it which travels from the heart to the brain via the neck region. This theory requires further, direct, investigation.

2.4.3.2. Data from human investigations

Brain temperature is primarily determined by the temperature of the blood reaching the brain and changes in cerebral arterial blood temperature result in similar changes throughout the brain within seconds (Baker, 1982). As a result of this relationship a cooling intervention that was able to lower the temperature of the arterial blood would be expected to reduce the temperature of the brain. Shiraki *et al.* (1988) measured the brain temperature of a 12-year old male patient subjected to passive hyperthermia with 20 min of facial cooling while undergoing surgery to relieve intracranial pressure caused by a pineal tumour. Facial fanning decreased the tympanic temperature however it had no effect on the temperature of the brain (Figure 2.10). Similarly, Nybo *et al.* (2002) reported that there was no change in the arterial-to-venous blood temperature across the brain after 5 min of facial fanning. Although both studies reported that facial fanning had no effect on the temperature of the brain, as proposed by Nybo *et al.* (2002), it is possible that the short periods of facial fanning were insufficient to cool the human brain and that a longer exposure may have an effect or that a more aggressive cooling method may elicit different results.

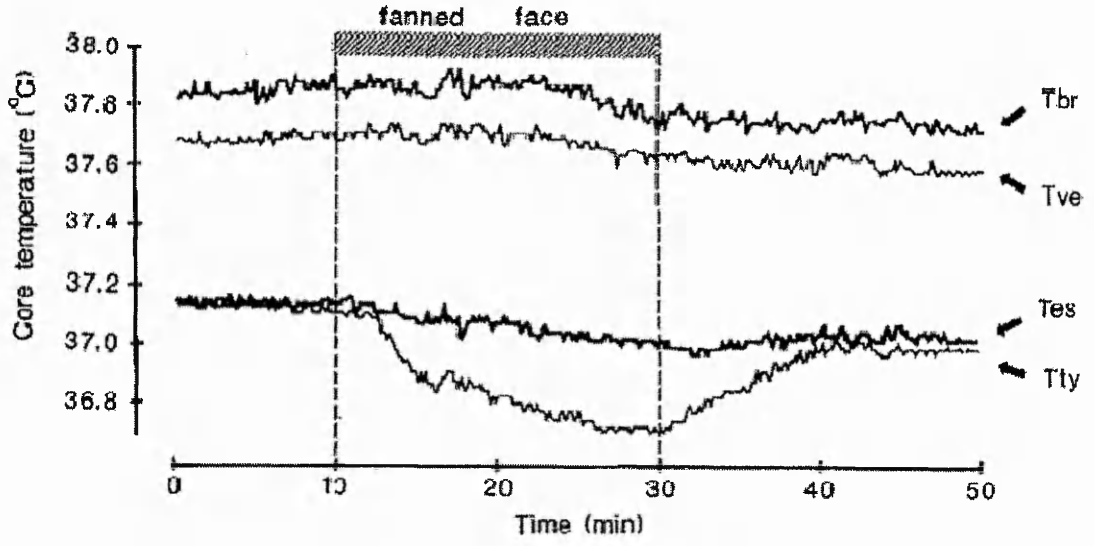

Figure 2.10. Temperatures recorded during fanning of the face. Tbr = brain temperature; Tve = temperature in lateral ventricle; Tes = oesophageal temperature; Tty = tympanic temperature [reproduced from Shiraki *et al.* (1988) with permission]

Direct cooling of the anterior aspect of the homolateral side of the forehead and the face with cotton-encased ice chips has been demonstrated to decrease the temperature of common carotid blood by 0.2-0.5°C in patients undergoing arteriography although breathing air cooled to 4°C did not have any effect (Rubenstein *et al.*, 1960). This is in contrast to Mariak *et al.* (1999) who investigated the effect of inhaling cool air (22°C) intensively (18-20 breaths per min) for 3 min. Mariak *et al.* (1999) reported that under mild passive hyperthermia (induced via a small heating pad) the passage of the inhaled cool air through the upper respiratory tract is able to lower the temperature of the frontobasal brain surface below the temperature recorded at the oesophagus. Further evidence disputing the ability to lower the temperature of the human brain via direct, external, cooling interventions was provided by Corbett and Laptook (1998). Localised cooling of the head via ice-water has been shown to decrease scalp temperatures to ~25°C and subsequently brain temperature to ~30°C in less than 25 min in rats (Barone *et al.*, 1997; Towfighi *et al.*, 1994) and cats (Horn *et al.*, 1991). Due to the difficulty in directly measuring human brain temperature following localised cooling Corbett and Laptook (1998) investigated the effects of localised head cooling on human brain temperature via a method called proton magnetic resonance spectroscopy (^{1}H-MRS) which is a validated non-invasive method to assess brain temperature (Corbett *et al.*, 1995). In the study participants had their scalp cooled to ~16°C (a reduction of ~19°C) via two cooling caps wrapped over the skull and neck worn for 50 min. They reported that the cooling intervention rapidly cooled the head

reaching its lowest value after only 5 min however despite the rapid and pronounced reduction there was no effect on deep or superficial brain temperature. It was postulated that a longer period of cooling may reduce brain temperature but this was not investigated. This data shows that further investigation is required however; from the literature to date it appears that direct cooling via a variety of means has no effect on the temperature of the brain.

2.4.4. Central fatigue and head cooling

Although the evidence with regards to head and neck cooling and the direct alleviation of cerebral hyperthermia is lacking, indirect assessment has suggested that cooling this region may have a direct effect on the central aspect of fatigue.

As reviewed earlier the role of the neurotransmitters serotonin and, more dominantly, dopamine in the premature termination of exercise in a hot environment has received recent interest. It appears that dopamine levels within the brain have a strong influence on exercise adherence (Roelands *et al.*, 2008a; Roelands *et al.*, 2008c; Watson *et al.*, 2005c). Direct measurement of cerebral levels during exercise is not possible in humans and therefore peripheral hormone concentrations have been employed as an index of central neurotransmission (Chandler & Blair, 1980). The most commonly used peripheral marker is prolactin. Prolactin is a polypeptide hormone involved in many homeostatic roles. Prolactin release is inhibited by dopaminergic activity, due to its interaction with the prolactin-releasing factors thyrotropin-releasing hormone and oxytocin, and stimulated by serotonergic pathways (Freeman *et al.*, 2000) and therefore it has been proposed as a peripheral marker of central fatigue in hot environments.

Hyperthermia has been implicated in the elevation of prolactin levels because it has been demonstrated that heat exposure (Frewin *et al.*, 1976; Mills & Robertshaw, 1981) and exercise (Brisson *et al.*, 1986b; Brisson *et al.*, 1986a) both result in increased peripheral prolactin concentrations. It has been proposed that the temperature of the brain may play a key role in the secretion of prolactin (Radomski *et al.*, 1998) and therefore the effect of cooling of the head and face region has on the prolactin response has been investigated. Cooling the head via cool air and misting has been shown to have little effect on the physiological response to exercise (Brisson *et al.*, 1987; Brisson *et al.*, 1989) however has been shown to attenuate the prolactin response to exercise performed in warm to hot (26 – 41°C) temperatures in most (Ansley *et al.*, 2007; Armada-da-Silva *et al.*, 2004; Brisson *et*

al., 1987; Brisson *et al.*, 1989; Mundel *et al.*, 2006) but not all (Brisson *et al.*, 1991) studies. In the study by Brisson *et al.* (1991) two methods of facial cooling were investigated during either 45 min of submaximal cycle ergometer exercise in high ambient temperatures (41°C) or 30 min hot water immersion (41°C). Participants were either cooled via the provision of cooled air (10°C) or fanned with ambient air (41°C). Neither method of cooling had any effect on rectal temperature and both the endogenous and exogenous heat loading methods significantly elevated prolactin concentrations. The elevation in prolactin was unaffected by the fanning (41°C ambient air) treatment however the cooling (10°C air) intervention significantly attenuated the prolactin response in both conditions (Figure 2.11.).

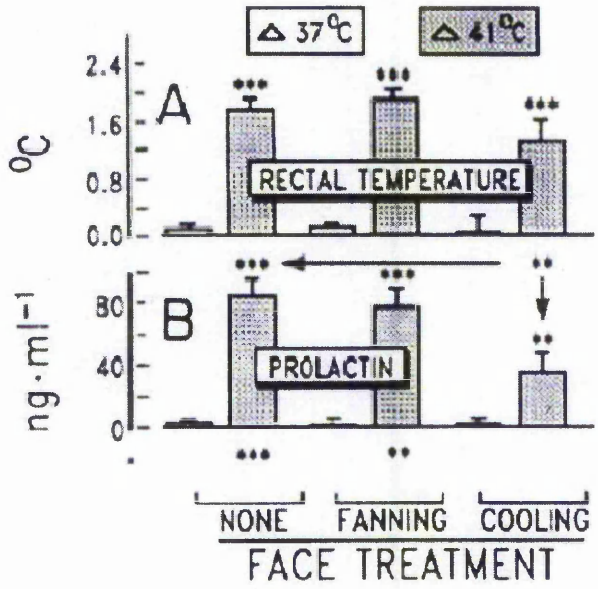

Figure 2.11. Changes in rectal temperature (A) and concentrations of prolactin following head-out water immersion in the absence or presence of face fanning and cooling. ** = $P < 0.01$; *** = $P < 0.001$ [reproduced from Brisson *et al.* (1991) with permission].

Based upon the data of Roelands *et al.* (2008) the attenuation of prolactin would be expected to be associated with enhanced exercise performance in the heat if prolactin represents dopaminergic activity however to the author's knowledge this has only be investigated in one recent study. Ansley *et al.* (2007) reported that cooling the head and face region via cooling fans resulted in an improvement of ~51% in cycle ergometer capacity in a hot environment and a significant attenuation in the prolactin response to the exercise bout from 10 min to the point of fatigue. The difference between trials was pronounced with the six-fold elevation observed in the control trial being reduced to a two-fold increase with the cooling. Care when using prolactin as a marker is required however as it has been demonstrated that cooling the skin can reduce skin temperature independently of core temperature (Armada-da-Silva *et al.*, 2004; Bridge *et al.*, 2003a; Low *et al.*, 2005) and that central serotonergic manipulation via nutritional or pharmacological interventions fails to alter the prolactin response to exercise (Strachan *et*

al., 2004; Strachan *et al.*, 2005). The data from Ansley *et al.* (2007) suggests that a cooling-induced reduction in prolactin concentrations may be able to explain the markedly improved performance in the heat however further research is required.

2.4.6. Theoretical basis for the application of a cooling collar summary

It is well documented that animals are able to reduce the temperature of the brain below that of the trunk via a method termed selective brain cooling and it has been proposed that due to the vulnerability of the cerebral tissues this mechanism is an evolutional adaptation protecting the brain from dangerous levels of hyperthermia (Cabanac, 1993). Anatomical reviews and direct cooling interventions in anesthetised humans suggest that humans do not have the ability to selectively brain cool (Brengelmann, 1993; Shiraki *et al.*, 1988) however mathematical modelling studies have suggested that peripheral brain temperature can be reduced by the application of a cooling device (Sukstanskii & Yablonskiy, 2004; Zhu, 2000). The obtainment of a high brain temperature has been proposed as a major cause of the premature termination of exercise in a hot environment and therefore it has been proposed that an external cooling device may enhance exercise ability via the reduction in brain temperature however this is speculative and warrants further investigation. Central concentrations of neurotransmitters have also been implicated in the development of fatigue in a hot environment (Meeusen *et al.*, 2006) and peripheral concentrations are often used as a surrogate measurement of central activity. Cooling the head and face region has been shown to improve exercise capacity and drastically attenuate the prolactin response to the exercise bout (Ansley *et al.*, 2007) however there is controversy regarding the use of prolactin as an indirect marker of central fatigue (Meeusen *et al.*, 2006) and so the ergogenic effect of facial cooling may result from alternative mechanisms.

2.5. Review of literature summary

The negative effect of elevated ambient temperatures on exercise performance and capacity is well documented (Galloway & Maughan, 1997) however the reasons for this are not fully understood. It has been demonstrated that during fixed-intensity exercise that voluntary termination occurs consistently at a core temperature of ~40°C (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993) and the literature from animal investigations suggests that it is the obtainment of a high brain temperature that ultimately limits exercise capacity (Caputa *et al.*, 1986; Fuller *et al.*, 1998). In variable-intensity exercise self-selected pace is down-regulated before the obtainment of a high core temperature and it has been proposed that in such exercise procedures a pacing strategy is adopted that will allow the task to be completed within homeostatic limits (Marino, 2004; Tucker *et al.*, 2004). The cause of the termination of exercise at high temperatures or the down-regulation of intensity to prevent hyperthermia developing is not fully understood however it is clear from neuromuscular physiology research that there is a central component to fatigue (Nybo & Nielsen, 2001a). Cerebral concentrations of neurotransmitters have been implicated in the onset of fatigue in a hot environment and recent investigations suggest that fatigue in such conditions is heavily linked to cerebral levels of dopamine (Roelands *et al.*, 2008a; Roelands *et al.*, 2008c).

Although the exact mechanisms limiting exercise capacity and performance in a hot environment are not fully understood it is clear that hyperthermia plays an integral role. Many interventions have attempted to attenuate the reduction in exercise performance and capacity observed by reducing the level of hyperthermia experienced. Pre-cooling has consistently been demonstrated to improve subsequent performance with and without reductions in physiological variables (Hessemer *et al.*, 1984; Lee & Haymes, 1995) however lack practical application in a sporting competition. Practical cooling devices have been less extensively investigated however when offering a sufficient magnitude of cooling have been demonstrated to be effective in alleviating levels of physiological and perceived strain (Arngrimsson *et al.*, 2004). Cooling the neck region has many proposed benefits over cooling elsewhere and it has been demonstrated that cooling this site can elicit a disproportionate cooling effect (Shvartz, 1976) possibly due to its close proximity to the thermoregulatory centre. Investigations that have cooled this region via the application of a practical cooling device have reported no beneficial physiological alterations however they did not look at the effect of cooling this site on exercise capacity or performance (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). Exercise performance and

capacity can both be enhanced in hot conditions via the application of a cooling intervention without alterations in physiological variables (Ansley *et al.*, 2007; Hessemer *et al.*, 1984) and therefore such changes are not essential to improve exercise performance in a hot environment. It has been demonstrated that cooling the face and head region via facial fanning and a water-perfused hood can improve exercise capacity and performance in a hot environment (Ansley *et al.*, 2007; Palmer *et al.*, 2001); however, the effect of practical devices on performance and capacity are unknown.

2.6. Research aims of the thesis

The research aims of the thesis were to investigate the effect of cooling the neck region, via the application of a practical neck-cooling collar, on exercise performance and capacity in a hot environment and to examine the physiological, perceptual and neuroendocrinological responses to the cooling intervention.

In order to achieve these aims, five investigations were conducted and these investigations are presented in the following six Chapters. Due to different hypotheses and to maximise clarity, data collected from the second experimental study is presented in two Chapters- Chapters five and six. The five investigations that were conducted are presented in the order in which they were conducted to demonstrate the progressive nature of the work and to highlight the organic nature of the study design process adopted.

Chapter 3: General Methods

3.1. Introduction

This Chapter explains the methodological procedures that were used in the experimental studies described within this thesis. Information regarding participant recruitment, experimental procedures and measurements, blood collection, treatment and analysis and statistical analysis is presented.

3.2. Participant recruitment

Volunteers for all studies were recruited by word of mouth; poster campaigns and electronic mailing. Participants were recruited from the male staff and students at Nottingham Trent University and from the male members of local running and triathlon clubs. All participants were recreationally active with experience of long distance running.

3.3. Procedures and measurements: all studies

3.3.1. Ethical approval

Ethical approval was granted by Nottingham Trent University's Ethical Advisory Committee. Potential volunteers for all studies were given participant information sheets detailing the experimental protocol and any potential risks and discomforts involved with participation. Prior to the start of the first experiment participants were asked to sign a consent form which confirmed that the protocol, and associated risks and/or discomforts, had been fully explained and that they were fully aware of the procedures that they would be expected to undertake. Prior to each laboratory visit participants completed and signed a full health screening questionnaire to confirm that they were in good health and physically able to perform the required exercise. All volunteers were made fully aware that they were free to withdraw their participation in the study at any time without having to provide a reason for doing so.

3.3.2. Estimation of maximal oxygen uptake

Maximal oxygen uptake was measured via indirect calorimetry on a motorised treadmill using an incremental protocol as per Jones and Doust (1996). Participants completed 5 - 7 3 min stages of increasing submaximal workload on a 1% gradient until the individual's lactate threshold was achieved. Participants began the incremental phase of the protocol at 10km·h^{-1} and ran at speeds increasing by 1 km·h^{-1} per 3 min stage. The "lactate threshold"

was determined by plotting whole blood lactate ($mmol \cdot l^{-1}$) versus speed ($km \cdot hr^{-1}$) and determining the speed at which the increase in lactate, compared to the previous speed, was greater than or equal to 1 $mmol \cdot l^{-1}$ (Bishop, 1997). Following a short interval (~10 min) the participants exercised at the speed immediately preceding the lactate threshold with an increase in the gradient of $1\% \cdot min^{-1}$ until volitional exhaustion. Expired air was collected via the Douglas bag technique during the last min of each sub-maximal and maximal stage. A Harvard dry gas meter (Harvard Apparatus Ltd, Kent, England, U.K.) was used to measure the volume and temperature of the expired air (Fisher Scientific Ltd, Loughborough, England, U.K.). Expired air was analysed for oxygen and carbon dioxide concentration using a Servomex 1440 gas analyser (Servomex Group Ltd, Crowborough, U.K.) calibrated prior to use with gas of known-concentration. Maximal oxygen uptake was defined as the value obtained during the final complete expired air collection period and was assumed to have been met if one of the following criteria was achieved; a maximum heart rate (HR) of no less than 15 beats below age-predicted (220-age) maximum (Jackson *et al.*, 1990) or a respiratory exchange ratio of equal to, or greater than, 1.1. The submaximal values were plotted and the speed required to elicit the desired percentage of maximal oxygen uptake was calculated using regression analysis.

3.3.3. Pre-trial standardisation

Participants were required to standardise their diet for 24h prior to each trial, abstaining from alcohol and caffeine. Participants were issued with a diet and physical activity diary at the initial meeting and instructed to record all food and fluid consumed during the 24h before the first trial. They were then requested to follow the same diet in the 24h before subsequent trials. Participants were also asked to refrain from strenuous physical activity during the 48h before each trial and repeat their physical activity patterns before following trials. For the studies in Chapters 4, 5, 6 and 8 participants reported to the laboratory $\geq$ 10h post-prandial following an overnight fast except for the ingestion of 500 ml of water ~1.5h before their arrival to ensure that the participants were euhydrated at the start of each trial. For the studies in Chapters 7 and 9 participants were $\geq$ 4h post-prandial.

3.3.4. Physiological and perceptual measurements

3.3.4.1. Rectal temperature

Rectal temperature was measured via a rectal thermistor (Grant Instruments (Cambridge) Ltd, England, U.K.) self-inserted ~10cm past the anal sphincter connected to a multi-port

data logger (Squirrel 2040, Grant Instruments (Cambridge) Ltd, England, U.K.). Rectal thermistors were calibrated against mercury thermometers using water-bath immersion prior to use. Participants used the same, sterilised, rectal thermistor for each trial.

3.3.4.2. Heart rate

Heart rate was monitored throughout all trials via heart rate a monitor (Polar Electro Oy, Kempele, Finland). A tight fit and good contact was ensured via the provision of a variety of strap sizes and the moistening of all contact points.

3.3.4.3. Oxygen uptake

Expired air was collected via the Douglas bag technique. A Harvard dry gas meter (Harvard Apparatus Ltd, Kent, England, U.K.) was used to determine expired air volume and temperature (Fisher Scientific Ltd, Loughborough, England, U.K.). Expired air was analysed for oxygen and carbon dioxide concentration using a Servomex 1440 gas analyser (Servomex Group Ltd, Crowborough, U.K.) calibrated with known-concentration gas.

3.3.3.4. Stature

Stature was measured using an upright stadiometer (Seca U.K., Birmingham, England, U.K.). Stature was measured without footwear and according to standard procedure.

3.3.3.5. Nude body mass

Nude body mass was recorded pre and post trials by the participants themselves. Prior to doing so participants were familiarised with the weighing scales. Over the course of the studies within the thesis different sets of weighing scales were used (all Seca, Birmingham, England, U.K.) although the same set was used for each participant within trials.

3.3.3.6. Sweat loss

Sweat loss was estimated using pre and post exercise bout dry body mass differences taking into account voluntary water consumption and, where necessary, urinary losses.

3.3.3.7. Rating of perceived exertion

Rating of perceived exertion was measured using the standard 6 - 20 scale (Borg, 1982). The scale was described and explained prior to their trials.

3.3.3.8. Thermal sensation

Thermal sensation was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987). The scale was described and fully explained using standard cues prior to each of their trials.

3.3.5. Environmental conditions

All main trials were conducted within a walk-in environmental chamber (Model: WIR52-20HS, Design Environmental Ltd, Gwent, Wales, U.K.) designed to control ambient temperature (range: -20°C to +40°C) and humidity (+5 to +95%). In addition to setting the walk-in environmental chamber to the required environmental conditions using the accompanying console unit additional measurements were recorded throughout.

3.3.5.1. Measurement of ambient temperatures

Ambient temperature was measured using a digital thermometer (Fisher Scientific Ltd, Loughborough, England, U.K.), fixed in a standardised position during each trial.

3.3.5.2. Measurement of relative humidity

Relative humidity was assessed using a whirling hygrometer (Zeal, London, U.K.) rotated for ~20-seconds prior to each reading as per the manufacturers recommendations.

Relative humidity was then calculated from the wet-bulb (wb) and dry-bulb (db) temperatures using the following formula:

$$(6.11*10.0*(7.5*T_{wb}/(237.7 + T_{wb}))) / (6.11*10.0*(7.5*T_{db}/(237.7 + T_{db}))) * 100$$

3.4. Main trial protocol (Chapters 4, 5, 6 and 8)

During each trial participants ran at a speed calculated to elicit 60% of their individual $\dot{V}O_{2max}$ for 75min immediately followed by a self-paced 15min time-trial on a motorised treadmill, which was standardised for each participant for each trial. No indication of distance ran or running speed was given throughout the preload phase of the trials. The only indication of the time elapsed given was during the 15 min time-trial when participants were able to see a countdown timer placed outside the direct line of vision. During the time-trial participants were encouraged to run as far as possible in the 15 min and were able to increase or decrease the running speed manually as desired, although they were unaware of the speeds at which they were running. Water was allowed *ad libitum* throughout the trials and the volume consumed was recorded. Feedback regarding time-trial performance and the response of physiological variables was only given after the completion of all trials. Trials were conducted at the same time of the day ± 30min and 7 - 14 days apart.

3.5. Main trial procedures and measurements (Chapters 5, 6, 7, 8 and 9)

3.5.1. Mean neck temperature

Mean neck temperature was calculated as the mean temperature of four skin thermistors (Grant Instruments (Cambridge) Ltd, England, U.K.) spaced equally across the posterior aspect of the neck. All thermistors were attached via a transparent dressing (Tagaderm, 3M Health Care, USA) and water-proof tape (Transpore, 3M Health Care, USA).

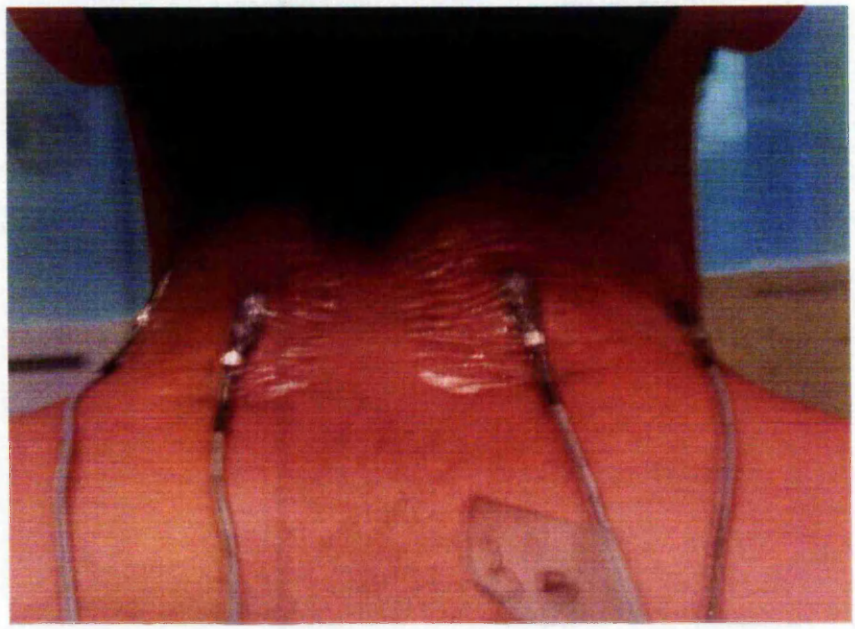

Figure 3.1. The location of the four skin thermistors used to determine mean neck temperature.

3.5.2. Neck cooling collar

3.5.2.1. Design and evaluation

Having established the theoretical basis for the ergogenic affect of the application of a cooling collar around the human neck it was imperative that an adequate cooling collar was used. As reviewed in Chapter 2, problems that have occurred during other cooling studies tend to have arisen due to the cooling intervention used either not cooling the relevant area sufficiently or for a long enough duration. To combat this, pilot work was undertaken to ensure that the collar investigated would significantly cool the area for a sustained duration of the test. After an extensive search, two commercially available neck cooling devices were sourced. These collars as well as modified versions of these two collars were investigated in the design and evaluation stage.

3.5.2.2. Pilot methods

One healthy, trained female volunteer participated in the pilot trials. Their age, body mass, height and maximal oxygen uptake ($\dot{V}O_{2max}$) was 31-years, 56.5 kg, 1.61 ± 0.07 m and 49.9 $ml \cdot kg^{-1} \cdot min^{-1}$. The volunteer completed five bouts of 60min of treadmill running at a speed designed to elicit 60% $\dot{V}O_{2max}$ (7.9$km.h^{-1}$) wearing one of the collars piloted and one bout without a collar for comparison. Due to the development process the exercise bouts were conducted over a five month period (July 2006 – January 2007).

3.5.2.3. Collars evaluated

Four variations of cooling collars were piloted during the design and evaluation process; a commercially available Total Cool ice cooling collar (TC) (www.Totalcool.biz); a modified version of the Total Cool ice cooling collar (TC-MOD); a commercially available Black Ice 'Smart Ice^{TM}' cooling collar (BI) (Black Ice LLC, Lakeland, USA) and a modification of the Black Ice Smart Ice^{TM} cooling collar (BI-MOD). The collars were frozen for 24 to 28h at -80°C for all collar pilot trials except for the first one (TC -20) for which the collar was frozen for 24 to 28h at -20°C. The collar was left in ambient conditions for 5min following removal from the freezer and wiped of any surface frost prior to being worn.

3.5.2.3.1. Total Cool Ice cooling collar

The TC was comprised of a fabric neck wrap (550mm (L) x 55-95mm (W)) with a meshed pocket on the skin-surface side into which a segmented strip of ice pockets (365mm (L) x 70mm (W) x 20mm (D)) (Figure 3.2) was inserted. The collar weighed 110g when dry.

Figure 3.2. Total Cool fabric neck wrap and segmented ice strip.

3.5.2.3.2. Modified Total Cool Ice cooling collar

The fabric neck wrap of the TC-MOD was identical in design to the commercially available version. The modification involved replacing the strip of ice pockets with three individual cooling packs (105mm (L) x 50mm (W) 10mm (D)) comprised of a double layer of thin plastic tubing sealed and filled with ~32g of gel refrigerant (BDH Laboratory Supplies, Poole, Dorset, England, U.K.).

3.5.2.3.3. Black Ice cooling collar

The Black Ice cooling collar (Black Ice LLC, Lakeland, USA) was comprised of two parts (Figure 3.3). The cooling section was made from a thin plastic and consisted of five compartments filled with ~180g of Smart Ice™, attached via three Velcro fastenings to a 600mm neoprene wrap secured with Velcro fastenings at the anterior aspect of the neck. The dimensions of the cooling section of the collar were 375mm (L) x 60mm (W) x 15mm (D) weighing 210g when dry.

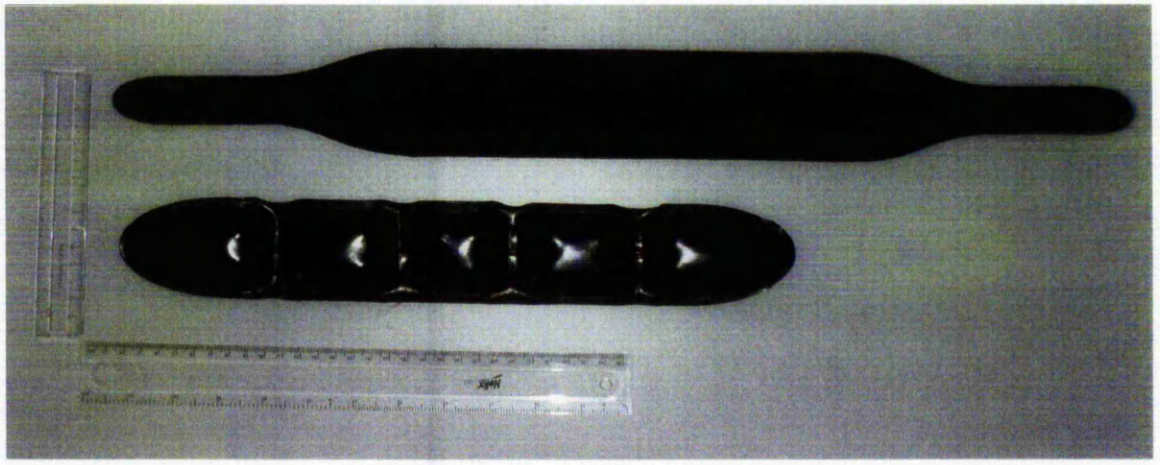

Figure 3.3. The Black Ice cooling collar system. The cooling section is shown below the neoprene wrap.

3.5.2.3.4. Modified Black Ice cooling collar

The BI-MOD was identical in design and dimensions to the commercially available model; however, the Smart Ice™ cooling fluid was drained and replaced with 120g of gel refrigerant (BDH Laboratory Supplies, Dorset, England, U.K.). The modified collar weighed 180g when dry.

3.5.2.4. Pilot results and discussions

3.5.2.4.1. Total Cool cooling collar

3.5.2.4.1.1. Results

The initial TC test was conducted with the collar frozen at -20°C. This resulted in cooling the neck by 4.33 ± 1.22°C. Freezing the collar at -80°C resulted in a better cooling effect

and lowered T_{neck} by 11.40 ± 2.63°C. The T_{neck} observed during the trials while wearing the TC frozen at -20°C (TC -20) and -80°C (TC -80) compared to no collar (without) is shown in Figure 3.4.

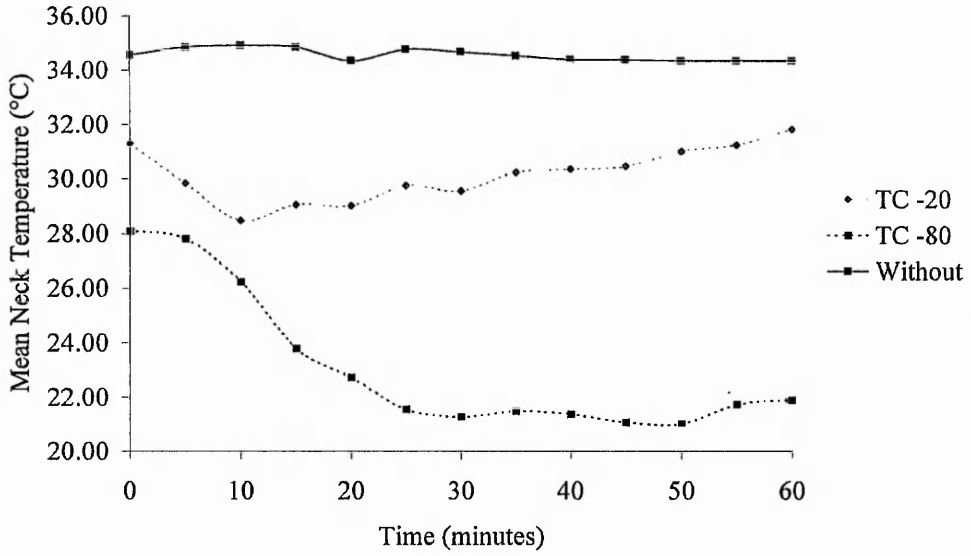

Figure 3.4. Neck temperature recorded during 60min of submaximal treadmill running in hot (30°C) conditions with and without the application of a Total Cool cooling collar frozen at -20°C (TC -20) and -80°C (TC -80).

3.5.2.4.1.2. Discussion

The data clearly shows the application of the TC frozen at either temperature can lower T_{neck} and that unsurprisingly freezing it at -80°C lowers the temperature to a greater degree than freezing it at -20°C. The degree of cooling achieved by TC -80 was substantial but resulted in extreme levels of discomfort and minor ice burns in areas of the neck. Due to this the TC -80 was considered inappropriate to use.

3.5.2.4.2. Modified Total Cool Ice cooling collar

3.5.2.4.2.1. Results

The results achieved by freezing the TC-MOD within a -80°C industrial freezer compared to the commercially available TC and no collar are shown in Figure 3.5. The cooling stimulus achieved was more constant in TC-MOD but was not as strong as the TC and the

T_{neck} began to rise after about 20min. TC-MOD maintained T_{neck} below the T_{neck} observed without the collar throughout.

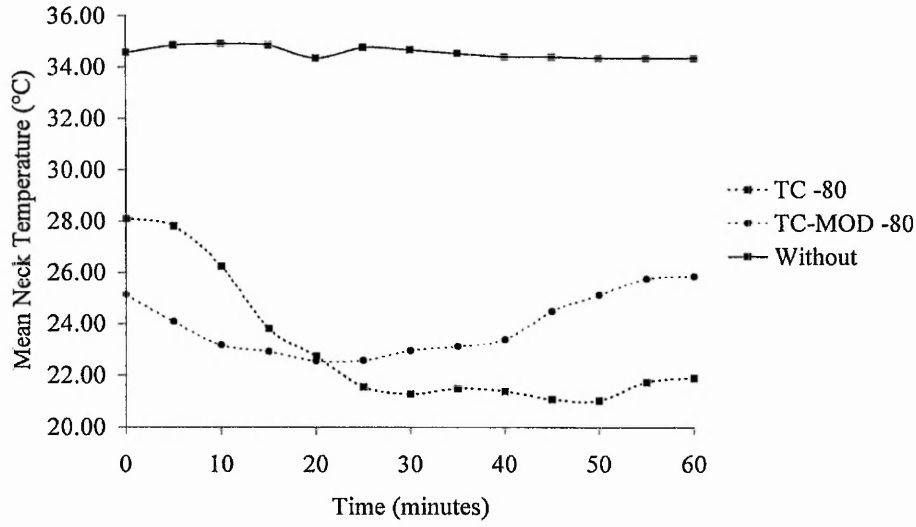

Figure 3.5. Neck temperature recorded during 60min of submaximal treadmill running in hot (30°C) conditions with and without the application of a Total Cool cooling collar (TC -80) and a modified Total cool cooling collar (TC-MOD -80) frozen at -80°C.

3.5.2.4.2.2. Discussion

The extreme discomfort and skin tissue damage that occurred as a result of the TC cooling collar led to its modification. The TC-MOD cooling collar cooled the neck region to a lesser degree than TC but resulted in no ice burns. The fabric neck wrap of the TC collar system was quite restrictive so, although it looked promising as a cooling device, a potential alternative was sourced and tested.

3.5.2.4.3. Black Ice cooling collar

3.5.2.4.3.1. Results

The T_{neck} achieved via the application of the Black Ice (BI) collar compared to the TC-MOD and T_{neck} without a collar is shown in Figure 3.6. It is clearly shown that the effectiveness of the BI cooling is far below that of the TC-MOD. T_{neck} is effectively the

same after 60min with BI cooling collar and without a collar and it can be assumed that T_{neck} would be higher with BI than without if the pilot extended past 60min.

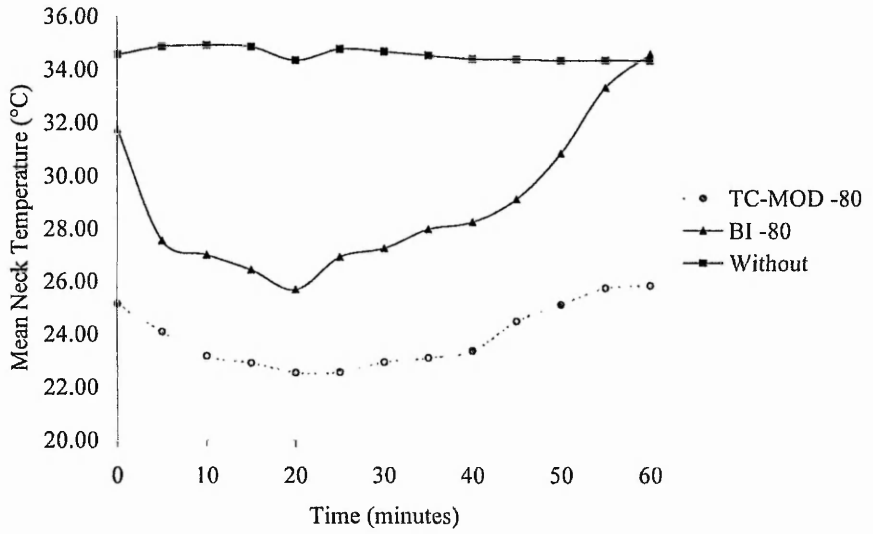

Figure 3.6. Neck temperature recorded during 60min of submaximal treadmill running in hot (30°C) conditions with and without the application of a Black Ice cooling collar (BI -80) and a modified Total cool cooling collar (TC-MOD -80) frozen at -80°C.

3.5.2.4.3.2. Discussion

The Black Ice collar was much more comfortable and the participant reported that the levels of associated discomfort were much attenuated. Unfortunately, the cooling elicited by the collar was much lower than provided by the TC and TC-MOD and T_{neck} was effectively the same with the BI collar than without any collar at 60min, meaning that the collar actually had a progressive warming, rather than cooling, effect.

3.5.2.4.4. Modified Black Ice cooling collar

3.5.2.4.4.1. Results

Modifying the BI cooling collar enabled a lower T_{neck} to be achieved and for it to be sustained for the duration of the protocol (Figure 3.7). The T_{neck} at the termination was 25.75°C with BI-MOD compared to 34.58°C with the commercially available BI collar.

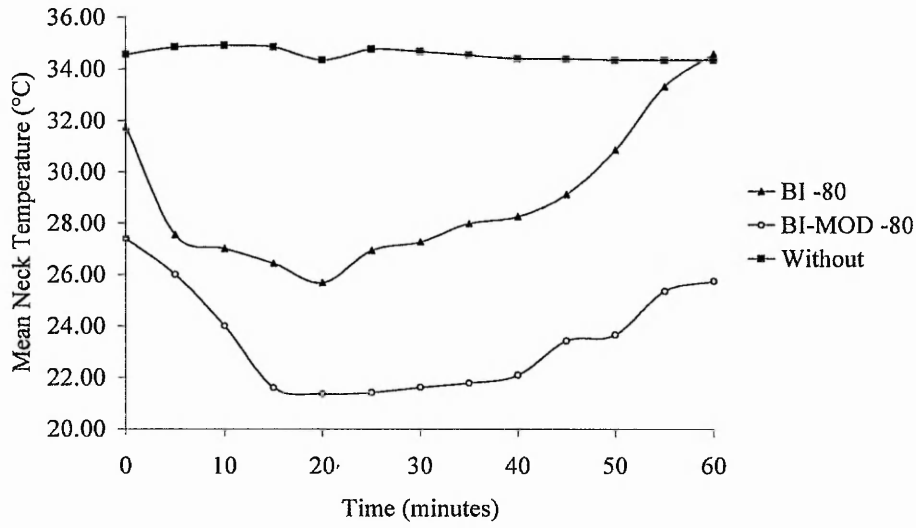

Figure 3.7. Neck temperature recorded during 60min of submaximal treadmill running in hot (30°C) conditions with and without the application of a Black Ice cooling collar (BI -80) and a modified Black Ice cooling collar (BI-MOD -80) frozen at -80°C.

3.5.2.4.4.1. Discussion

Modifying the commercially available BI cooling collar resulted in a greater cooling effect than the commercially available BI collar and a similar cooling effect to the TC-MOD. Unlike the TC-MOD collar the participant reported that discomfort associated with the BI collar was minimal. This is likely to be due to the smaller nature of the BI collar which allows the T_{neck} to be lowered with minimal restriction to movement and therefore minimal disruption to the natural running style.

3.5.2.5. Collar design conclusion

The magnitude of the cooling achieved varied greatly depending on the collar used (Figure 3.8). Out of all the collars investigated, TC -80 achieved the greatest reduction in T_{neck}; however, this was associated with discomfort and skin tissue damage. The application of the BI-MOD -80 collar lowered T_{neck} to a greater extent than TC -80 between 0 and 20min and a similar extent between 25 and 40min. After 40min the T_{neck} was higher with BI-MOD -80 than TC -80 although it remained lower than with the other collars investigated for the remaining 20min of the pilot trial. The BI-MOD -80 collar lowered T_{neck} to the

greatest extent without resulting in skin damage or restricting running motion and so this collar was investigated

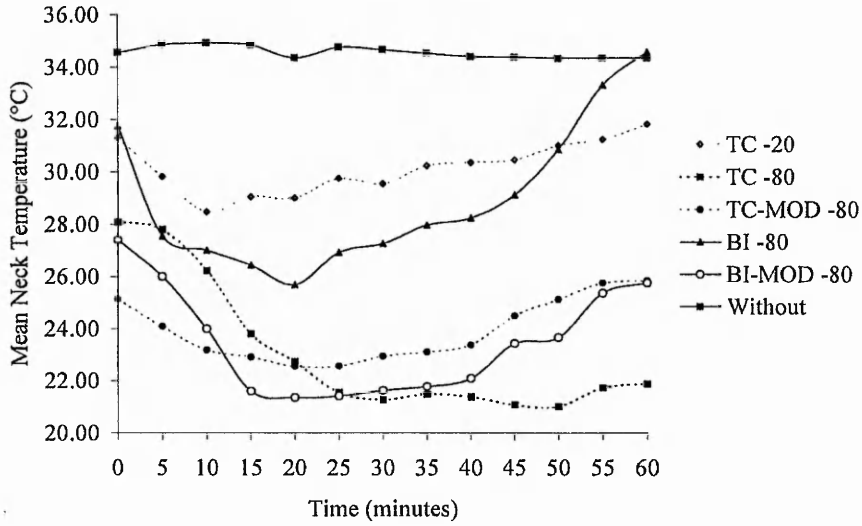

Figure 3.8. Neck temperatures recorded during 60 min of submaximal treadmill running in hot (30°C) conditions with and without the application of the cooling collars investigated.

3.5.2.6. The specifications of the cooling collar investigated

The cooling section of the modified Black Ice cooling collar was made from a thin plastic casing consisting of five compartments which were drained of the Black Ice cooling reagent and filled with 120g of gel refrigerant (BDH Laboratory Supplies, Poole, Dorset, England). The cooling section of the collar was held in place by a 600mm neoprene wrap secured with Velcro fastenings at the anterior aspect of the neck. The dimensions of the cooling section of the collar were 375mm (L) x 60mm (W) x 15mm (D) and it weighed 155g when dry at room temperature. For the collar cold trials in Chapters 4, 5, 6 and 8 the collar was frozen for 24 - 28h in a freezer at -80°C and was then left in ambient conditions (for 5min in Chapters 4 and 5 and 10min in Chapters 6, 7 and 8) before being cleared of any surface frost prior to application. For collar trials in Chapter 4, the collar was kept at ambient laboratory temperature for at least 24h prior to application. For the collar cold replaced trials in Chapter 7 the collar was initially applied as per collar cold trials and replaced, having been frozen at -20°C and left for 5 min in ambient conditions prior to application, at 30 and 60 min.

3.6. Blood treatment and storage

3.6.1. Blood sample collection, treatment, storage and analysis

Venous blood samples were taken via an indwelling cannula (Venflon, Becton Dickinson U.K. Ltd, Oxford, England, U.K.) inserted by a trained phlebotomist into an antecubital vein. During the experiments indwelling cannulas were kept patent by an injection of saline (5 ml) after each sample. Prior to each sample 2ml was drawn to clear the cannula and tubing before 5 ml aliquots were drawn and dispensed immediately after sampling into appropriate tubes. For serum samples the aliquots were dispensed into plain, anticoagulant-free, tubes (Sarstedt Ltd, Leicester, England, U.K.) and allowed to clot for 30 - 60min at room temperature prior to centrifuging. For plasma samples the aliquots were dispensed into K_3-EDTA tubes (Sarstedt Ltd, Leicester, England, U.K.). Samples were centrifuged at 4000g for 10min at 4°C. After centrifuging the supernatant was removed and then frozen at -80°C until the analyses were performed.

3.6.2. Blood sample analysis

3.6.2.1. Whole-blood lactate and glucose

Blood lactate and glucose was measured prior to centrifuging using a whole-blood analyser (Yellow Springs Instrument 2300 STAT plus, Yellow Springs Instruments Inc., Ohio, USA). The coefficient of variation for the whole-blood analyser was 3.0% and 2.1% for lactate and glucose. The coefficient of variation for whole-blood lactate and glucose was calculated from the repeat sampling of venous blood sampled from 8 volunteers. The samples were run six times within 10 min of sampling. The samples were kept on ice between sampling. The ranges from which the variation was calculated for whole blood lactate and whole blood glucose were 0.825 – 8.26 $mmol \cdot l^{-1}$ and 3.29 – 5.25 $mmol \cdot l^{-1}$ (Appendix B1).

3.6.2.2. Changes in blood, plasma and red cell volume

Changes in blood, plasma and red cell volume (BV, PV and CV respectively) were calculated from the mean haemoglobin (Hb) concentration (B-hemoglobin photometer, Hemocue AB, Angelholm, Sweden, measured in triplicate) and the mean haematocrit (Hct) (Micocentrifugation, Hawksley, Sussex, U.K., in triplicate) (Dill & Costill, 1974). The coefficient variation of the B-hemoglobin photometer was calculated in the same way as the variation of the whole blood analyser. The coefficient variation for samples analysed

within 10 min of sampling was 0.68% calculated from haemoglobin samples ranging from 12.0 to 16.5g·dL^{-1}.

Equations for the calculation of changes in blood volume, cell volume and plasma volume (Dill and Costil, 1974)

$$BV_{post} = BV_{pre}*(Hb_{pre}/Hb_{post})$$
$$CV_{post} = BV_{post}*(Hct_{post})$$
$$PV_{post} = BV_{post} - CV_{post}$$

$$\Delta BV\ \% = 100*(BV_{post} - BV_{pre})/BV_{pre}$$
$$\Delta CV\ \% = 100*(CV_{post} - CV_{pre})/CV_{pre}$$
$$\Delta PV\ \% = 100*(PV_{post} - PV_{pre})/PV_{pre}$$

BV = blood volume; CV = cell volume; PV = plasma volume; Hb = haemoglobin; Hct = haematocrit.

3.6.2.3. Enzyme-linked immunosorbent assay analysis (ELISA)

All neuroendocrinological analysis was conducted within 12-months of the sample collection via enzyme-linked immunosorbent assay. Haemolysed blood samples were not used for the analysis. Data from individual participants were analysed on the same plate for each ELISA. The intra-assay coefficient of variation for each assay was calculated from 8-15 pipetted wells. The variation of each assay is reported within the relevant section of each experimental Chapter. To accurately reflect the actual exposure of the target tissues to the hormones, concentrations are reported as measured values not corrected for plasma volume shifts (Judelson *et al.*, 2008).

3.7. Statistical analysis

A variety of statistical procedures were used to analyse the data presented in the experimental Chapters contained within this thesis.

The majority of data were either interval (*e.g.* temperature) or ratio (*e.g.* distance) and therefore parametric. Ordinal data were also collected (TS and RPE) but analysed using parametric tests as per standard practice. Data were initially checked for normality via

histogram plotting, measurement of skewness and kurtosis and the Kolomogov-Smirnov test. A Z-score of less than 1.96 was considered normally distributed at the $P < 0.05$ level of significance for skewness and kurtosis data. Student *t*-tests were used to compare the means of two normally distributed data sets whereas one, two and three-way repeated measures analysis of variance were used to compare the means of three or more data sets. Tukey's Honestly Significant Difference (HSD) tests were conducted *post hoc* after establishing a significant difference. Tukey's HSD was used to determine pairwise differences. The variation of ratio data (exercise performance, exercise capacity and assay precision) was calculated via the coefficient of variation. Relationships between pairs of variables were assessed via the Pearson's product moment correlation. The relationship was considered moderate if $r = 0.3 - 0.5$, high if $r = 0.5 - 0.7$ and very high if above 0.7 (Cohen, 1988). All statistical analysis was conducted using either SPSS v.15 (SPSS Inc., Chicago, IL, USA) or Statistica v.8 (StatSoft Ltd, Tulsa, OK, USA). The specific statistical analysis procedures used within each study is presented in the methods section of each Chapter. Data are presented as means and one standard deviation throughout and are based upon the population stated. The level of significance was set at $P < 0.05$ throughout.

Chapter 4: The effect of ambient temperature on the reliability of a preloaded treadmill time-trial

4.1. Introduction

Exercise performance is one of the most commonly reported measures in sports physiology research. When exercise performance is measured it is imperative that researchers use tests with high levels of reliability in order to be able to detect meaningful changes and to ensure that any changes are due to the intervention rather than measurement error or inter-individual differences (Atkinson & Nevill, 2001; Hopkins *et al.*, 2001); however, there is much debate over the best method to use to measure performance.

Many investigators have used the traditional exercise capacity test during which participants exercise at a set percentage of maximal oxygen uptake ($\dot{V}O_{2max}$) or maximal workload to volitional fatigue or "exhaustion". Tests to exhaustion (T_{exh}) have regularly been used to investigate basic physiological mechanisms during steady-state activity; however, they are not directly applicable to sporting settings and assess exercise capacity rather than exercise performance. In addition, T_{exh} have generally been shown to be unreliable methods of endurance assessment (Jeukendrup *et al.*, 1996). Within-individual variation affects the precision of estimates of change and is, therefore, the most important type of reliability measure for researchers, coaches, physicians and other professions in which the monitoring of a change in performance or health is a fundamental aspect of the role (Hopkins, 2000). The are a number of statistical approaches that can be adopted to represent within-individual error although three that are commonly used are the re-test correlation, the limits of agreement (LoA) and the coefficient of variation (CV). The test re-test correlation is a measure of relative, rather than absolute, reliability and is sensitive to the heterogeneity of values between participants (Hopkins, 2000). Due to this sensitivity the results obtained depend greatly on the range of values in the sample (Atkinson & Nevill, 1998). For example, the re-test correlation coefficient could be artificially low (representing poor reliability) in a test with a low CV (representing good reliability) if the athletes were all of a similar ability or it could be artificially high (representing good reliability) in a test with a high CV (representing poor reliability) if the athletes possess a wide range of abilities (Hopkins *et al.*, 1999). Due to the potential for inaccurate interpretation of test reliability when using the test re-test correlation tests of absolute reliability, such as LoA and CV are favoured. LoA (also known as the coefficient of repeatability when applied to measurement error) calculates the range within which the individual difference scores would fall 95% of the time (Bland & Altman, 1999). It is a test designed to be used with large data sets with a tendency for statistical bias in tests with low degrees of freedom (Hopkins, 2000) and is therefore, an unsuitable statistical method to

use in such circumstances. It has also been argued that the 95% limit of agreement is too stringent for a decision limit for performance tests geared towards elite performers and is therefore too conservative for the majority of performance assessment tests (Hopkins, 2000). Due to these problems many sports investigations use the CV to quantify the reliability of a performance test. The CV is a measure of the typical error of the measurement expressed as a percentage of their respective mean. As with test re-test correlation and limits of agreement it is a dimensionless measure which allows for extrapolation to new individuals (Atkinson & Nevill, 1998) and comparisons between studies of differing designs (Hopkins *et al.*, 2001). Unlike LOA it is not affected by statistical bias in studies with fewer degrees of freedom and is not too conservative for investigations in which performance is a key outcome variable (Hopkins, 2000). Many studies have shown that T_{exh} have poor test-retest reliability and high levels of within-subject variation during cycling (Graham & McLellan, 1989; Jeukendrup *et al.*, 1996; Krebs & Powers, 1989; McLellan *et al.*, 1995), running (Billat *et al.*, 1994) and swimming (Alberty *et al.*, 2006). Due to the high CVs reported (6.5 – 26.6%) and the lack of ecological validity associated with T_{exh} protocols researchers have moved from exercise capacity to exercise performance tests and have looked at the reliability of such.

Simulated time-trials (TT) have been shown to be reliable testing procedures for many sports, including cycling (Bishop, 1997; Hickey *et al.*, 1992; Palmer *et al.*, 1996; Smith *et al.*, 2001), running (Schabort *et al.*, 1998), swimming (Alberty *et al.*, 2006) and rowing (Schabort *et al.*, 1999) with CVs of 0.56-2.7% reported in these studies. TTs have also been shown to be more reliable than T_{exh} in studies that have directly compared the two methods of endurance assessment for cycling (Jeukendrup *et al.*, 1996) and treadmill running (Laursen *et al.*, 2006).

Variations on the TT protocol have included the addition of sport-specific sprints within the protocol (Marino *et al.*, 2002; Schabort *et al.*, 1998) and the addition of a submaximal exercise bout, or "preload", immediately preceding a TT (Doyle & Martinez, 1998; Jeukendrup *et al.*, 1996; Russell *et al.*, 2004). The preload is designed to better replicate the demands of prolonged exercise than TTs, which tend to be shorter in duration ($\leq$ 60 min) and preloaded time-trials (TT_{pre}) have been shown to be reliable tests of endurance performance. A marginally larger range of CVs (0.54-4%) have been reported for TT_{pre} during cycling (Jeukendrup *et al.*, 1996) and treadmill running (Doyle & Martinez, 1998; Russell *et al.*, 2004) compared to fixed time or distance versions, although they have been used to assess longer exercise bouts and the CVs reported are still well below those

reported for T_{exh}. The mean CVs T_{exh}, TT and TT_{pre} calculated from the cited literature are shown in Figure 4.1.

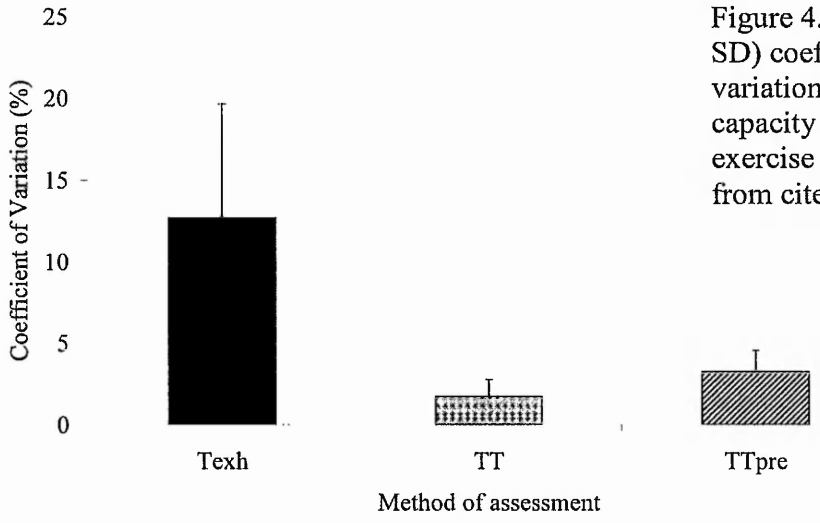

Figure 4.1. The mean (± SD) coefficient of variation reported for capacity and performance exercise tests. [calculated from cited literature]

Only six of the reliability studies cited stated the environmental conditions in which the trials took place and all but one of those conducted their trials in ambient conditions of approximately 20°C and 50% relative humidity (rh). It is well documented that endurance performance is impaired in hot, compared to temperate, environments during T_{exh} (Febbraio *et al.*, 1994; Galloway & Maughan, 1997; Nielsen *et al.*, 1993) and time-trial protocols (Altareki *et al.*, 2006). Many competitive sporting events take place during the warmer periods of the year (*e.g.* Summer Olympics) when temperatures are in excess of the typical laboratory temperatures reported (~20°C); however, to the author's knowledge, only one study has investigated the effect of elevated ambient temperature on the reliability of an endurance assessment protocol (Marino *et al.*, 2002). Marino *et al.* (2002) studied the reproducibility of a 1h cycling TT with the addition of six 1 min sprints at 10 min intervals in high ambient conditions (33°C, 63 ± 2% RH). Marino *et al.* (2002) reported a CV for the two experimental trials of 1.34%, similar to the CV reported for a cycling protocol of similar distance conducted in temperate conditions (Palmer *et al.*, 1996), and showed no significant difference between the distances cycled between the two main trials.

The data of Marino *et al.* (2002) suggests that TT are reliable methods of prolonged cycling performance in elevated ambient conditions; however, no studies have directly investigated this and none have looked at the reliability of treadmill performance in

elevated ambient conditions. The aim of this study was to investigate the reliability of a TT_{pre} treadmill test in moderate (MOD) (~14°C) and hot (HOT) (~30°C) ambient conditions. Due to the impaired performance observed in hot conditions it was hypothesised that the participants would cover less distance in the time-trial phase in the hot trials and that the reliability of the protocol would be enhanced as a result of the reduced potential for variation.

4.2. Methods

4.2.1. Participants

Nine healthy, endurance trained males volunteered for the study. The mean (± 1 SD) age, body mass (BM) and maximal oxygen uptake ($\dot{V}O_{2max}$) of the participants was 32 ± 8 yr, 72.8 ± 6.1 kg and 57.75 ± 6.43 $ml \cdot kg^{-1} \cdot min^{-1}$. The participants were fully informed of any risks and discomforts associated with the study before giving their informed written consent to participate and completing a health screen. The health screening was repeated prior to each laboratory visit to ensure the health status of the runners. The study was approved by the institution's Ethical Advisory Committee.

4.2.2. Experimental procedures

Prior to the main trials participants completed an incremental motorised treadmill test in which they ran at a fixed velocity with an increase in the treadmill gradient of $1\% \cdot min^{-1}$ to determine $\dot{V}O_{2max}$ (Jones & Doust, 1996). Following this test, participants visited the laboratory on six occasions for the main trials. For 8 of the participants each main trial was separated by 7 days. Due to mechanical problems MOD 2 and 3 for one volunteer were separated by 14 days. Trials were conducted at the same time of the day on each occasion ± 30 min. Participants completed three HOT trials (30.4 ± 0.1°C; 53 ± 2% rh) and three MOD trials (14.4 ± 0.1°C; 59 ± 4% rh). The environmental conditions were consecutive (*e.g.* three HOT trials separated by 7 days followed by three MOD trials separated by 7 days) but the trial order of the environmental condition was randomised.

During each trial participants ran at a speed calculated to elicit 60% of their individual $\dot{V}O_{2max}$ for 75 min immediately followed by a self-paced 15 min TT on a motorised treadmill (Powerjog, Sport Engineering Ltd., Birmingham, England, U.K. or Woodway ELG 55, Weil am Rhein, Germany), which was standardised for each runner for each trial.

No indication of distance ran or running speed was given throughout the preload phase of the trials. The only indication of the time elapsed given was provided during the 15 min TT when participants were able to see a countdown timer placed outside the direct line of vision. During the TT participants were encouraged to run as far as they could in the 15 min and were able to increase or decrease the running speed manually as desired, although they were unaware of the speeds at which they were running. Participants were unaware of the focus of the study, believing that the investigation was looking between, rather than within, conditions until they had completed all six trials when a full de-briefing was given. The level of verbal encouragement given and any background music listened to was the same for each trial. Water was allowed *ad libitum* throughout the trials and the volume consumed was recorded.

Participants completed a food record and physical activity diary and adopted the same diet for 24h (refraining from alcohol and caffeine), while abstaining from strenuous exercise for 24h, prior to each main trial. Participants arrived at the laboratory ~30 min before the commencement of the trial in a fasted ($\geq$ 10 h postprandial) state. On arrival, nude BM was recorded. A rectal probe (Grant Instruments (Cambridge) Ltd, England, U.K.) was self-inserted ~10cm past the anal sphincter and a heart rate (HR) monitor (Polar Electro Oy, Kempele, Finland) was attached prior to entering the environmental chamber (Design Environmental WIR52-20HS, Design Environmental Ltd., Gwent, Wales, U.K.). Participants rested inside the environmental chamber for ~10 min prior to the commencement of the trial during which time resting values for HR, rectal temperature (T_{rectal}) and thermal sensation (TS) were obtained. TS was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987).

During the trials, HR, T_{rectal}, rating of perceived exertion (RPE) (27) and TS were recorded at 5 min intervals whilst oxygen uptake ($\dot{V}O_2$) was measured every 15 min via the Douglas bag method. Capillary blood samples were taken while the participants were in a standing position for analysis of blood glucose and lactate (Yellow Springs Instrument 2300 STAT plus, Yellow Springs Instruments Inc., Ohio, USA) immediately before and after each trial. Distance ran during the 75 min preload phase was recorded as was the distance ran during the subsequent 15 min TT and the total distance covered during the 90 min trial. Following the completion of each trial participants towel-dried and recorded a post-exercise BM from which sweat loss and percentage BM loss was calculated taking into account voluntary fluid consumption.

4.2.3. Statistical analysis

Descriptive data are presented as mean ± one standard deviation (1 SD). Two-way (condition x trial) repeated-measures ANOVA was used to establish differences between and within conditions for distances ran. Three-way (condition x trial x time) repeated-measures ANOVA was used to identify differences between and within conditions for the physiological and perceptual variables. To investigate any potential acclimation effect one-way repeated-measures ANOVA was used on the distances ran and the resting core temperature observed in the hot trials. Individual CVs were calculated for each participant between pairs of trials in the same environmental conditions (e.g. between HOT1 and HOT2, HOT2 and HOT3 *etc*) and averaged to obtain a mean CV for each pair of trials. The difference between pairings was established using a paired, two-tailed t-test. For physiological and perceptual data trial 1 was considered a habituation trial and excluded from analysis. Following a significant F value, Tukey's HSD *post hoc* tests were conducted to identify pairwise differences. Significance was set at the $P < 0.05$ level.

4.3. Results

4.3.1. Running distances

4.3.1.1. Preload-phase

During the 75 min preload participants ran at an mean speed of 9.4 ± 1.3km·h^{-1}, eliciting 57.8 ± 4.8 and 58.4 ± 6.4 % $\dot{V}O_{2max}$ ($P = 0.875$) in HOT and MOD conditions respectively.

4.3.1.2. Total distance ran and time-trial performance

Participants ran significantly further in MOD compared to HOT during the 15 min TT phase ($P = 0.017$) (tables 4.1 and 4.2). The reliability of the 15 min TT section was significantly improved when trial 1 was considered a habituation session and the CV was calculated between trials 2 and 3 rather than trials 1 and 2 in HOT ($P = 0.045$) but not in MOD ($P = 0.534$). The distances ran and the CVs for the 15 min TT phase for all paired trials are shown in table 4.1 (HOT) and table 4.2 (MOD). There was no significant difference between the distances ran in the three time-trials conducted in the hot conditions ($P = 0.294$).

Table 4.1. Individual and mean time-trial performance in hot (30.4 ± 0.1°C; 53 ± 2% rh) conditions

Participant	1	2	3	4	5	6	7	8	9	Mean (1 SD)
TT1	2779	2903	3132	3160	3235	4171	2531	2765	3076	3085 (467)
TT2	2492	3016	3384	3470	3016	4081	3007	2976	3354	3200 (442)
TT3	2442	2817	3286	3715	3036	4164	3057	2810	3563	3210 (531)
CV TT1/2 (%)	7.7	2.7	5.5	6.6	5.0	1.5	12.2	5.2	6.1	5.8
CV TT2/3 (%)	1.4	4.8	2.1	4.8	0.5	1.4	1.2	4.1	4.3	2.7

Values represent the distances covered (m) in the 15 min TT stage of each trial. Mean CV (%) between TT1 and TT2 (TT1/2), TT1 and TT3 (TT1/3) and TT2 and TT3 (TT2/3) was calculated as the SD/mean * 100 from individual distances

Table 4.2. Individual and mean time-trial performance in moderate (14.4 ± 0.1°C; 59 ± 4% rh) conditions

Participant	1	2	3	4	5	6	7	8	9	Mean (1 SD)
TT1	2928	3407	3458	3611	3244	4298	3467	3377	3551	3482 (365)
TT2	2902	3754	3281	3296	3395	4435	3334	3497	3601	3499 (423)
TT3	2909	3624	3439	3745	3325	4430	3390	3513	3789	3574 (414)
CV TT1/2 (%)	0.6	6.9	3.7	6.4	3.2	2.2	2.8	2.5	1.0	3.3
CV TT2/3 (%)	0.2	2.5	3.3	9.0	1.5	0.1	1.2	0.3	3.6	2.4

Values represent the distances covered (m) in the 15 min TT stage of each trial. Mean CV (%) between TT1 and TT2 (TT1/2), TT1 and TT3 (TT1/3) and TT2 and TT3 (TT2/3) was calculated as the SD/mean * 100 from individual distances

4.3.2. Physiological variables

4.3.2.1. Heart rate

Heart rate significantly increased over time ($P < 0.001$). There were no significant differences in the heart rate response between conditions ($P = 0.117$) or trials ($P = 0.481$) however there were significant interaction effects for condition x time ($P < 0.001$) and trial x time ($P = 0.024$). There was no significant difference in the resting heart rate observed in the three hot trials ($P = 0.769$) (Figure 4.2).

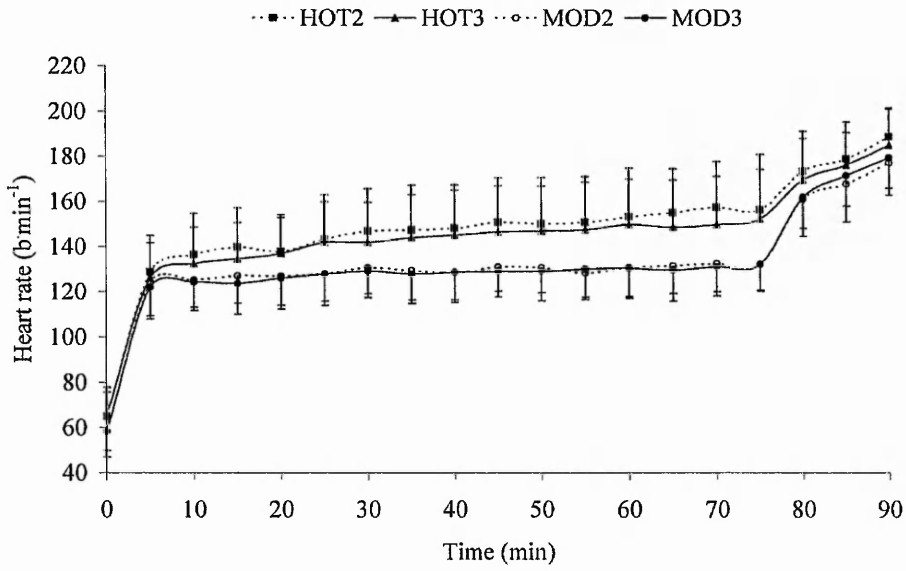

Figure. 4.2. The mean (± 1 SD) heart rate observed during the second and third hot and moderate trials. Main effect of time ($P < 0.001$), condition x time interaction ($P < 0.001$) and trial x time interaction ($P = 0.024$).

4.3.2.2. Rectal temperature

Rectal temperature significantly increased over time ($P < 0.001$). There were no main effect differences between conditions ($P = 0.231$) or trials ($P = 0.490$) however there were significant interaction effects for condition x time ($P < 0.001$), trial x time ($P = 0.002$) and condition x time x trial ($P < 0.001$). There was no difference in the resting core temperature in the three hot trials ($P = 0.128$) (Figure 4.3).

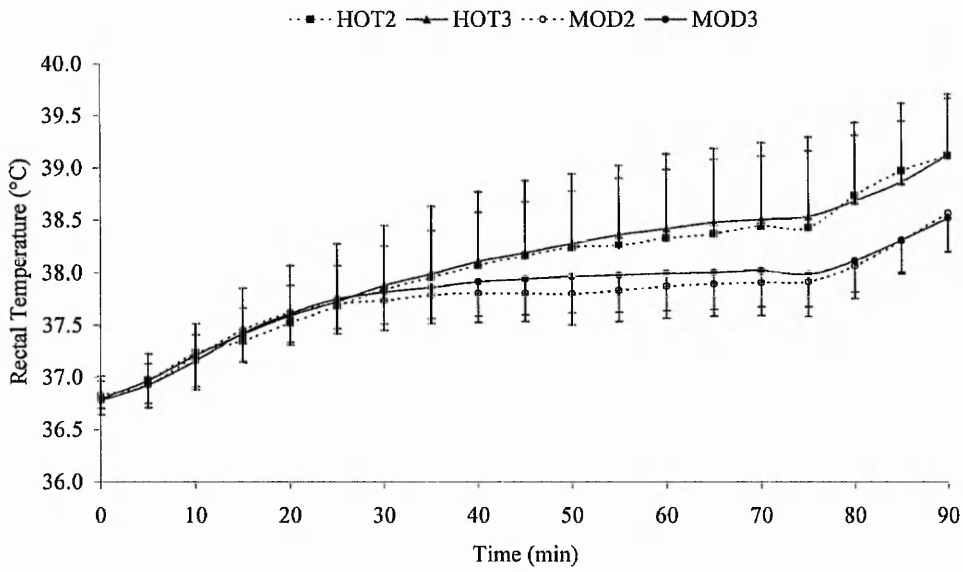

Figure. 4.3. The mean (± 1 SD) rectal temperature observed during the second and third hot and moderate trials. Main effect time ($P < 0.001$), condition x time interaction ($P < 0.001$), trial x time interaction ($P = 0.002$) and condition x time x trial interaction ($P < 0.001$).

4.3.3. Perceptual responses

Rating of perceived exertion (Figure 4.4) and thermal sensation (Figure 4.5) both increased over time ($P < 0.001$). The rating of perceived exertion was significantly different between trials ($P = 0.02$) being higher in the 2nd trial than the 3rd ($P = 0.027$). There was no significant difference between conditions ($P = 0.086$). Thermal sensation was significantly different between conditions ($P = 0.001$) and post hoc analysis revealed that it was lower in the moderate trials ($P = 0.001$). There was a significant interaction effect for trial x time ($P = 0.042$).

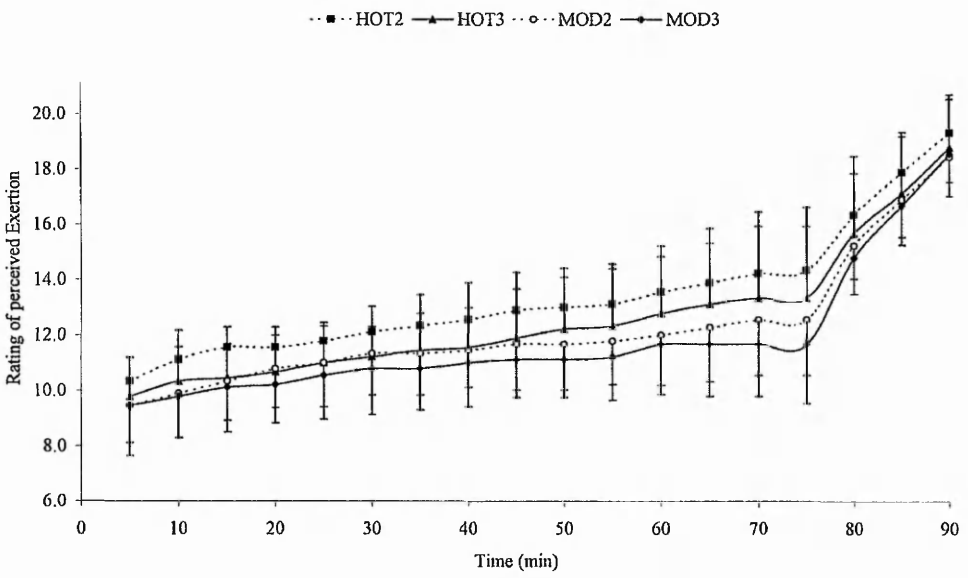

Figure. 4.4. The mean (± 1 SD) rating of perceived exertion reported during the second and third hot and moderate trials. Main effect trial (P = 0.027) and time (P < 0.001).

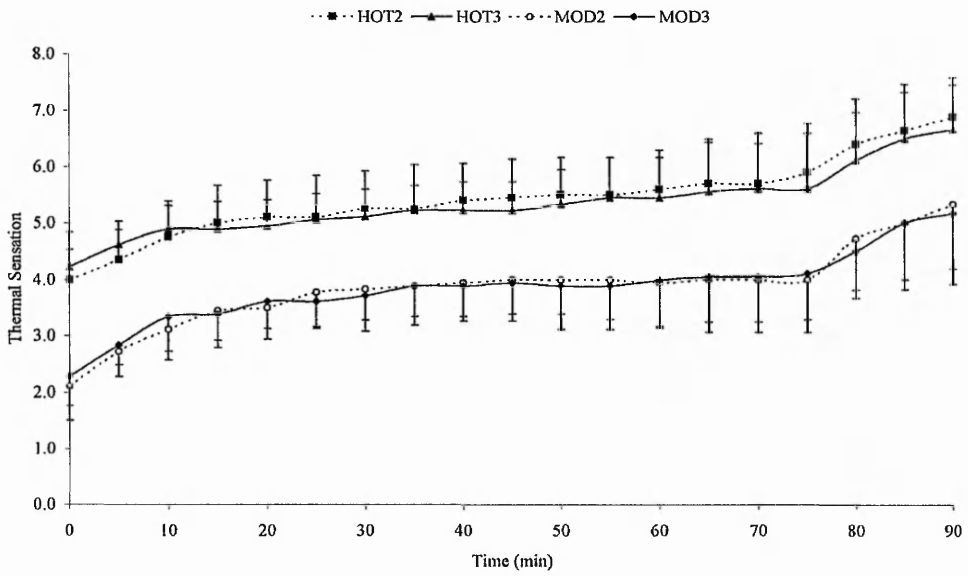

Figure. 4.5. The mean (± 1 SD) thermal sensation reported during the second and third hot and moderate trials. Main effect condition (P = 0.001), time (P < 0.001) and trial x time interaction (P = 0.042).

4.3.4. Blood data

There were no significant differences between conditions in the levels of blood lactate (MOD: 1.04 ± 0.33; HOT: 0.98 ± 0.40 $mmol \cdot l^{-1}$) and blood glucose (MOD: 4.42 ± 0.46; HOT: 5.16 ± 1.36 $mmol \cdot l^{-1}$) at rest (P = 0.598). At fatigue there was also no significant difference between the conditions for blood lactate (MOD: 6.00 ± 2.77; HOT: 4.58 ± 1.89 $mmol \cdot l^{-1}$) or blood glucose concentrations (MOD: 6.20 ± 1.41; HOT: 5.04 ± 1.57 $mmol \cdot l^{-1}$) (P = 0.487).

4.3.5. Body fluid balance

There were no significant differences in the percentage BM lost (HOT, 1.57 ± 0.81%; MOD, 1.28 ± 1.48%, P = 0.284) or in the mean sweat losses observed between HOT (1,853 ± 463 ml) and MOD (1,249 ± 1,039 ml) (P = 0.321) trials. There was a significantly greater voluntary water consumption in the heat (703 ± 403 v 318 ± 312 ml; P = 0.008).

4.4. Discussion

The main finding of the current study is that a 90 min TT_{pre} performed on a motorised treadmill is a reproducible test of endurance performance for well-trained, familiarised male runners in hot and moderate ambient conditions. The adoption of a reliable test is crucial for the effective assessment of a given intervention (*e.g.* a training programme, dietary manipulation) and the low mean CVs reported of 2.4 (MOD) and 2.7% (HOT) for the 15 min TT section demonstrate that any performance changes can be accurately tracked using the protocol.

The CVs shown are comparable to those observed in the similar running TT_{pre} trials (Doyle & Martinez, 1998; Russell *et al.*, 2004). Russell *et al.* (2004) investigated the reliability of a 10km TT following a 90 min constant intensity preload in well-trained males and females. When computing the CV for the entire TT_{pre} protocol they reported a CV of 1% when sexes were combined, which improved to 0.54% when the reliability was calculated for males alone. If the CV is calculated for the entire 90 min TT_{pre} in the current study values of 0.7% are shown for MOD and HOT conditions and so are similar to those previously reported. The CV of 4.4% reported for the TT_{pre} of Doyle and Martinez (1998) (~70% $\dot{V}O_{2max}$ followed by participant specific TT (mean distance 5.05km)) was calculated for the TT section alone and therefore is higher than that reported by Russell *et*

al. (2004). The CVs reported in the current study are smaller than those reported by Doyle and Martinez (1998). This may have been due to the shorter TT distance completed in the current study (mean distance 3.35km) which would inherently result in a decreased potential for variance to occur.

The CVs reported in the current study are also similar to those previously reported in TT investigations for cycling (Bishop, 1997; Jeukendrup *et al.*, 1996; Palmer *et al.*, 1996; Smith *et al.*, 2001), running (Schabort *et al.*, 1998) swimming (Alberty *et al.*, 2006) and rowing (Schabort *et al.*, 1999) and far lower than those reported for T_{exh} for cycling (Graham & McLellan, 1989; Jeukendrup *et al.*, 1996; McLellan *et al.*, 1995), running (Billat *et al.*, 1994) and swimming (Alberty *et al.*, 2006).

Familiarisation with the protocol has been shown to be an important determinant of the reliability of the test used. The TT studies in which the lowest CVs have been reported were conducted in swimming (Alberty *et al.*, 2006) and rowing (Schabort *et al.*, 1999) using protocols commonly used during training and competition in these sports. Doyle and Martinez (1998) reported that when classifying trial one as a habituation trial, and excluding it from the statistical analysis, the mean CV for their treadmill-based test was reduced from 10.1% to 4.4%. Other reliability studies (Marino *et al.*, 2002; McLellan *et al.*, 1995; Schabort *et al.*, 1999) have also reported a better reliability for their protocol when excluding the first trial (and the last in McLellan *et al.* 1995) from statistical; however, Currell *et al.* (2006) reported no learning effect in their cycling TT. The authors suggested that this may have been due to the highly trained, elite nature of their participants (Currell *et al.*, 2006). The current findings suggest that a habituation trial is essential when conducting experiments in unfamiliar conditions, as shown by the enhanced reliability of the test in HOT when trial one was considered a habituation trial, however they support the suggestion that one may not be required when conducting experiments in familiar conditions (*e.g.* MOD) with well-trained, experienced participants.

One potential problem associated with conducting repeated trials in the heat, from a controlled experimental viewpoint, is the potential for acclimation to occur. Heat acclimation decreases HR, T_{rectal}, skin temperature (T_{skin}) and increases sweat response during exercise in the heat (Pandolf *et al.*, 1988) and so can influence the data collected. Barnett and Maughan (1993) investigated the effect of three trials in the heat (34.6 ± 0.6°C, 60 ± 7% rh), conducted at weekly intervals, on human acclimation. They showed no differences in HR, T_{rectal}, T_{skin} or total sweat loss between the trials. The lack of any

significant differences between the T_{rectal}, HR and sweat loss data collected from the three trials in HOT in the current study supports the suggestion that trials conducted in the heat with individuals from temperate climates can be conducted at weekly intervals without heat acclimation occurring (Barnett & Maughan, 1993).

4.5. Conclusion

The TT_{pre} investigated in the current study is a reliable test of running performance in both hot and moderate conditions. The need for a habituation trial may depend upon the familiarity of the participant with the experimental conditions and protocols. When conducting trials in hot conditions weekly intervals between trials are sufficient to prevent acclimation occurring.

Chapter 5: Practical neck cooling improves preloaded time-trial running performance in the heat

5.1. Introduction

As reported in the experimental study presented in the previous Chapter, 15 min time-trial performance is impaired in hot compared to moderate conditions by ~10%. The reason for the impairment is not fully understood, although the impairment is often attributed to the development of hyperthermia. It has been suggested that there is a core temperature of ~40°C above which voluntary exercise is terminated (Nielsen *et al.*, 1993); however, it is more likely that it is the development of a high hypothalamic, rather than body, temperature that ultimately limits exercise (Caputa *et al.*, 1986). The observation that exercise appears to be terminated due to the onset of hyperthermia has led many researchers to investigate a variety of cooling interventions administered prior to, and during, exercise in an attempt to offset the reduction in performance observed during exercise in the heat.

Whole-body cooling prior to exercise (pre-cooling) has received significant attention and has been shown to enhance both exercise performance (Booth *et al.*, 1997) and capacity (Lee & Haymes, 1995) in hot conditions; however, its practical application has been questioned and the need for more viable alternatives has recently been highlighted (Quod *et al.*, 2006).

Cooling the head and neck can be achieved without causing excessive disruption to the individual or their sporting attire. Due to the neck's close proximity to the thermoregulatory centre, cooling this region may be more effective than cooling elsewhere (Shvartz, 1970). Cooling the head and neck region has been shown to be more efficient than cooling the torso in the alleviation of heat strain during exposure to high ambient temperatures (~40°C) (Shvartz, 1976). Despite this, relatively few studies have investigated the effect of cooling the neck region during exercise.

It has been demonstrated that if the magnitude of cooling provided is sufficient and/or the thermal strain is severe enough cooling the head, neck and face region can elicit beneficial thermoregulatory (Gordon *et al.*, 1990; Shvartz, 1970), cardiovascular (Shvartz, 1970) and perceptual (Mundel *et al.*, 2006; Simmons *et al.*, 2008) adjustments; however, commercially available neck and head cooling mechanisms rarely provide sufficient cooling (Bulbulian *et al.*, 1999; Desruelle & Candas, 2000). Cooling the whole head via water-misting improves exercise capacity in the heat by approximately 51% (Ansley *et al.*, 2007) while cooling via a water-perfused hood can improve 15 min time-trial performance

by 3.3% (Palmer *et al.*, 2001). Palmer *et al.* (2001) reported that cooling the head via a cooling hood during 60 min of seated rest, a 30 min submaximal preload and a 15 min time-trial improved running performance significantly compared to no-cooling or cooling at rest only. The cooling-intervention lowered rectal temperature and improved thermal comfort but had no effect on the heart rate recorded. In contrast, Ansley *et al.* (2007) reported that head cooling had no effect on any of the physiological variables associated with peripheral fatigue; however, the cooling significantly attenuated the prolactin response to exercise (a finding consistently reported in the literature (Brisson *et al.*, 1987; Mundel *et al.*, 2006)) and suggested this supported the ideal of a central component of fatigue in the heat. A variety of hormones and neurotransmitters have been proposed as mediators of exercise performance and adherence as well as markers of stress and central fatigue. The activity of cerebral serotoninergic and dopaminergic pathways has been linked to feelings of arousal and motivation; however, due to practical limitations that prevent their measurement, prolactin is often used as a surrogate marker. Prolactin is often used as such a surrogate because its secretion is stimulated by serotonergic and inhibited by dopaminergic activity within the hypothalamus (Bridge *et al.*, 2003b; Freeman *et al.*, 2000). Many of the hormones and neurotransmitters linked to the regulation of exercise performance and fatigue have been shown to increase in concentrations when exposed to a stressor such as exercise or heat (Bridge *et al.*, 2003a). Cooling the head and neck region has been shown to decrease both the physical and perceptual stress of exercising in hot conditions (Ansley *et al.*, 2007; Nunneley *et al.*, 1971) and therefore it seems prudent to suggest that cooling during exercise may have an effect on the response of a number of these hormones and neurotransmitters to exercise in hot conditions, although only prolactin has been investigated to date.

The aim of the present study is to investigate the effect of a practical neck-cooling device on preloaded time-trial performance in hot conditions and on the physiological, perceptual and neuroendocrinological responses to the exercise bout. It was hypothesised that cooling the neck would enhance performance by positively affecting the perceptual and hormonal response to the bout.

5.2. Methods

5.2.1. Participants

Eleven healthy, trained males volunteered for the study. Due to injury two volunteers failed to complete all trials and therefore their data has been omitted. The mean (± 1 SD)

age, body mass, height and relative maximal oxygen uptake ($\dot{V}O_{2max}$) of the nine participating volunteers was 25 ± 4 y, 76.5 ± 5.9 kg, 1.81 ± 0.07 m and 54.2 ± 4.6 ml·kg^{-1}·min^{-1}. All participants were fully informed of any risks and discomforts associated with the study before giving their oral and written informed consent to participate and completing a health screen. The health screening procedure was repeated prior to each laboratory visit to ensure the health status of the participants remained the same. The study was approved by the Ethical Advisory Committee of Nottingham Trent University.

5.2.2. Experimental procedures

Prior to the main trials, participants completed an incremental motorised treadmill test to determine $\dot{V}O_{2max}$, as per Jones and Doust (1996). Following a full habituation trial, they visited the laboratory on three occasions for the main experimental trials. The trials were conducted in a randomised and counter-balanced order. The experimental trials were conducted at the same time of the day on each occasion ± 30 min and were separated by 7-days for all participants except for number 7. Participant number 7 completed his final trial after a 1-month break (due to mechanical failure occurring during his original 'final' trial) however was re-familiarised 7-days prior to the repeated final trial. The distance covered in the re-habituation trial was within the established coefficient of variation of the protocol (2.7%) compared to the initial habituation trial and so data from participant 7 has been included. The participants were not acclimated- the mean outdoor temperature on the mornings of the main trials was 6.41 ± 3.06°C.

During the habituation and experimental trials, participants completed a 90 min preloaded time-trial (TT_{pre}) in hot conditions (30.4 ± 0.1°C; 53 ± 2% rh). The TT_{pre} consisted of 75 min of treadmill running at ~60% $\dot{V}O_{2max}$ (9.0 ± 1.1km·h^{-1}) followed by a self-paced 15-min time-trial. During the time-trial participants were able to manually increase and decrease their speed and they were instructed to cover as much distance as they could during the time. The time remaining was displayed via a countdown timer. The distances covered were not revealed until the completion of all four trials. All four trials were conducted with the runners wearing the same clothing.

During the three experimental trials, volunteers completed the TT_{pre} whilst wearing either a cold collar (CC), an uncooled collar (C) or no collar (NC). The collar was comprised of

two parts and was modified from a commercially available neck cooling device (Black Ice LLC, Lakeland, USA) (see section 3.5.2.6.).

Participants abstained from alcohol and caffeine and completed a food record for the day prior to the initial experimental trial. They adopted the same diet and abstained from strenuous exercise for 24h, prior to each main trial. Participants arrived at the laboratory ~30 min before the commencement of each trial in a fasted ($\geq$ 10h postprandial) state and having ingested 500 ml of water ~1.5h previously. On arrival, nude body mass was recorded. A rectal probe (Grant Instruments (Cambridge) Ltd, England, U.K.) was self-inserted ~10cm past the anal sphincter; a heart rate (HR) monitor (Polar Electro Oy, Kempele, Finland) was attached and an indwelling cannula (Venflon, Becton Dickinson U.K. Ltd, Oxford, U.K.) was inserted into a vein in the antecubital fossa prior to the participant entering the environmental chamber (Design Environmental WIR52-20HS, Design Environmental Ltd., Gwent, Wales, U.K.). The indwelling cannula was kept patent by an injection of saline (5 ml) after each sample. Participants rested in the environmental chamber in an upright position for 10 min, wearing the collar when appropriate (CC and C trials), after which, resting values for HR, rectal temperature (T_{rectal}), mean neck temperature (T_{neck}) and thermal sensation (TS) were obtained. Mean neck temperature was calculated as the mean temperature of four skin thermistors (Grant Instruments (Cambridge) Ltd, England, U.K.) spaced equally across the posterior aspect of the neck. Skin thermistors were attached to the sternum, forearm, thigh and calf for the calculation of weighted mean skin temperature (Ramanathan, 1964). However, due to difficulty in the fixing of the thermistors for the duration of the trial insufficient data was collected to compute mean skin temperature and therefore the data is not presented. All thermistors were attached via a transparent dressing (Tagaderm, 3M Health Care, USA) and water-proof tape (Transpore, 3M Health Care, USA). TS was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987).

During the 75 min preload phase of the trials, HR, T_{rectal}, T_{neck}, rating of perceived exertion (RPE) (Borg, 1982) and TS were recorded at 5 min intervals. Oxygen uptake was measured at 40 and 60 min via the Douglas bag method. During the time-trial phase HR, T_{rectal}, T_{neck}, RPE and TS were recorded at 2.5 min intervals allowing for closer monitoring, during the higher-demanding phase. Distances ran during the 75 min preload phase and the subsequent 15 min time-trial were recorded. The mean self-selected speed for the time-trial phase was calculated from this data. Following the completion of each trial, participants

towel-dried and recorded a dry post-exercise nude body mass from which sweat loss and the percentage change in body mass was calculated, taking into account voluntary fluid consumption during the protocol.

5.2.3. Collection and analysis of blood samples

Due to sampling difficulty with one individual the blood data presented is from 8 of the 9 volunteers who participated in the study. Blood samples were taken at 0, 10, 30, 50, 70 and 90 min. Whole blood was initially analysed in triplicate for lactate and glucose (Yellow Springs Instrument 2300 STAT plus, Yellow Springs Instruments Inc., Ohio, USA) and then aliquots were dispensed into K_3-EDTA tubes (Sarstedt Ltd, Leicester, U.K.) for the subsequent obtainment of platelet-free plasma. Samples were centrifuged at 4000g for 10 min at 4°C. After centrifuging, the supernatant was removed and then frozen at -80°C until the analyses were performed. Changes in blood, plasma and red cell volume (BV, PV and CV respectively) were calculated from the mean haemoglobin (Hb) concentration (B-hemoglobin photometer, Hemocue AB, Angelholm, Sweden, measured in triplicate) and the mean haematocrit (Hct) (Micocentrifugation, Hawksley, Sussex, U.K., in triplicate), using the methods of Dill and Costill (1974).

Plasma concentrations of prolactin, cortisol, dopamine, adrenaline, noradrenaline and serotonin were determined via enzyme-linked immunosorbent assays; prolactin and cortisol (DRG Instruments GmbH, Marburg, Germany); dopamine, adrenaline, noradrenaline and serotonin (IBL Hamburg, Hamburg, Germany). The intra-assay variation (CV) based upon laboratory work for the prolactin, cortisol, dopamine, adrenaline, noradrenaline and serotonin assays were 10.0%, 6.0%, 1.5%, 8.2%, 8.2% and 7.6% respectively.

5.2.4. Statistical analysis

Descriptive data is reported as mean ± one standard deviation (1 SD). One-way repeated-measures analysis of variance (ANOVA) tests were conducted to evaluate differences between the distances ran, sweat-loss and fluid consumption while two-way (trial x time) tests were performed to evaluate differences between trials for thermoregulatory, cardiovascular, neuroendocrinological and perceptual variables. Following a significant F value, Tukey's HSD *post hoc* tests were conducted to identify pair-wise differences. Statistical analyses were conducted for the 90 min trial and additional analyses were

conducted for 0, 75 and 90 min time-points which represented the beginning and the end of the preload and time-trial phases. Significance was set at the $P < 0.05$ level.

5.3. Results

5.3.1. Time-trial performance

During the time-trial phase participants covered 3,030 ± 485m; 2,741 ± 537m and 2,884 ± 571m in the CC, C and NC trials (main effect trial $P < 0.001$) (Figure 5.1). Significantly more distance was covered in CC compared to C ($P = 0.008$) and in CC compared to NC ($P = 0.041$). There was no significant difference in the distance covered in NC compared to C ($P = 0.079$). The time-trial performance of all participants was improved with the application of CC (mean improvement +5.9%), and impaired (mean impairment -4.8%) with C compared to NC. There was no trial order effect ($P = 0.124$).

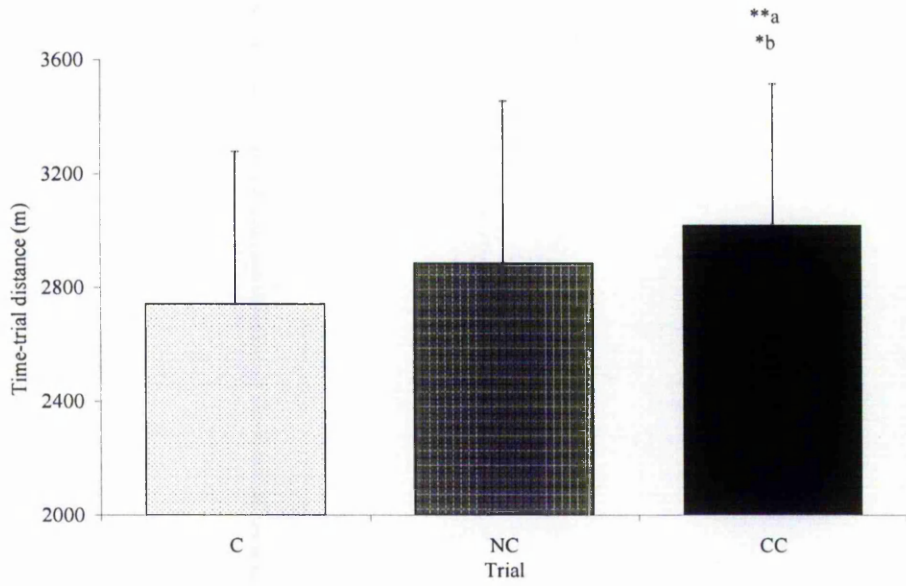

Figure 5.1. The mean distances covered during the 15 min time-trials in the no collar (NC), uncooled collar (C) and cold collar (CC) trials. * = $P < 0.05$; ** = $P < 0.01$. [a] = compared to C; [b] = compared to NC.

5.3.2. Neck temperature

Mean neck temperature (T_{neck}) is shown in Figure 5.2. There was a significant main effect for trial and time and time x trial interaction ($P < 0.001$ for all). T_{neck} was significantly

colder during the 90 min TT_{pre} in CC compared to NC ($P < 0.001$) and C ($P < 0.001$) but there was no difference between NC and C ($P = 0.449$). There was a significant interaction effect between trials ($P < 0.001$). At the commencement of the time-trial (t = 75 min) neck temperature was significantly lower in NC and CC compared to C trials ($P = 0.001$ for both); however, there was no significant difference between NC and CC ($P = 0.512$).

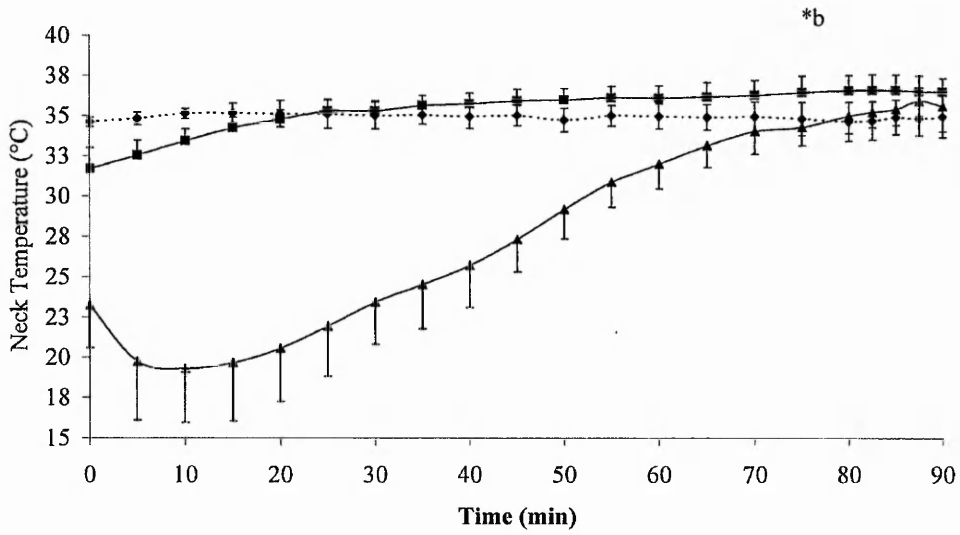

Figure 5.2. Mean neck temperature during the 90 min preloaded time-trial. ♦ = no collar (NC); ■ = uncooled collar (C); ▲ = cold collar (CC). * = $P < 0.05$; ^{b} = C significantly different compared to NC and CC. Main effect trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P < 0.001$).

5.3.3. Heart rate and rectal temperature

Heart rate and T_{rectal} increased significantly over the duration of the trial ($P < 0.001$) however there were no significant differences between trials for T_{rectal} ($P = 0.266$) or HR ($P = 0.780$). The T_{rectal} and HR at the beginning and end of the 90 min exercise bout, as well as at the beginning of the time-trial phase, are shown in Table 5.1.

Table 5.1. Rectal temperature and heart rate at 0, 75 and 90 min

Time (min)	Trial	Rectal temperature (°C)	Heart rate ($b \cdot min^{-1}$)
0	CC	36.81 ± 0.10	60 ± 13
	C	36.82 ± 0.21	68 ± 14
	NC	36.90 ± 0.29	67 ± 12
75	CC	$38.64 \pm 0.29^{**a}$	$163 \pm 13^{**a}$
	C	$38.69 \pm 0.37^{**a}$	$165 \pm 12^{**a}$
	NC	$38.78 \pm 0.37^{**a}$	$165 \pm 11^{**a}$
90	CC	$39.14 \pm 0.27^{**ab}$	$188 \pm 6^{**ab}$
	C	$39.08 \pm 0.37^{**ab}$	$185 \pm 8^{**ab}$
	NC	$39.25 \pm 0.28^{**ab}$	$186 \pm 6^{**ab}$

Values are means ± 1 SD. CC = cold collar trial; C = collar trial; NC = no collar trial. ** = P < 0.01. [a] = significant difference compared to 0-min; [b] = significant difference compared to 70-min

5.3.4. Perceptual measurements

Thermal sensation and rating of perceived exertion data are shown in Figures 5.3 and 5.4. There was a significant main effect of trial ($P < 0.001$), and time ($P < 0.001$) and interaction ($P = 0.11$) for thermal sensation and main effects for trial ($P = 0.021$) and time ($P < 0.001$) for rating of perceived exertion. Thermal sensation and rating of perceived exertion were significantly lower in CC compared to C (TS: $P = 0.001$; RPE: $P = 0.016$). Thermal sensation was also lower in CC compared to NC ($P = 0.014$). There was a significant interaction effect for thermal sensation between trials ($P = 0.011$). There were no significant differences for the thermal sensation reported between NC and C ($P = 0.208$) or for the rating of perceived exertion reported between NC and C ($P = 0.343$) or NC and CC ($P = 0.234$). At the onset of the time-trial phase (75 min) both RPE and TS were significantly lower in CC than C ($P = 0.031$ and $P = 0.016$). There were no significant differences between the other trial pairings ($P > 0.05$).

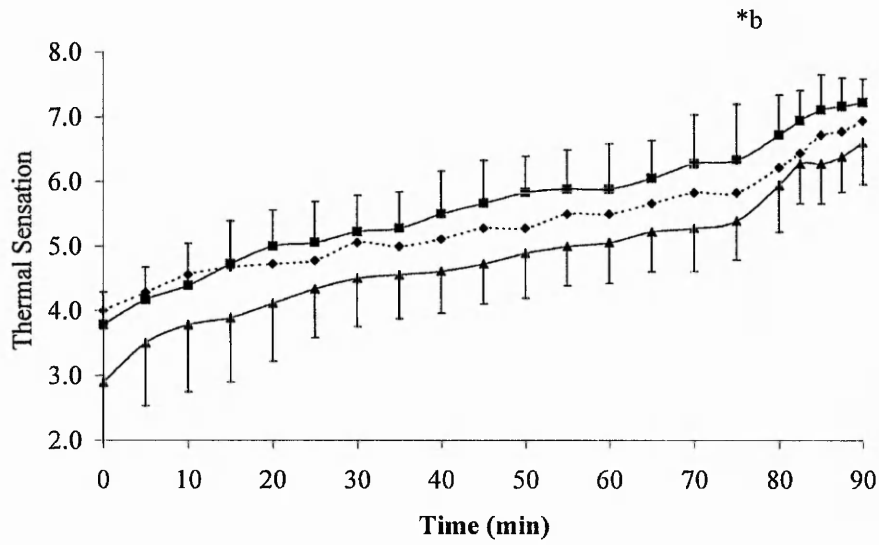

Figure 5.3. Mean (± 1 SD) thermal sensation reported during the 90 min preloaded time-trial. ♦ = no collar (NC); ■ = uncooled collar (C); ▲ = cold collar (CC). For clarity SD is not shown for NC trials, mean SD for NC = ± 0.6. * = $P < 0.05$; [b] = significant difference between C and CC. Main effect trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P = 0.11$).

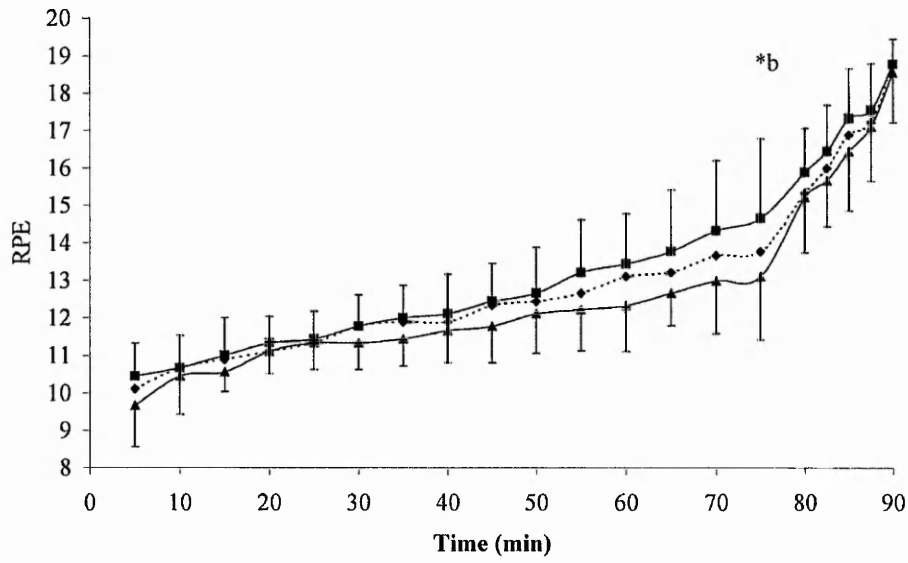

Figure 5.4. Mean (± 1 SD) rating of perceived exertion (RPE) reported during the 90 min preloaded time-trial. ♦ = No Collar (NC); ■ = Uncooled collar (C); ▲ = Collar cold (CC). For clarity SD is not shown for NC trials, mean SD for NC = ± 1.4. * = $P < 0.05$; ^{b} = significant difference between C and CC. Main effect trial ($P = 0.021$) and time ($P < 0.001$).

5.3.5. Body fluid balance

There were no significant differences in the baseline body mass of the participants ($P = 0.414$), the volume of water voluntary consumed ($P = 0.400$) or in the volume of sweat lost ($P = 0.951$) between trials. Participants consumed 0.761 ± 0.378L; 0.828 ± 0.406L and 0.749 ± 0.418L of water and lost 2.09 ± 0.33L; 2.08 ± 0.24L and 2.05 ± 0.26L of sweat during the NC, C and CC trials respectively.

5.3.6. Blood data

All measured blood variables increased significantly over time ($P < 0.001$). There were no significant main effects between trials for whole blood lactate ($P = 0.221$) or glucose ($P = 0.153$) or for plasma concentrations of cortisol ($P = 0.323$), prolactin ($P = 0.775$), adrenaline ($P = 0.744$), noradrenaline ($P = 0.323$), dopamine ($P = 0.784$) (Table 4.2) or

serotonin (P = 0.08, Figure 5.5). There was a significant difference in the interaction observed for cortisol concentrations (trial x time, P < 0.001). Wearing the cold collar in CC trials resulted in an initial elevation that was not observed in the other two conditions (Figure 5.6). There were no differences in plasma volume change between trials (P = 0.518). The mean plasma volume changes were -1.4 ± 6.4, 0.4 ± 4.7 and -2.8 ± 5.3% in NC, CC and C trials respectively.

Table 5.2. The neuroendocrinological response to the preloaded time-trial

Time (min)	Trial	Prolactin ($ng \cdot ml^{-1}$)	Adrenaline ($nmol \cdot l^{-1}$)	Noradrenaline ($nmol \cdot l^{-1}$)	Dopamine ($nmol \cdot l^{-1}$)
0	CC	5.52 ± 2.21	0.83 ± 0.43	3.10 ± 0.73	0.18 ± 0.03
	C	4.40 ± 1.85	0.70 ± 0.35	2.70 ± 0.84	0.19 ± 0.07
	NC	4.92 ± 1.85	0.82 ± 0.45	2.54 ± 0.55	0.26 ± 0.12
70	CC	8.59 ± 3.25	1.08 ± 0.23	12.77 ± 9.91^{**a}	0.92 ± 0.59
	C	8.48 ± 2.99	1.03 ± 0.34	9.92 ± 6.56^{**a}	1.62 ± 1.75
	NC	8.59 ± 3.92	1.03 ± 0.13	9.72 ± 3.71^{**a}	1.17 ± 0.36
90	CC	20.85 ± 8.24^{**ab}	1.92 ± 0.94^{**ab}	20.90 ± 7.00^{**ab}	2.91 ± 1.86^{**ab}
	C	20.77 ± 10.52^{**ab}	1.71 ± 0.54^{**ab}	18.41 ± 5.69^{**ab}	2.62 ± 1.84^{**ab}
	NC	19.71 ± 8.01^{**ab}	1.60 ± 0.39^{**ab}	24.57 ± 7.57^{**ab}	3.22 ± 2.42^{**ab}
Main effect (trial)		P = 0.775	P = 0.744	P = 0.323	P = 0.784

Values are means ± 1 SD. ** = P < 0.01. ^{a} = significant difference compared to 0 min; ^{b} = significant difference compared to 70 min

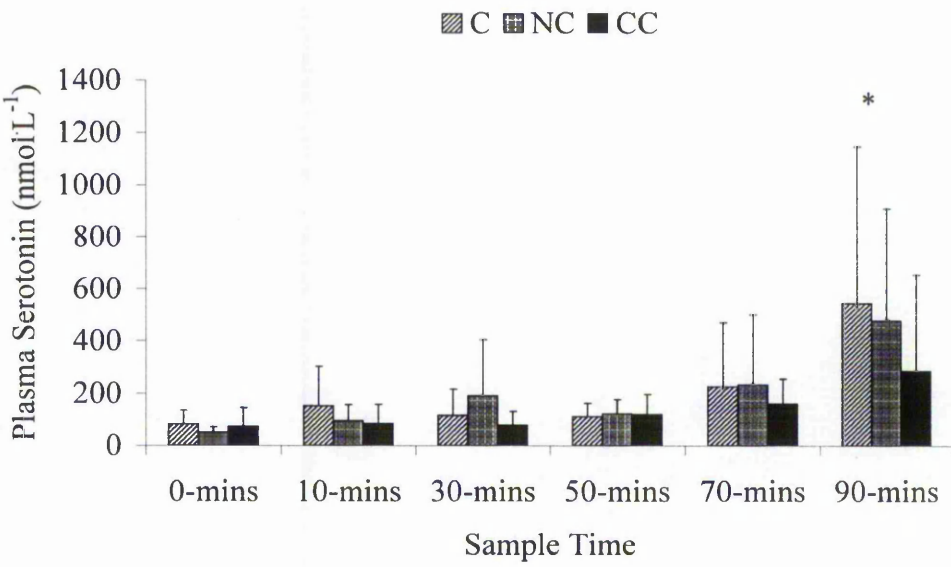

Figure 5.5. The serotonin concentrations observed during the preloaded time-trial. * significant difference from all other time-points ($P < 0.05$). Main effect time ($P < 0.001$).

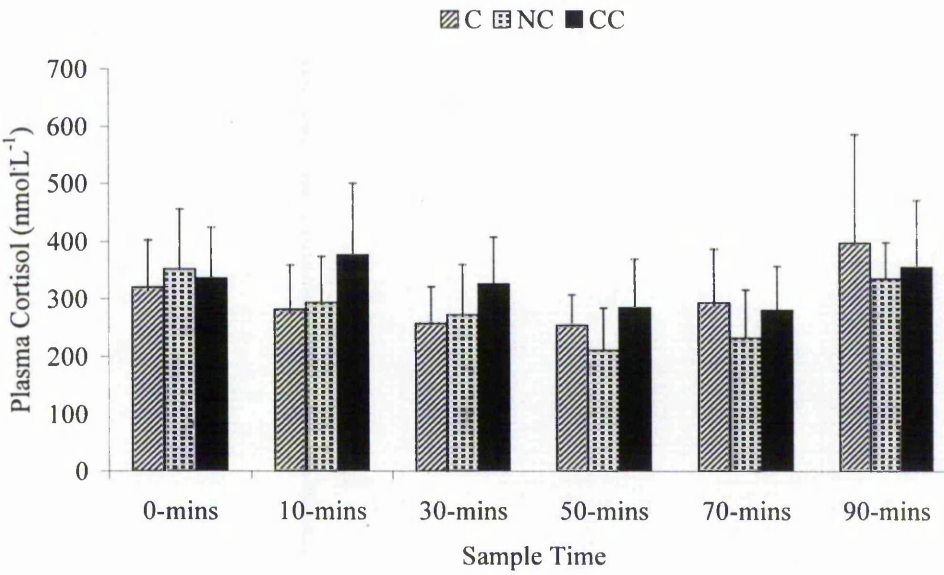

Figure 5.6. The plasma cortisol concentrations observed during the preloaded time-trial. Main effect time ($P < 0.001$) and interaction ($P < 0.001$).

5.4. Discussion

The main finding of the current study is that reducing T_{neck} can improve time-trial performance in the heat without any significant alterations in the physiological or neuroendocrinological response to the exercise bout. Palmer *et al.* (2001) reported that cooling the head via a water-perfused hood could improve 15 min time-trial performance by 3.3% but this is the first study to show that cooling the neck via a practical neck-cooling collar, which could be worn during a wide range of exercise disciplines, also has a beneficial effect upon exercise performance.

It has been suggested that exercise in the heat is limited by the attainment of a high core temperature (Nielsen *et al.*, 1993); however despite differences in time-trial performance in the current study there were no significant differences in the T_{rectal} between the three trials. The T_{rectal} and HR profiles observed in the present study match those reported using this protocol in the first study (presented in Chapter 4). The results from the current study are in line with other studies who reported that neck cooling had no effect on T_{rectal} or HR during submaximal (60% $\dot{V}O_{2max}$) cycling despite significant reductions in T_{neck} (Bulbulian *et al.*, 1999; Hamada *et al.*, 2006). The study by Hamada *et al.* (2006) had design similarities to the preload phase of the current study and showed that despite significantly lowering T_{neck} at the termination of exercise compared to the control trial (23.59 ± 2.18°C v 36.07 ± 0.12°C, $P < 0.05$), the application of the ice cooling packs had no significant effect on T_{rectal}, skin temperature, sweat loss, HR or TS during 40 min of cycle ergometer exercise at ~60% $\dot{V}O_{2max}$ in a hot environment (30 ± 1°C; 40% rh). Hamada *et al.* (2006) investigated the physiological, rather than performance, response to neck cooling. The findings of the present study support theirs but demonstrate a performance benefit in the absence of cooling-induced physiological alterations. Palmer *et al.* (2001) reported a performance-benefit following head-cooling with associated reductions in T_{rectal} but not in HR however a reduction in T_{rectal} is not a requirement for an enhanced ability to sustain exercise in the heat (Hessemer *et al.*, 1984).

It has been demonstrated that heat tolerance can be enhanced by the maintenance of a cooler brain (Carithers & Seagrave, 1976) and it has been proposed that brain temperature (T_{brain}) may be more important than core temperature in the regulation of thermal tolerance (and therefore perhaps exercise adherence) in hyperthermic conditions (Caputa *et al.*, 1986). This idea has been suggested following the observations that many animal and bird species protect the brain from excessive heat loads by maintaining T_{brain} at levels below

that of the core, via a method termed selective brain cooling (Cabanac, 1998). Brain temperature is primarily dependent on the temperature of the blood within the carotid arteries (Baker, 1982) and it has been suggested that the application of a cooling collar may be able to reduce the temperature of the carotid blood and therefore reduce T_{brain} (Caputa, 2004; Gordon *et al.*, 1990; Zhu, 2000). The measurement of human brain temperature is only possible in exceptional circumstances so mathematical modelling has been used as a surrogate method of quantifying changes in T_{brain} as a result of external cooling interventions. Zhu (2000) proposed that, theoretically, external cooling could lower the temperature of arterial blood on route to the brain, although a more recent mathematical modelling paper (Sukstanskii & Yablonskiy, 2007) reported that human T_{brain} is practically homogenous and that any alterations in T_{brain} could only occur at the surface to a depth of ~3 - 4mm (Sukstanskii & Yablonskiy, 2007). Although the collar may not reduce deep T_{brain} it is possible that the temperature of the blood reaching the peripherally-located thermoregulatory centre is reduced by the external cooling and this may result in the masking of signals regarding the temperature of the body (Brisson *et al.*, 1987). Further investigation is required to explore this proposal.

It has been suggested that during exercise, pacing strategies are adopted to allow for the completion of the task within homeostatic limits (Lambert *et al.*, 2005). This is often described as the central governor theory (Noakes & St Clair, 2004). It is unknown what stimulus this governor may act in response to but if pacing strategies are governed by an internal mechanism to prevent the overloading of the body it is possible the presentation of a cooling, or indeed heating, intervention may disrupt this mechanism by providing a false representation of the body's thermal status. It has been proposed that any false signal may be amplified by altering the temperature of the neck, rather than elsewhere, due to the neck's close proximity to the thermoregulatory centre (Shvartz, 1976) and the 6% improvement following neck-cooling in the present study compared to the 3.3% improvement observed following head-cooling (Palmer *et al.*, 2001) offer support for this idea. The amplification of a false signal could result in the selection of a 'false' or 'un-natural' pace which could potentially result in an improved or impaired sporting performance. Arngrimsson *et al.* (2004) and Booth *et al.* (1997) showed that pre-cooling resulted in an improved performance because it allowed the participants to maintain a faster mean pace during the time-trial and allowed them to further increase their pace towards the end of the bout despite a reduced effect of the pre-cooling. Although the pacing strategies adopted during the time-trial were not looked at in the current study, elevations in T_{neck} resulted in a significantly reduced self-selected mean time-trial speed,

while lowering T_{neck} allowed participants to maintain a significantly faster mean speed during the time-trial even when the cooling stimulus was reduced or no longer present. It appears that altering T_{neck} may provide a 'false' signal allowing for the selection of a pace faster than that which would be naturally selected and this may be due to an altered perception of thermal comfort.

Palmer *et al.* (2001) reported an improvement in time-trial performance and thermal sensation with head cooling and, in the present study, thermal sensation was lower (signifying that participants felt cooler) when T_{neck} was reduced at the beginning of the TT_{pre}, via the application of CC compared to NC and C. Thermal sensation remained significantly reduced at the beginning and end of the time-trial phase in CC compared to C; however, there was no difference between the TS reported at the same time-points between CC with NC. This directly matches differences in T_{neck} at the corresponding time points and shows that reducing T_{neck} consistently reduces TS. The findings of the current study are inline with those of Arngrimsson *et al.* (2004) as the beneficial effect of the cooling intervention remained even when the cooling stimulus was no longer present suggesting that the benefit may have occurred cumulatively during the preload phase of the exercise.

It appears that the perception of thermal sensation, rather than actual body temperature, may have a central role in pace selection however it is unclear by what mechanism this works. Cooling the neck improved the perception of thermal comfort but had no significant effect on any of the hormones or neurotransmitters measured in the current study despite many being often classified as "stress hormones". Plasma prolactin levels were unaffected by the cooling intervention in the current study despite facial-cooling being shown to attenuate the hyperprolacteamia often observed during exercise in the heat (Ansley *et al.*, 2007; Mundel *et al.*, 2006). The mechanisms governing the prolactin response to exercise and the effects that cooling the head and face has on reducing prolactin levels is not clear, although it has been shown that it is dependent on the magnitude of the cooling (Brisson *et al.*, 1991). The current study suggests that cooling the neck provides an insufficient magnitude of cooling to alter the prolactin response to exercise in the heat. Adrenaline, noradrenaline, serotonin and dopamine concentrations were all unaffected by the cooling collar and followed well documented patterns of change over the duration of the trial. Cortisol was elevated initially by the cold collar and there was a significant interaction effect. As previously documented adrenaline and dopamine levels were only significantly elevated during the final sampling when the intensity was greater than the 60% $\dot{V}O_{2max}$

elicited during the preload, while noradrenaline showed a progressive increase over time throughout the trial (Kotchen *et al.*, 1971).

5.5. Conclusion

Time-trial performance in the heat can be significantly enhanced by reducing T_{neck}, without significantly altering the physiological or neuroendocrinological response to the exercise bout. It seems that the running performance improvements are due to an up-regulation of pacing as a result of an alteration in perceived, rather than actual, thermal strain.

Chapter 6: The serum S100β response to practical neck cooling during exercise in the heat

6.1. Introduction

As shown in Chapter 4, running performance is impaired in hot conditions. The exact mechanism(s) that limit exercise performance and capacity in a hot environment remain unclear although the data from the previous chapter (Chapter 5) adds support for the suggestion that there is a central element to the regulation of exercise. It has been shown that hyperthermia has a profound impact on cerebral function during exercise altering brain activity (Nielsen *et al.*, 2001), reducing voluntary muscle activation (Nybo & Nielsen, 2001a) and increasing the perception of effort (Nybo & Nielsen, 2001c). Watson *et al.* (2005) proposed that the reasons for the reduction in performance and capacity in a hot environment may be attributed to mechanisms within the central nervous system and possibly due to alterations in the direct transfer of molecules between the brain and the peripheral circulation.

Cerebral homeostasis is protected by the presence of the blood-brain barrier. The blood-brain barrier is a dynamic structure formed from microvascular cells with tight junctions reinforced by the foot processes of surrounding astrocytes and the presence of pericytes. As a result of the tight junctions, the diffusion of molecules across the blood-brain barrier is tightly regulated when the barrier is intact. The integrity of the blood-brain barrier is maintained in the majority of circumstances; however, there are situations when the permeability of the blood-brain barrier may be increased. These situations include ischemic stroke, bacterial and viral infection, brain trauma (Marchi *et al.*, 2003) and exercise performed in a hot environment (Watson *et al.*, 2005a; Watson *et al.*, 2005b). An increase in the permeability of the blood-brain barrier leads to the transfer of molecules across the membrane and the leakage of detectable substances from the central nervous system into the circulation. One such substance in particular which has been proposed as a marker of blood-brain barrier integrity is S100β (Kapural *et al.*, 2002).

S100β is a cytosolic calcium-binding protein primarily found in astrocytes and Schwann cells (Zimmer & Landar, 1995) and accounts for the majority of the serum S100 protein found in the human brain (Isobe *et al.*, 1983; Jensen *et al.*, 1985). Although Kapural *et al.* (2002) showed, via the infusion of mannitol, that S100β concentrations increase as a result of an increase in blood-brain barrier permeability the reasons for the increase in permeability are not fully understood. Recently it has been shown that S100β concentrations are elevated after 1hr submaximal cycle ergometer exercise in warm (Watson *et al.*, 2005a; Watson *et al.*, 2006), but not moderate (Watson *et al.*, 2005a),

conditions although consuming enough water to maintain euhydration can attenuate the increases observed (Watson *et al.*, 2006). The reasons for the increased concentrations of S100β in hot, but not moderate, conditions remain unclear as the response is not related to increases in core temperature (Watson *et al.*, 2005a). Watson *et al.* (2006) proposed that the increase may be due to the hyperosmolarity caused by the loss of large volumes of hypertonic sweat during such exercise. Osmotic movement of fluid across the blood-brain barrier, coupled with the dehydration-induced shrinkage of the structural endothelial cells, may result in the opening of the usually tight junctions and the increased release of S100β into the peripheral circulation.

In addition to exercise in a hot environment (Watson *et al.*, 2005a; Watson *et al.*, 2005b), ischemic stroke (Abraha *et al.*, 1997; Butterworth *et al.*, 1998; Buttner *et al.*, 1997; Cunningham *et al.*, 1996) has been shown to impair the integrity of the blood-brain barrier and elevate peripheral serum S100β concentrations. Ischemia occurs during hyperthermic exercise and it as been shown that cerebral blood flow decreases by ~20% during exercise performed in a hot environment (Nybo *et al.*, 2002b). It has been proposed that the reduced exercise performance and capacity observed in a hot environment could be due to a reduction in the amount of oxygen delivered to the cerebral tissue as a result of the reduced blood flow (Nybo & Nielsen, 2001d; Nybo *et al.*, 2002b) and/or the movement of substances implicated in fatigue across the damaged blood-brain barrier (Sharma *et al.*, 1991). If this suggestion is correct, an intervention which could increase cerebral blood flow and help maintain blood-brain barrier integrity would be advantageous to exercise performance and capacity in hot environments.

In stroke patients hypothermia is used as a treatment to maintain and/or increase the blood supply to ischemic regions (Adams *et al.*, 2005). Whole-body hypothermia is often used but it has been proposed that local cooling of the area superior to the carotid arteries via a neck collar may be the optimal intervention due to the hypothermia-induced vasodilation of the carotid artery (Mustafa & Thulesius, 2002). It seems prudent to suggest that the application of a cooling collar may increase cerebral blood flow which may help maintain blood-brain barrier integrity during exercise performed in a hot environment. If so, cooling the neck region would be expected to attenuate the serum S100β response observed and improve exercise performance.

Exercise performance was enhanced with the application of a cooling collar in Chapter 5 without alterations in the physiological or peripheral neuroendocrinological variables

measured. The aim of the current study is to investigate whether the application of a cooling collar improves exercise performance by offering protection to the blood-brain barrier, as assessed by concentrations of serum S100β, during prolonged exercise performed in a hot environment. It was hypothesised that if the performance enhancement was due to a cooling-induced maintenance of the blood-brain barrier then concentrations of S100β would be attenuated with the application of the collar.

6.2. Methods

6.2.1. Participants

The data presented in this experimental chapter were collected during the data collection of the previous experimental chapter. Serum samples were collected from eight of the nine participants during two of the experimental trials. The mean (± 1 SD) age, body mass, height and relative maximal oxygen uptake ($\dot{V}O_{2max}$) of the participants was 25 ± 5 y, 77.4 ± 5.6 kg, 1.81 ± 0.08 m and 53.7 ± 4.7 $mL \cdot kg^{-1} \cdot min^{-1}$. All of the participants were fully informed of any risks and discomforts associated with the study before giving their oral and written informed consent to participate and completing a health screen. The health screening was repeated prior to each laboratory visit. The study was approved by the institution's Ethical Advisory Committee.

6.2.2. Experimental procedures

Blood samples were collected from participants who completed the 90 min TT_{pre} cold collar (CC) and non-collar (NC) trials from the second experimental study (reported in Chapter 5). The trials were conducted in a randomised and counter-balanced order.

6.2.3. Collection and analysis of blood samples

Blood samples were drawn at 0, 70 and 90 min during both trials. The 70 min sample represented the pre-time-trial sample and was drawn at 70 rather than 75 min to prevent undue disruption to the participant at the commencement of the performance section of the protocol. 5 ml aliquots were dispensed into plain tubes (Sarstedt Ltd, Leicester, U.K.) and were allowed to clot for ~60 min at room temperature. Samples were centrifuged at 4000g for 10 min at 4°C. After centrifuging the supernatant was removed and then frozen at -80°C until the analyses were performed. Concentrations of S100β were measured using a commercially available enzyme-linked immunosorbant assay (Fujirebo Diagnostics AB,

Goteborg, Sweden). The intra-assay coefficient of variation established by in-house laboratory tests was 8.0%.

6.2.4. Statistical analysis

Descriptive data are reported as mean ± standard deviation (1 SD). Blood samples were analysed using paired students t-tests and two-way ANOVA for repeated measurements. Tukey's HSD *post hoc* tests were conducted to identify pair-wise differences when appropriate. Pearson correlation analysis was performed to investigate associations between serum S100β concentrations and other variables of interest. It was calculated that a sample size greater than N = 7 would provide sufficient statistical power (β = 0.2) to detect a difference larger than the typical S100β standard deviation (0.04 $\mu g \cdot L^{-1}$) for a normal population with a meaningful change for practical significance estimated as 0.12 $\mu g \cdot L^{-1}$ (Anderson *et al.*, 2001). Significance was accepted at the $P < 0.05$ level.

6.3. Results

6.3.1. Physiological and time-trial performance data

The physiological and time-trial performance data from this study is presented in the previous Chapter. To summarise, participants ran significantly further in CC (3,030 ± 485m) than NC trials (2,884 ± 571; P = 0.047). Mean neck temperature was significantly lower when wearing the cold collar ($P < 0.001$). There were no differences between trials for T_{rectal}, heart rate, sweat loss or voluntary fluid consumption ($P > 0.05$).

6.3.2. Serum S100β data

Concentrations significantly increased over time ($P < 0.001$) but there was no significant main trial (P = 0.700) or interaction effect (P = 0.284) for serum S100β concentrations (Figure 6.1). There was no difference in the post-time-trial (90 min) serum S100β concentrations in the cold collar trial compared to the no collar trial (0.089 ± 0.017 v 0.079 ± 0.027 $\mu g \cdot L^{-1}$; P = 0.056). All values were within normal range (0.035-0.124 $\mu g \cdot L^{-1}$ (Anderson *et al.*, 2001; Cheuvront *et al.*, 2008)). The mean pre-post trial difference (0.032 $\mu g \cdot L^{-1}$) was below the 0.08 $\mu g \cdot L^{-1}$ estimated for practical significance (Anderson *et al.*, 2001). Post-trial serum S100β concentrations were significantly correlated with baseline S100β concentrations, mean rectal temperature and the total distance covered but they were not correlated with post-trial (*i.e.* peak) rectal temperature, the change in rectal

temperature observed over the course of the trial or % dehydration (Table 6.1). The change in serum S100β observed during the time-trial phase was not correlated with either the time-trial distance ran or the change in core temperature over the same time-period (Table 6.1).

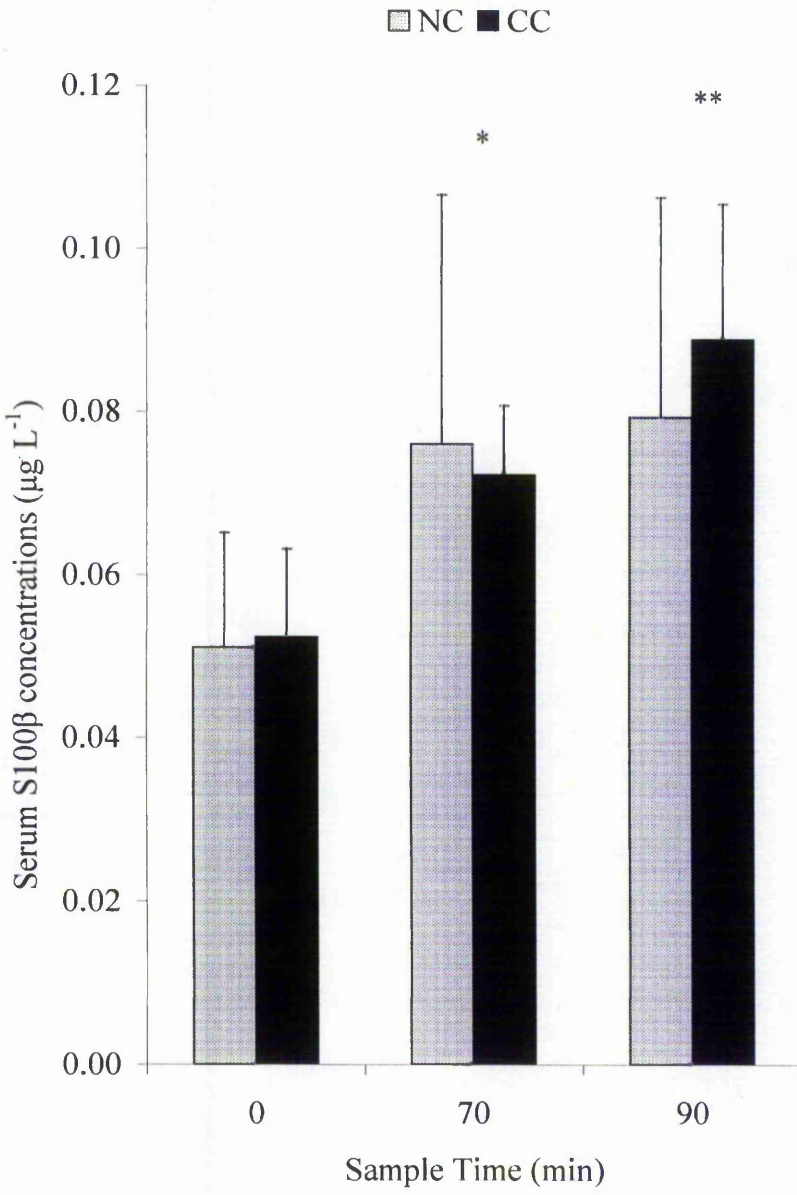

Figure 6.1. Serum S100β concentrations at 0, 70 and 90 min. * = significant difference from baseline ($P < 0.05$); ** significant difference from baseline ($P < 0.01$). Main effect time ($P < 0.001$).

Table 6.1. Relationship between serum S-100β and potential variables of influence

	r	P
Baseline S100β concentration ($\mu g \cdot L^{-1}$)[a]	0.748	0.001**
Total distance (m)[a]	0.741	0.001**
Time-trial distance (m)[b]	0.453	0.078
Peak T_{rectal} (°C)[a]	-0.463	0.071
0-90min ΔT_{rectal} (°C)[a]	-0.222	0.408
70-90min ΔT_{rectal} (°C)[b]	0.142	0.507
Dehydration (%)[a]	0.086	0.751

[a] Pearson correlation with final S100β ($\mu g \cdot L^{-1}$)
[b] Pearson correlation with 70-90min ΔS100β ($\mu g \cdot L^{-1}$)

6.4. Discussion

The present study investigated the effects of cooling the neck via a practical neck cooling collar on the serum S100β response to exercise in a hot environment. Time-trial performance was significantly improved with the application of a neck-cooling collar but the collar had no effect on the S100β response to the exercise bout.

Serum S100β concentrations are elevated following exercise in elevated ambient conditions and although much of the increase is attenuated via the maintenance of euhydration, increases are still observed in such conditions (Watson *et al.*, 2006). S100β concentrations increase as a result of blood-brain barrier disruption which may be caused by hyperosmolality (Watson *et al.*, 2006) and/or cerebral ischemia (Reynolds *et al.*, 2003). It has been proposed that the impaired performance observed in hot conditions may be due to the reduced cerebral blood flow which is observed during exercise in the heat (Nybo *et al.*, 2002b) and/or the leakage of substances across the damaged blood-brain barrier (Sharma *et al.*, 1991). During ischemic stroke the blood-brain barrier integrity is compromised by endothelial cell death and there is an increased potential for cytosolic contents released by the damaged brain to cross this barrier (Reynolds *et al.*, 2003). Cooling the neck is an effective way to increase cerebral blood flow, and potentially maintain blood-brain barrier integrity, (Mustafa & Thulesius, 2002) but there was no difference in the S100β response to the exercise bout with the collar in the current study. This suggests that cooling the neck has no effect on blood-brain barrier integrity during prolonged exercise in a hot environment.

Watson *et al.* (2006) investigated the effect of fluid ingestion on blood-brain barrier permeability during prolonged intermittent exercise in the heat and showed that dehydration (~2.8% body mass loss) significantly increased serum S100β concentrations (~10 $\mu g \cdot L^{-1}$) and that the changes in S100β concentrations were significantly correlated

with the change in serum osmolality ($r = 0.662$; $P = 0.005$). In the present study there was no significant difference in the amount of water consumed or sweat lost between trials and although Osmolality was not measured there was no correlation between the percentage dehydration and S100β concentrations ($r = 0.086$; $P = 0.751$). It is possible that the S100β elevations may not reflect changes in blood-brain barrier integrity during exercise-heat stress but instead simply reflect the stress of the exercise (Cheuvront *et al.*, 2008; Hasselblatt *et al.*, 2004).

The β subunit of the S100β protein is highly specific to the central nervous system (Ali *et al.*, 2000); however, it is expressed in small quantities in adipose and skeletal muscle tissues (Zimmer *et al.*, 1995) . As a result, elevations may occur following exercise in the absence of blood-brain barrier disruption. Elevations in serum S100β levels have been reported following bouts of boxing, soccer, ice-hockey, jogging, running and cycling in most (Dietrich *et al.*, 2003; Otto *et al.*, 2000; Stalnacke *et al.*, 2003; Stalnacke *et al.*, 2006; Watson *et al.*, 2005a; Zetterberg *et al.*, 2007) but not all (Cheuvront *et al.*, 2008; Watson *et al.*, 2005a) studies. In the present study, S100β concentrations were significantly elevated by the exercise bout but unaffected by the cooling collar. Significantly more distance was covered in the CC trial and there was a positive correlation between S100β concentrations and the total distance covered (Table 6.1). In contrast, Cheuvront *et al.* (2008) reported that ~100 min of treadmill walking ($1.56 m \cdot s^{-1}$; 4% grade) in very hot conditions (45°C; 20% rh) did not significantly elevate concentrations of S100β and saw no relationship between S100β concentrations and the amount of work done (exercise duration) ($r = 0.40$; $P = 0.09$). Despite the suggestion that S100β levels are predominantly increased due to blood-brain barrier disruption (Kapural *et al.*, 2002) it has been proposed that the elevated concentrations of S100β observed following exercise may have a peripheral, rather than central, source and be elevated as a result of muscle damage (Hasselblatt *et al.*, 2004; Schulpis *et al.*, 2007) and/or axial vibration of the brain (Otto *et al.*, 2000). Sports with less acceleration/deceleration events have lower serum S100β responses than those with a higher incidence of such events (Stalnacke *et al.*, 2003; Stalnacke *et al.*, 2006) and so the differing responses between the data from the current study and that of Cheuvront *et al.* (2008) may have been due to the lower ground forces and/or acceleration and deceleration forces on the brain or muscles experienced during walking than running. In the current study serum S100β concentrations were elevated after a sustained bout of moderate intensity running suggesting that running provides a sufficient stimulus for the release of serum S100β. It remains unclear whether the increase observed following running is due to

higher levels of brain trauma occurring due to axial vibration of the brain or increased muscle damage as a result of the exercise itself.

Hyperthermia has also previously been proposed as a stimulus for the release of S100β (Watson *et al.*, 2005a; Watson *et al.*, 2006) although passive hyperthermia, via water-immersion, does not elevate serum S100β concentrations (Watson *et al.*, 2005b). This supports the idea that the increase is caused by the stress of the exercise rather than the hyperthermia. In the current study there was no relationship between serum S100β concentrations and the change in rectal temperature observed (Table 1) which supports other recent data (Cheuvront *et al.*, 2008). Cheuvront *et al.* (2008) investigated the serum S100β response to a 10-day acclimation protocol and found that despite acclimation-induced reductions in core-temperature there were no significant differences in serum S100β concentrations pre- to post-acclimation. These results were observed despite a higher hyperthermic strain on day 1. As with the data from the current study there was also no correlation between serum S100β concentrations and peak rectal temperature. In addition, there was also no relationship between the acclimation-induced reductions in core temperature and the S100β response ($r = 0.10$) (Cheuvront *et al.*, 2008). The findings of the current study support the recent literature that elevations in S100β are not body-temperature dependent.

6.5. Conclusion

It has been suggested that ischemia-induced impairments in blood-brain barrier integrity elevates serum S100β concentrations. Cooling the neck region has been proposed as a way to maintain the integrity of the blood-brain barrier which could have performance benefits. Data from the current study show that the performance benefits gained by cooling the neck during exercise in the heat can not be attributed to a cooling-induced attenuation of the S100β response as concentrations were unaffected by the collar.

Chapter 7: Short-duration neck cooling whilst running does not improve time-trial performance in the heat

7.1. Introduction

Data from the study presented in Chapter 4 and from other studies have consistently demonstrated that exercise performance is impaired in hot, compared to temperate, conditions (Tucker *et al.*, 2004; Tatterson *et al.*, 2000; Tucker *et al.*, 2004). Many studies have indicated that there is an internal temperature of ~ 40°C above which exercise is voluntarily terminated (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993); however, it is likely that it is the obtainment of a high hypothalamic, rather than body, temperature that ultimately limits exercise (Caputa *et al.*, 1986). The temperature of the brain is primarily dependent on the temperature of the carotid blood that supplies it (Baker, 1982). Many animal species are able to lower the temperature of the blood reaching the brain below that of their body via a mechanism called selective brain cooling (Baker, 1979) and most do so due to the presence of a carotid rete (a network of medium-sized arteries which lay in a lake of venous blood) in either the cavernous sinus or in the pterygoid plexus.

Anatomical reviews and direct cooling interventions in anesthetised humans suggest that humans do not have the ability to selectively brain cool (Brengelmann, 1993; Shiraki *et al.*, 1988) although mathematical modelling studies have suggested that peripheral brain temperature can be reduced by the application of a cooling device (Sukstanskii & Yablonskiy, 2004; Zhu, 2000). Nybo *et al.* (2002) reported that during 40 min of exercise in which participants cycled while wearing water-proof attire to produce a heat stress the temperature of the jugular venous blood was always greater than the temperatures measured at the oesophagus and aortic arch, demonstrating that the cerebral temperature exceeded that of the body's core throughout exercise. Although it seems unlikely that selective brain cooling occurs in humans (Brengelmann, 1993), it has been proposed that artificial cooling of the carotid blood, via external cooling devices, may be able to reduce the temperature of the carotid blood (Caputa, 2004; Gordon *et al.*, 1990; Zhu, 2000) and, in turn, possibly mimic the selective brain cooling mechanism.

Cooling the neck region via water-perfused garments has been shown to reduce cardiovascular and thermoregulatory strain during exercise in hot conditions in some (Palmer *et al.*, 2001; Shvartz, 1970; Shvartz, 1976; Watanuki, 1993) but not all (Nunneley *et al.*, 1971) studies; however, only one study has investigated the effect of cooling the neck region via a water-perfused garment on exercise performance in the heat. Palmer *et al.* (2001) reported that cooling the head via a water-perfused hood during 60 min of seated rest followed by 30 min of submaximal treadmill running (60% $\dot{V}O_{2max}$) improved

subsequent 15 min time-trial performance in a hot environment (32°C; 55% rh) by 2.3% compared to cooling at rest and by 3.3% compared to no-cooling. The head-cooling intervention reduced rectal temperature and improved thermal sensation but had no effect on heart rate or ratings of perceived exertion. Water-perfused garments can alter the physiological response to exercise and improve exercise performance in a hot environment; however, cooling mechanisms are impractical in a sporting setting and so more practical alternatives have been investigated.

In Chapter 5 the application of a practical neck-cooling device was shown to improve 15 min time-trial performance in the heat when applied during a 90 min preloaded time-trial by ~6%. It was demonstrated that the lowering of neck temperature had no effect on core temperature, heart rate, sweat loss or fluid consumption but did alter the rating of perceived exertion (RPE) (Borg, 1982) and perception of thermal comfort. Participants felt significantly cooler with the application of a cooling collar and were able to cover significantly more distance with the cold collar (3,030 ± 537m) compared to no collar (2,884 ± 571m; P = 0.041). It is worth noting that the collar significantly lowered mean neck temperature during the early stages of the 75 min preload phase, although there was no difference in the neck temperature at the commencement of, or during, the time-trial phase. In addition to the study conducted in Chapter 4, only three other studies have investigated the effect of cooling the neck via a practical neck-cooling device, although none of these studies investigated the effects of such a cooling intervention on exercise performance (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990; Hamada *et al.*, 2006).

Studies that have adopted constant-intensity exercise models have demonstrated that the attainment of a high core temperature can limit exercise in hot conditions (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993); however, when participants exercise in self-paced models the results differ (Tatterson *et al.*, 2000; Marino *et al.*, 2004). Tatterson *et al.* (2000) reported that 30 min time-trial performance was impaired in hot (32°C) compared to moderate (23°C) conditions, despite no difference in rectal temperatures, while down-regulation of self-selected pace has been shown to occur before the onset of high core temperatures (Marino *et al.*, 2004). Based upon these findings, it has been proposed that self-selected pace may be regulated based upon feedback relating to the rate at which core temperature is rising to prevent the onset of dangerously high temperatures (Marino *et al.*, 2004; Tatterson *et al.*, 2000). Providing a cooling stimulus has been demonstrated to alter self-selected pacing and it has been proposed that the pace may have been mediated by thermoregulatory cues (Arngrimsson *et al.*, 2004). On this basis it would seem prudent to

suggest that the enhanced performance reported in Chapter 4, that was observed despite the collar having no significant effect on the physiological response to the exercise, may have been due to the provision of false thermoregulatory feedback, as supported by the significant benefit of the collar on subjective feelings of thermal comfort.

In Chapter 5 the cooling of the neck region via a practical cooling collar was shown to improve exercise performance in the heat even when the mean neck temperature is no longer cooler at the onset of the performance phase of the trial. It would be sensible to suggest that if cooling the neck *per se* enhances exercise performance then maintaining a cool neck for the duration of a time-trial would be beneficial when running in the heat. The present study has been designed to investigate the effect of short-term cooling of the neck region on time-trial performance in hot conditions.

7.2. Methods

7.2.1. Participants

Ten healthy, trained males volunteered for the study. Due to injury two volunteers failed to complete all trials and their data has been omitted. The mean (± 1 SD) age, body mass, height and relative maximal oxygen uptake ($\dot{V}O_{2max}$) of the eight participants was 25 ± 3 y, 75.5 ± 7.0 kg, 1.80 ± 0.05 m and 54.9 ± 3.1 $ml \cdot kg^{-1} \cdot min^{-1}$. The runners were fully informed of any risks and discomforts associated with the study before giving their oral and written informed consent to participate and completing a health screen. The health screening was repeated prior to each laboratory visit to ensure that the health status of the participants was constant throughout the duration of the investigation. The study was approved by the institution's Ethical Advisory Committee.

7.2.2. Experimental procedures

Prior to the main trials participants completed an incremental motorised treadmill test to determine $\dot{V}O_{2max}$ as per Jones and Doust (1996). Following a full habituation trial, participants visited the laboratory on two occasions for the main experimental trials. The experimental trials were conducted at the same time of day on each occasion ± 10 min to control for circadian rhythm fluctuations. Seven days has been shown to be a sufficient duration between trials in hot conditions to prevent acclimation occurring (Chapter 4) and

therefore all trials were separated by 7-days. Participants were not acclimatised and the outdoor temperature on the days of the trials was 5.6 ± 1.5°C.

Participants completed two experimental trials; one trial whilst wearing a cold collar (CC) (see section 3.5.2.6) and one without wearing the collar (NC). The trials were conducted in a randomised and counter-balanced order. It was not possible to blind the trials and so in an attempt to prevent any related problems participants were given information regarding why the collar could improve or impair performance and the lead investigator was careful to not give any information regarding his opinion of the effectiveness of such an intervention.

During the habituation and experimental trials participants completed a 5 min warm-up followed by a 15 min time-trial in hot conditions (30.4 ± 0.1°C; 53 ± 2% rh). The standardised warm-up consisted of 5 min of treadmill running at 9.0km·h^{-1} (~60% $\dot{V}O_{2max}$). Participants were instructed to cover as much distance as they could during the time-trial and were able to increase and decrease the treadmill speed as desired. The duration remaining was displayed via a countdown timer. The distances covered were not revealed until the completion of all trials.

Participants abstained from alcohol and caffeine and completed a food record for the 24h prior to the initial experimental trial. They adopted the same diet and abstained from strenuous exercise for 24h, prior to each main trial. Participants arrived at the laboratory ~30-mins before the commencement of the trial, >4h postprandial, having ingested 500 ml of water ~1.5h previously. On arrival, nude body mass was recorded. A rectal probe (Grant Instruments (Cambridge) Ltd, England, U.K.) was self-inserted ~10cm past the anal sphincter and a heart rate (HR) monitor (Polar Electro Oy, Kempele, Finland) was attached prior to the participant entering the environmental chamber (Design Environmental WIR52-20HS, Design Environmental Ltd., Gwent, Wales, U.K.). Participants rested in the environmental chamber in an upright position for 10 min, wearing the collar when appropriate (cold collar trial), prior to the commencement of the standardised warm-up.

During the time-trial, HR, rectal temperature (T_{rectal}), mean neck temperature (T_{neck}) were recorded at 1 min intervals; rating of perceived exertion (RPE) (Borg, 1982) and thermal sensation (TS) were recorded at 3 min intervals and the distance covered and the self-selected speed were recorded at 30s intervals. Heart rate, distance covered and the self-selected speed were recorded by the HP-Cosmos Para Graphics software package (HP-

Cosmos Sports and Medical, Nussdorf-Traunstein, Germany). Mean neck temperature was calculated as the mean temperature of four skin thermistors (Grant Instruments (Cambridge) Ltd, England, U.K.) spaced equally across the posterior aspect of the neck. All thermistors were attached via a transparent dressing (Tagaderm, 3M Health Care, USA) and water-proof tape (Transpore, 3M Health Care, USA). TS was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987).

Following the completion of each trial, participants towel-dried and recorded a dry post-exercise body mass from which sweat loss and the percentage change in body mass was calculated taking into account *ad libitum* fluid consumption.

7.2.3. Statistical analysis

Descriptive data is reported as mean ± 1 standard deviation (1 SD). Paired t-tests were used to examine differences between trials for the distances ran, sweat lost and fluid consumed, while two-way repeated-measures ANOVA were performed to evaluate differences in the physiological and perceptual responses between trials. Following a significant F value Tukey's HSD *post hoc* tests were conducted to identify pair-wise differences. Significance was set at the $P < 0.05$ level.

7.3. Results

7.3.1. Time-trial performance

During the time-trial, participants covered 3,180 ± 271m and 3,239 ± 267m in the no collar and cold collar trials respectively. There was no significant difference in the distance covered between trials ($P = 0.351$). The performance of five of the eight volunteers was improved with the application of cold collar while the performance of the other three was impaired. There was no significant difference between the self-selected time-trial speeds selected between trials ($P = 0.298$).

7.3.2. Neck temperature, rectal temperature and heart rate

Mean neck temperature (T_{neck}) is shown in Figure 7.1. There was a significant main effect for trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P < 0.001$). T_{neck} was significantly

colder throughout the 15 min time-trial in CC compared to NC trials ($P < 0.001$). There were no significant differences between trials for heart rate ($P = 0.780$; Figure 7.2) or rectal temperature ($P = 0.665$; Figure 7.3).

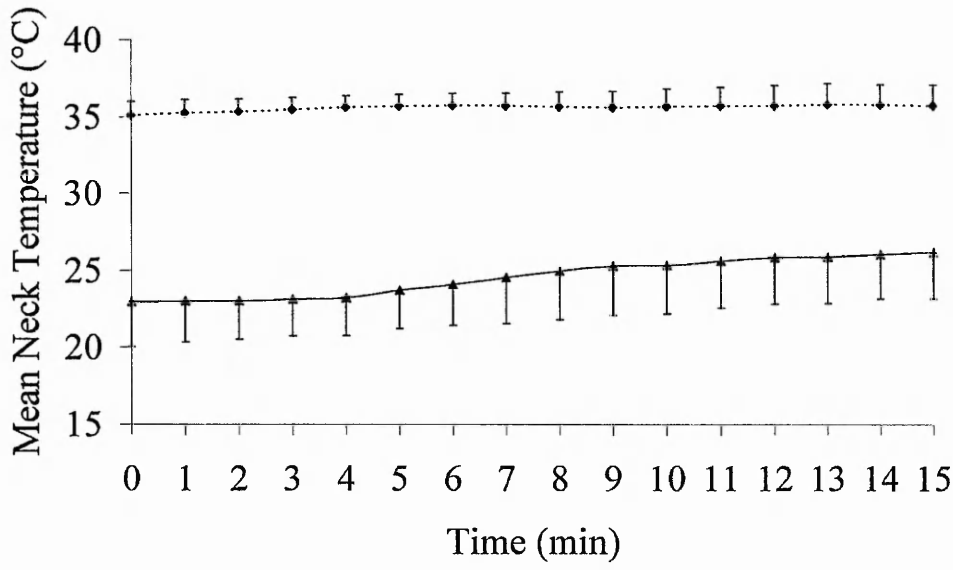

Figure 7.1. Mean neck temperature during the time-trial. ♦ = no collar (NC); ▲ = cold collar (CC). Main effect trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P < 0.001$).

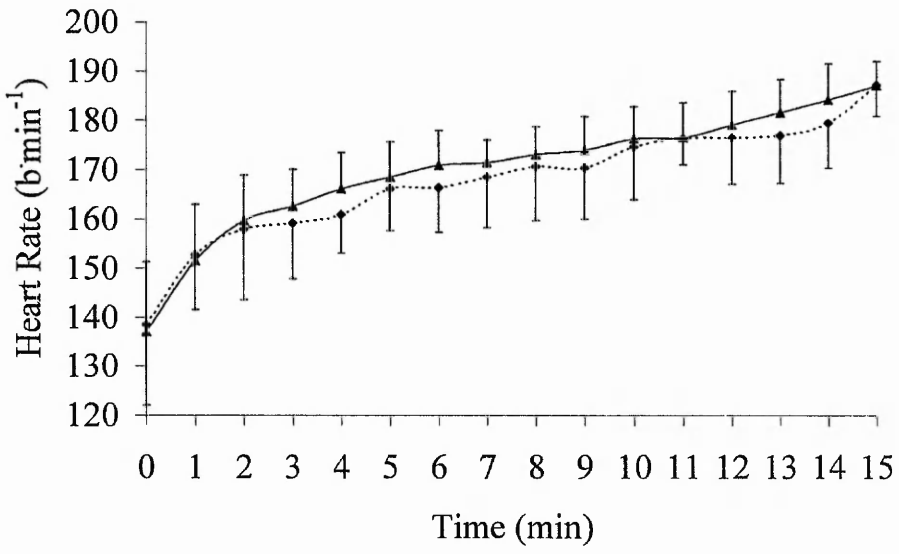

Figure 7.2. Mean heart rate during the time-trial. ♦ = no Collar (NC); ▲ = cold collar (CC).

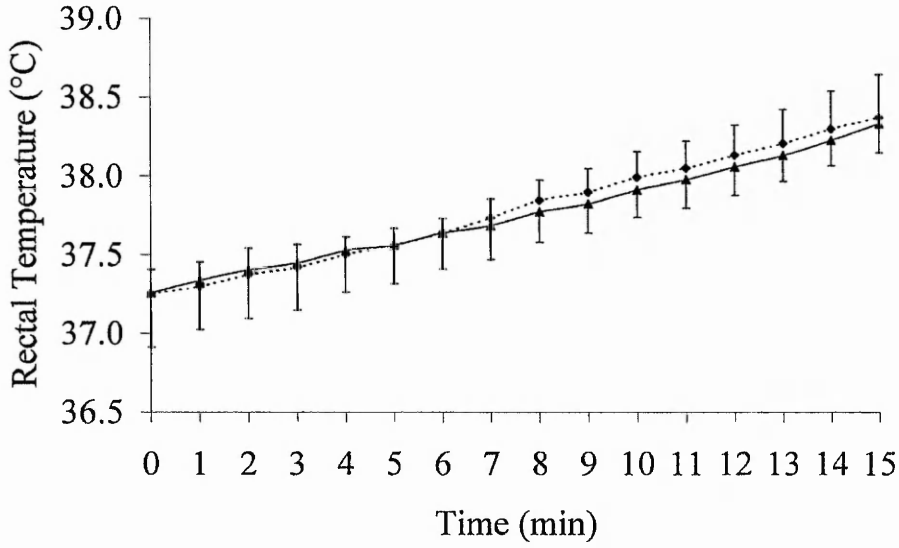

Figure 7.3. Mean rectal temperature during time-trial. ♦ = no collar (NC); ▲ = cold collar (CC).

7.3.3. Perceptual measurements

Rating of perceived exertion was unaltered by the application of the cooling collar (P = 0.925). There was a significant main effect for trial (P = 0.009) and time for thermal sensation (P < 0.001) (Figure 7.4). Participants felt cooler in the cold collar trials (P = 0.009).

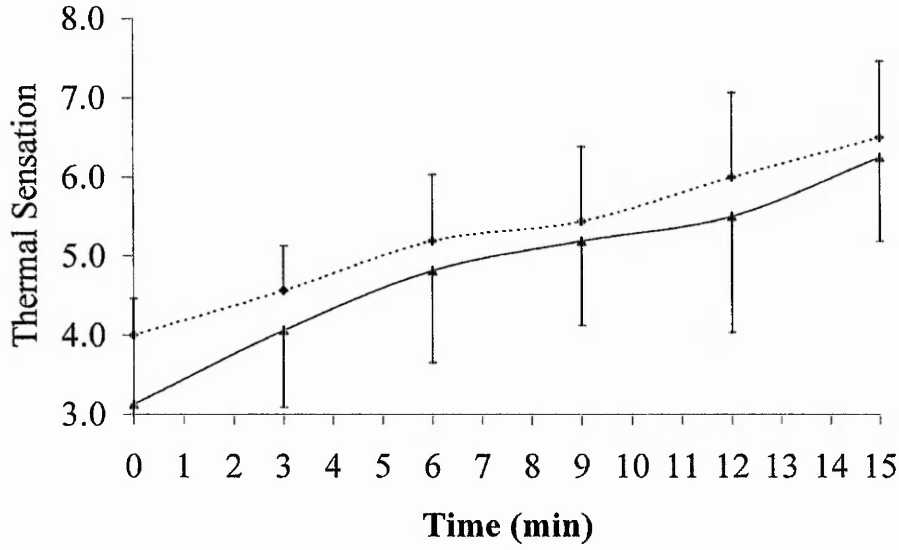

Figure 7.4. Thermal sensation during the time-trial. ♦ = no collar (NC); ▲ = cold collar (CC). Main effect trial (P = 0.009) and time (P < 0.001).

7.3.4. Body fluid regulation

Sweat loss was not significantly different between trials (no collar: 0.638 ± 0.183 L; cold collar: 0.602 ± 0.245 L; P = 0.756) however voluntary water consumption was significantly reduced by wearing the cold collar (0.083 ± 0.111 v 0.113 ± 0.100 L; P = 0.033).

7.4. Discussion

Data from Chapter 5 shows that the lowering of T_{neck} elicits beneficial changes during prolonged exercise; however, the main finding of the current study is that reducing T_{neck} does not improve short-duration time-trial performance in the heat.

In accordance with other cooling-collar studies (Bulbulian *et al.*, 1999; Hamada *et al.*, 2006), T_{rectal}, HR, RPE and sweat rate were unaffected by the application of a cooling collar. Unlike the study in Chapter 5, time-trial performance was also unaffected. In Chapter 5, the cooling device was worn during exercise for a total of 90 min (a 75 min preload (60% $\dot{V}O_{2max}$) immediately followed by a 15 min time-trial) during which time the collar had lost its cooling effect by the commencement of the performance test. Despite the loss of cooling, time-trial performance was improved. This is in contrast to Palmer *et al.* (2001) who reported that cooling during a period of rest and exercise improved time-trial performance compared to cooling exclusively at rest, although this may have been due to body temperature reductions. In the current study the collar was only worn for 20 min of exercise and therefore the neck remained significantly cooler throughout. The results show that the application of a cold collar *per se* does not improve exercise performance in the heat.

Heat tolerance can be enhanced by the maintenance of a cooler brain, which has led to the proposal that brain temperature may be more important than core temperature in the regulation of thermal tolerance and possibly exercise adherence in hot conditions (Caputa *et al.*, 1986). Brain temperature is primarily dependent on the temperature of the blood within the carotid arteries (Baker, 1982) and it has been suggested that the application of a cooling collar may lower carotid blood temperature and therefore reduce the temperature of the brain (Caputa, 2004; Gordon *et al.*, 1990; Zhu, 2000). Human brain temperature is practically homogenous and alterations can only occur at the surface to a depth of ~3 - 4mm (Sukstanskii & Yablonskiy, 2007a). Although the collar is highly unlikely to reduce deep brain temperature it has been proposed that the temperature of the blood reaching the thermoregulatory centre could be reduced by the external cooling which may result in the masking of signals regarding the thermal state of the body (Brisson *et al.*, 1987).

It has been suggested that during exercise, pacing strategies are adopted to allow for the completion of the task within homeostatic limits (Tucker *et al.*, 2004). Tucker *et al.* (2004) investigated the pacing strategy adopted during a 20km time-trial in hot (35°C) compared to moderate (15°C) conditions and showed that a reduction in pace occurred in the hot trials well in advance of the obtainment of a high core temperature. They proposed that this reduced pace was selected as part of an anticipatory mechanism down-regulating muscle recruitment to prevent the development of excessive thermal strain (Tucker *et al.*, 2004). If the pacing mechanism is disrupted by the provision of a masked signal giving a false

representation of the body's thermal status it could result in the selection of a "false" or "un-natural" pace. Pre-cooling the torso (Arngrimsson *et al.*, 2004) and neck cooling during exercise (Chapter 5) have both been shown to allow participants to adopt a faster self-selected pace and so it appears that the provision of a cooling stimulus may allow for the selection of a pace faster than that which would be naturally selected. In the study in Chapter 5 the rectal temperature was higher in the cold collar trial compared to that observed in the present study at the beginning (38.64 ± 0.29°C v 37.25 ± 0.15) and end (39.14 ± 0.27°C v 38.37 ± 0.27°C) of the time-trial. The lower core temperatures observed in the current study are due to the shorter duration spent exercising in the high ambient temperatures and it possible that there was no effect upon self-selected pace, and therefore performance, with the application of the collar because the runners were under insufficient thermal stress to necessitate the masking of signals relating to the thermal state of the body.

7.5. Conclusion

Short-duration neck cooling does not alter 15 min time-trial performance in the heat. It seems prudent to suggest that neck-cooling may only be beneficial when the body is under a greater level of thermal stress than that observed in the current study.

Chapter 8: Sustained cooling of the neck offers no cumulative benefit to running performance in a hot environment compared to acute cooling

8.1. Introduction

It is well documented that high ambient temperatures result in an impaired exercise performance (Tatterson *et al.*, 2000; Tucker *et al.*, 2004). The data from the first experimental study within the thesis (presented in Chapter 4) demonstrated that 15 min time-trial performance is impaired in hot (30°C) compared to moderate (14°C) conditions by ~10% when undertaken following a 75 min submaximal preload bout of exercise. Many top class sporting events take place in elevated ambient temperatures (*e.g.* the Athens and Beijing Olympic Games) and as a result any intervention which can attenuate the reduction in exercise performance observed, therefore giving the athlete an advantage, would be of great interest to any individual performing in hot conditions. Hyperthermia has been implicated in the reduced ability to perform and therefore a number of different cooling strategies have been investigated and adopted (Marino, 2002; Quod *et al.*, 2006).

Pre-cooling has consistently been shown to enhance subsequent prolonged exercise performance in the heat; however, it has limited practical application due to the associated disruption and time required to implement. Due to this, more practical alternatives have been investigated, such as the wearing of a cooling jacket or vest (Arngrimsson *et al.*, 2004; Duffield *et al.*, 2003; Duffield & Marino, 2007; Webster *et al.*, 2005) or neck cooling collar (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). The majority of these studies have reported that the practical devices assessed provided an insufficiently sustained cooling effect and this is particularly the case with the neck cooling devices investigated (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990). Cooling the neck region has many theoretical benefits compared to cooling elsewhere due to its close proximity to the thermoregulatory centre, located at the base of the brain, and cooling this region via water perfused garments has been shown to be more effective at alleviating heat strain than cooling a section of the chest of the same surface area (Shvartz, 1976). Cooling the head via a water-perfused garment improves time-trial performance in a hot environment (Palmer *et al.*, 2001); however, to the author's knowledge, the studies reported in Chapters 5 and 7 are the first to look at the effects of using a practical neck-cooling device on exercise performance in the heat. The neck-cooling device significantly improved 15 min time-trial performance in the heat in Chapter 5 when the collar was worn during a 90 min preloaded time-trial but had no effect on running performance when worn for the shorter duration of the study in Chapter 7. In both studies, cooling the neck via the practical cooling collar had no effect on any of the physiological or neuroendocrinological variables measured, as was reported in the other two studies (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990).

Palmer *et al.* (2001) reported that sustained cooling of the head during a bout of rest, followed by exercise, improved time-trial performance in the heat compared to when the cooling stimulus was removed prior to exercise. No data was provided regarding the effect of cooling the neck exclusively at rest compared to no cooling; however, it appears that sustained cooling might offer a cumulative benefit. The effectiveness of any cooling intervention appears to be dependent on the difference between the magnitude of the cooling and the thermal strain experienced and the greatest benefits are often reported when the thermal strain is at its most severe (Nunneley *et al.*, 1971). Based upon this, it was hypothesised that the differences in the performance effect observed was due to differing levels of thermal strain experienced in the two trials. There was a difference of approximately 1.44°C in the rectal temperature observed at the beginning of the 15 min performance test in Chapters 5 and 7 (38.64 ± 0.29°C v 37.25 ± 0.15) and therefore it seems that, as has been observed for physiological responses to head cooling (Nunneley *et al.*, 1971), there is a threshold of thermal strain that needs to be breached before a neck-cooling intervention has a positive effect on time-trial performance.

Time-trial performance was improved by cooling the neck region in the study in Chapter 5 despite the fact that the cooling effect was no longer in existence at the onset of the performance test period. If the benefit of a cooling intervention is largely dependent on the difference between the level of cooling provided and the level of thermal strain experienced (Nunneley *et al.*, 1971), it would seem prudent to suggest that providing a sustained bout of cooling during the 90 min preloaded time-trial would have a cumulatively beneficial effect on time-trial performance. It was hypothesised that the additional performance improvement would occur as a result of a greater improvement in the perceived level of strain experienced and not due to the additional cooling affecting the physiological or hormonal response to the exercise. The aim of the current study was to test this hypothesis and to investigate the effect of maintaining the neck at a reduced temperature, via the replacement of the neck cooling collar, throughout the 90 min protocol.

8.2. Methods

8.2.1. Participants

Seven healthy, trained males volunteered for the study. The mean (± 1 SD) age, body mass, height and relative maximal oxygen uptake ($\dot{V}O_{2max}$) of the participants was 25 ± 2 y, 75.3 ± 8.4 kg, 1.79 ± 0.05 m and 55.3 ± 3.6 $ml \cdot kg^{-1} \cdot min^{-1}$. The participants were fully

informed of any risks and discomforts associated with the study before giving their oral and written informed consent to participate and completing a health screen. The health screening procedure was repeated prior to each laboratory visit to ensure the health status of the individual. The study was approved by the institution's Ethical Advisory Committee.

8.2.2. Experimental procedures

Prior to the main trials, participants completed an incremental motorised treadmill test to determine $\dot{V}O_{2max}$ (Jones & Doust, 1996). Following a full habituation trial participants visited the laboratory on three occasions for the main experimental trials. The trials were conducted in a randomised and counter-balanced order. The experimental trials were conducted at the same time of the day on each occasion ± 30 min and were separated by 7 days. Participants were not naturally acclimatised to a hot environment and the mean outdoor temperature on the mornings of the main trials was 8.1 ± 3.6°C.

During the habituation and experimental trials participants completed a 90 min preloaded time-trial (TT_{pre}) in hot conditions (30.4 ± 0.1°C; 53 ± 2% rh). The TT_{pre} consisted of 75 min of treadmill running at ~60% $\dot{V}O_{2max}$ (9.0 ± 1.0km·h^{-1}) followed by a self-paced 15 min time-trial during which participants were manually able to increase and decrease their speed and were instructed to cover as much distance as they could during the time, displayed via a countdown timer. The distances covered were not revealed until the completion of all four trials (one habituation and three experimental).

During the three experimental trials participants completed the TT_{pre} whilst wearing either a cold collar for the duration of the trial (CC), a cold collar worn from the start and replaced twice during the TT_{pre} at 30 and 60 min (CC_{rep}), or no collar (NC). The collar was comprised of two parts and was modified from a commercially available neck cooling device (Black Ice LLC, Lakeland, USA) (see section 3.5.2.6).

Participants abstained from alcohol and caffeine and completed a food record for the day prior to the initial experimental trial. They adopted the same diet and abstained from strenuous exercise for 24h, prior to each main trial. Participants arrived at the laboratory ~30 min before the commencement of the trial in a fasted (≥ 10h postprandial) state and having ingested 500 ml of water ~1.5h previously. On arrival, nude body mass was recorded. A rectal probe (Grant Instruments (Cambridge) Ltd, England, U.K.) was self-inserted ~10cm past the anal sphincter; a heart rate (HR) monitor (Polar Electro Oy, Kempele, Finland) was attached and an indwelling cannula (Venflon, Becton Dickinson

U.K. Ltd, Oxford, U.K.) was inserted into a vein of the antecubital fossa prior to the participant entering the environmental chamber (Design Environmental WIR52-20HS, Design Environmental Ltd., Gwent, Wales, U.K.). The indwelling cannula was kept patent by an injection of saline (~5 ml) after each sample. Participants rested in the environmental chamber in an upright position for 10 min, after which resting values for HR, rectal temperature (T_{rectal}), mean neck temperature (T_{neck}), whole-body thermal sensation (TS) and thermal sensation of the neck (TS_{neck}) were obtained. The collar was then placed around the neck in the CC and CC_{rep} trials. Mean neck temperature was calculated as the mean temperature of four skin thermistors (Grant Instruments (Cambridge) Ltd, England, U.K.) spaced equally across the posterior aspect of the neck. All thermistors were attached via a transparent dressing (Tagaderm, 3M Health Care, USA) and water-proof tape (Transpore, 3M Health Care, USA). TS and TS_{neck} was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987). Chilled water was allowed *ad libitum* during all of the trials.

During the 90 min TT_{pre}, HR, T_{rectal}, T_{neck}, rating of perceived exertion (RPE) (Borg, 1982), TS and TS_{neck} were recorded at 5 min intervals. The distances ran during the 75 min preload phase and the subsequent 15 min time-trial were noted. The self-selected pace was recorded every min during the time-trial. Following the completion of each trial participants towel-dried and recorded a dry post-exercise body mass from which sweat loss and the percentage change in body mass was calculated taking into account voluntary fluid consumption during the protocol.

8.2.3. Collection and analysis of blood samples

Blood samples were taken at 0, 10, 40, 70 and 90 min at which times participants were stationary. All blood samples were taken within 2 min therefore participants were stationary for < 6 min during the 90 min trial. Whole blood was initially analysed for lactate and glucose (Yellow Springs Instrument 2300 STAT plus, Yellow Springs Instruments Inc., Ohio, USA) and then aliquots were dispensed into K_3-EDTA tubes (Sarstedt Ltd, Leicester, U.K.). Samples were centrifuged at 4000g for 10 min at 4°C. After centrifuging the supernatant was removed and then frozen at -80°C until the analyses were performed.

Changes in blood, plasma and red cell volume (BV, PV and CV respectively) were calculated from the mean haemoglobin (Hb) concentration (B-hemoglobin photometer,

Hemocue AB, Angelholm, Sweden, measured in triplicate) and the mean haematocrit (Hct) (Micocentrifugation, Hawksley, Sussex, U.K., in triplicate), measured using the methods of Dill and Costill (1974).

Plasma concentrations of cortisol, dopamine and serotonin were determined via enzyme-linked immunosorbent assays; cortisol (DRG Instruments GmbH, Marburg, Germany); dopamine and serotonin (IBL Hamburg, Hamburg, Germany). The intra-assay variation (CV) for the cortisol, dopamine and serotonin assays were 4.3%, 8.1%, and 5.7% respectively.

8.2.4. Statistical analysis

Descriptive data is reported as mean ± 1 standard deviation (1 SD). One-way repeated-measures analysis of variance (ANOVA) tests were conducted to evaluate differences between the distances ran, sweat-loss and fluid consumption while two-way (trial x time) tests were performed to evaluate differences between trials for thermoregulatory, cardiovascular, neuroendocrinological and perceptual variables. Following a significant F value Tukey's HSD *post hoc* tests were conducted to identify pair-wise differences. Significance was set at the $P < 0.05$ level.

8.3. Results

8.3.1. Time-trial performance

During the time-trial phase participants covered 2,597 ± 291m; 2,779 ± 299m and 2,776 ± 331m in the NC, CC and CC_{rep} trials (Figure 8.1). There was a significant main effect for trial ($P = 0.003$). Significantly more distance was covered in CC compared to NC ($P = 0.007$) and in CC_{rep} compared to NC ($P = 0.008$). There was no significant difference in the distance covered in CC compared to CC_{rep} ($P = 0.998$). There was no significant difference between trials for the pacing strategy selected ($P = 0.283$) (Figure 8.2). Individual percentage time-trial performance differences compared to NC trials are presented in Figure 8.3. The mean percentage improvement in time-trial performance compared to NC was 7.3% in CC and 6.9% in CC_{rep}. No trial-order effect was observed for the distances ran ($P = 0.926$).

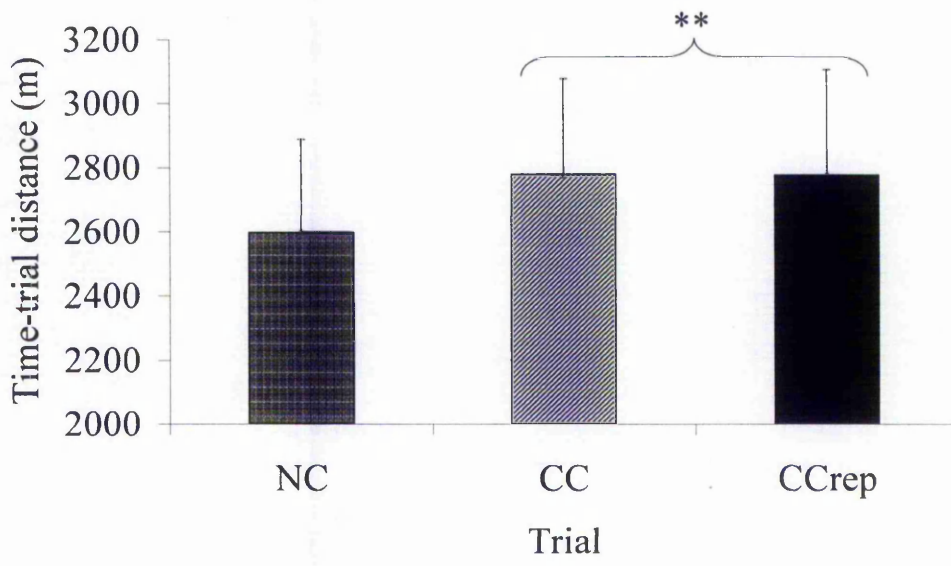

Figure 8.1. The mean (± 1 SD) distances covered during the 15 min time-trials in the no collar (NC), cold collar (CC) and cold collar replaced (CC_{rep}) trials. ** = $P < 0.01$; [a] = compared to NC. Main effect trial ($P = 0.003$).

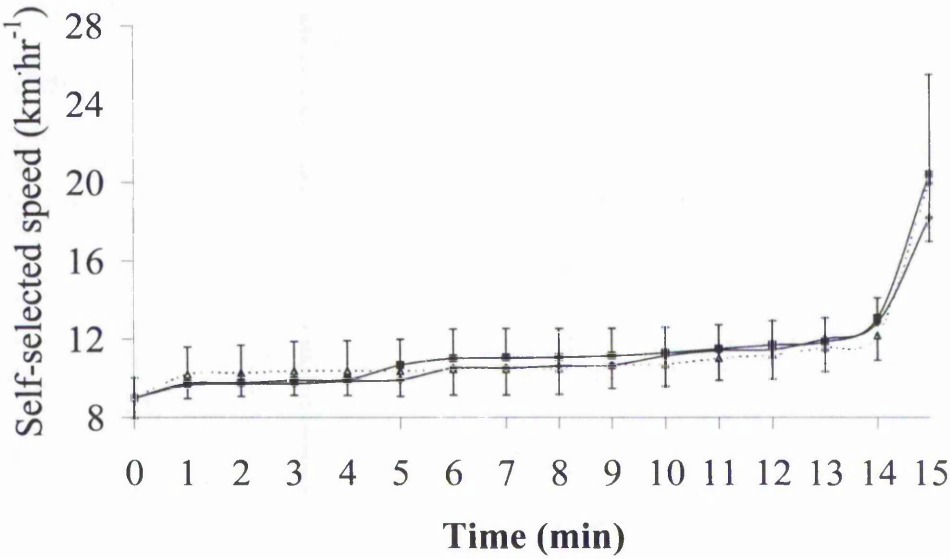

Figure 8.2. The mean (± 1 SD) treadmill speed selected during the 15 min time-trials. ♦ solid line = no collar; ■ = cold collar; ▲ dashed line = cold collar replaced.

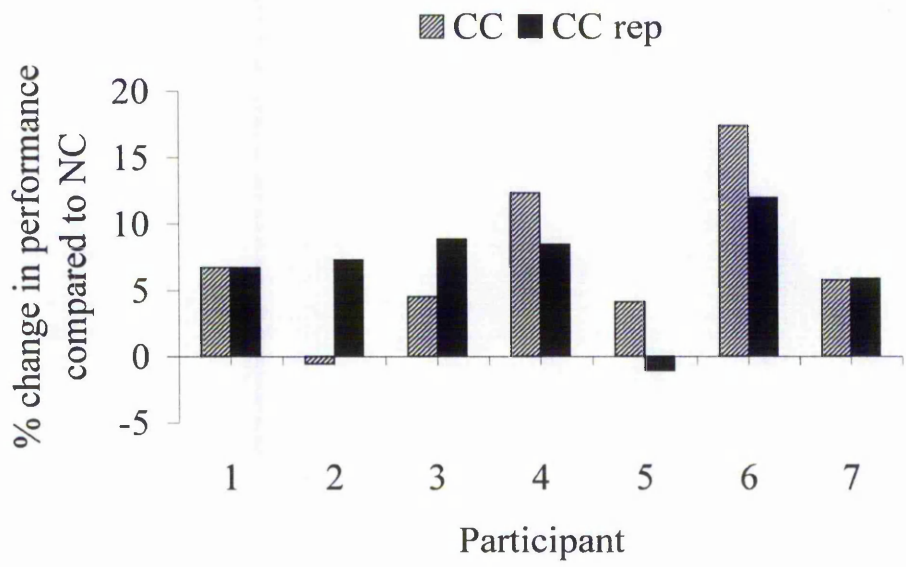

Figure 8.3. Individual performance changes (%) compared to the no collar (NC) trial for the cold collar (CC) and collar cold replaced (CC_{rep}) trials.

8.3.2. Neck temperature

Mean neck temperature (T_{neck}) is shown in Figure 8.4. There was a significant main effect for trial ($P < 0.001$), time ($P < 0.001$) and trial x time interaction ($P < 0.001$). T_{neck} was significantly colder during the 90 min TT_{pre} in CC compared to NC ($P = 0.006$); CC_{rep} compared to NC ($P < 0.001$) and CC_{rep} compared to CC ($P = 0.036$) for the 90 min protocol. There was a significant interaction effect between trials ($P < 0.001$). At the commencement of the time-trial ($t = 75$ min) neck temperature was significantly lower in CC_{rep} compared to NC and CC trials ($P < 0.001$ for both); however, there was no significant difference between NC and CC ($P = 0.467$).

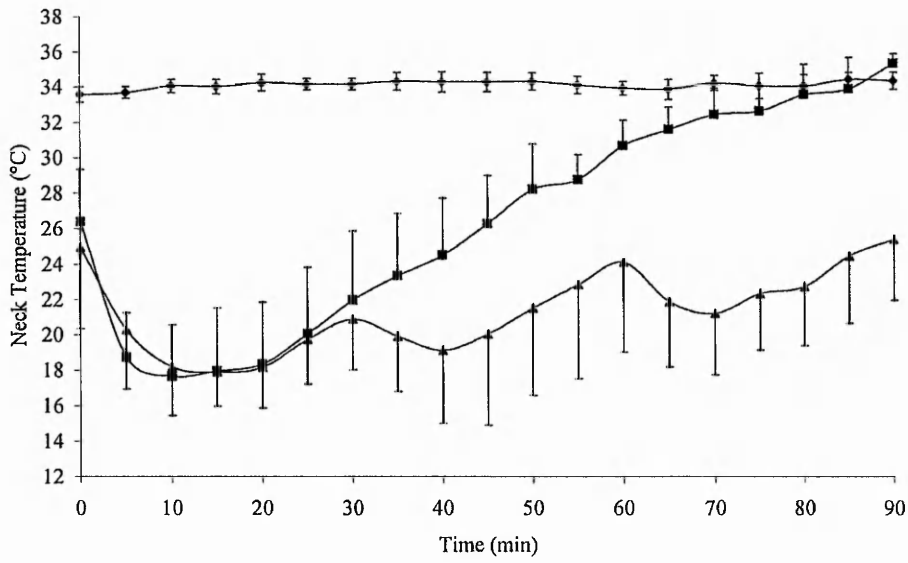

Figure 8.4. The mean (± 1 SD) neck temperature during the 90 min preloaded time-trials. ♦ = no collar (NC); ■ = cold collar (CC); ▲ = cold collar replaced (CC_{rep}). Main effect time ($P < 0.001$), trial ($P < 0.001$) and interaction ($P < 0.001$).

8.3.3. Heart rate and rectal temperature

Heart rate and T_{rectal} increased significantly over the duration of the trial ($P < 0.001$) however there were no significant differences between trials for T_{rectal} ($P = 0.320$) or HR ($P = 0.454$). Further analysis revealed that there were no significant differences between trials for T_{rectal} or HR at the commencement of the time-trial phase ($t = 75$ min) ($P = 0.496$; $P = 0.583$ respectively). The T_{rectal} and HR data observed at 0, 75 and 90 min is shown in Table 8.1.

Table 8.1. Rectal temperature and heart rate at 0, 75 and 90 min

Time (min)	Trial	Rectal temperature (°C)	Heart rate ($b \cdot min^{-1}$)
0	NC	36.68 ± 0.25	68 ± 12
	CC	36.53 ± 0.67	76 ± 16
	CC_{rep}	36.72 ± 0.28	77 ± 11
75	NC	$38.50 \pm 0.35^{**a}$	$160 \pm 14^{**a}$
	CC	$38.5 \pm 0.51^{**a}$	$158 \pm 8^{**a}$
	CC_{rep}	$38.58 \pm 0.38^{**a}$	$157 \pm 9^{**a}$
90	NC	$38.91 \pm 0.29^{**ab}$	$185 \pm 9^{**ab}$
	CC	$38.90 \pm 0.53^{**ab}$	$186 \pm 11^{**ab}$
	CC_{rep}	$38.97 \pm 0.36^{**ab}$	$185 \pm 11^{**ab}$

Values are means ± 1 SD. No significant differences existed between trials at any time-point (T_{rectal}: P = 0.391; HR: P = 0.355). ** = P < 0.01. [a] = significant difference compared to 0 min; [b] = significant difference compared to 75 min

8.3.4. Perceptual measurements

Thermal sensation (TS), neck thermal sensation (TS_{neck}) and rating of perceived exertion data are shown in Figures 8.5, 8.6 and 8.7. There were no significant main effects for TS or RPE (P = 0.069; P = 0.540); however, there was a significant interaction effect observed for TS (trial x time; P = 0.017). The TS_{neck} was significantly lower in CC_{rep} compared to NC (P < 0.001) and CC (P = 0.036) and in CC compared to NC (P = 0.006). At the beginning of the time-trial (75 min) there was no significant difference for RPE but participants reported feeling significantly cooler in CC_{rep} compared to CC (P = 0.006). These results were replicated with TS_{neck}. Participants reported a significantly lower TS_{neck} in CC_{rep} compared to CC (P = 0.001) and NC (P < 0.001) with no difference between CC and NC (P = 0.141).

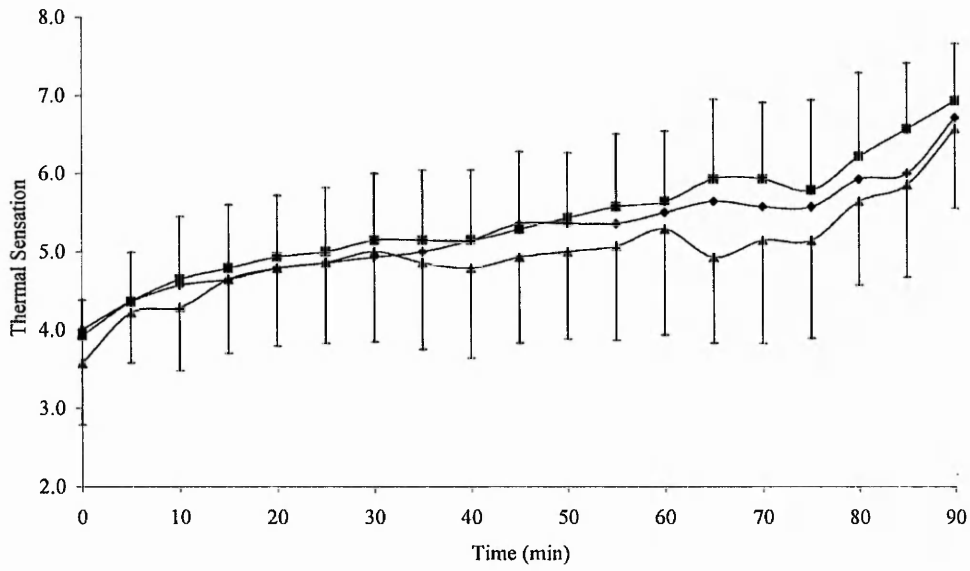

Figure 8.5. The mean (± 1 SD) thermal sensation reported during the 90 min preloaded time-trial. ♦ = no collar (NC); ■ = cold collar (CC); ▲ = collar cold replaced (CC_{rep}). For clarity SD is not shown for no collar trials, mean SD = ± 1.0. Main effect time ($P < 0.001$) and interaction ($P = 0.017$)

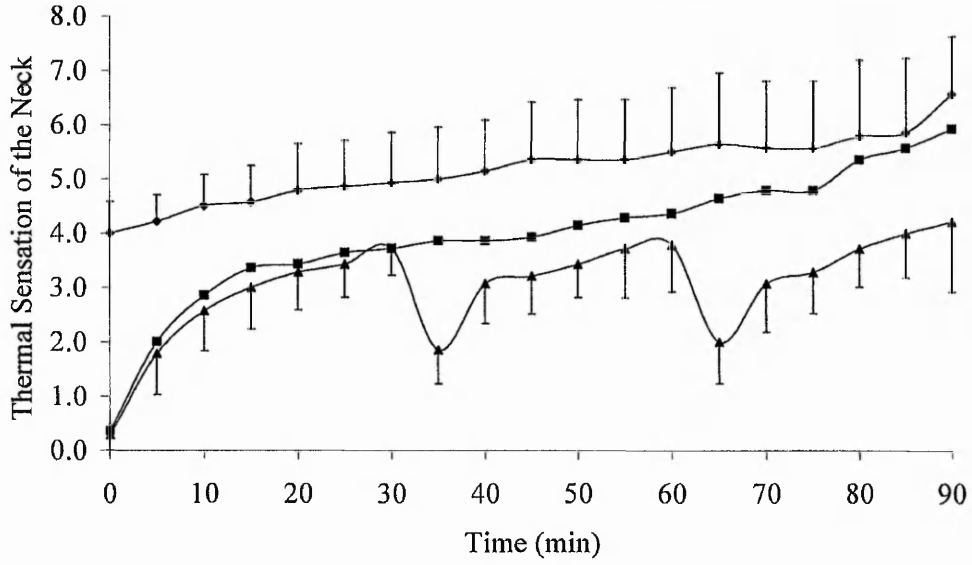

Figure 8.6. The mean (± 1 SD) thermal sensation of the neck reported during the 90 min preloaded time-trial. ♦ = no collar (NC); ■ = cold collar (CC); ▲ = cold collar replaced (CC_{rep}). For clarity SD is not shown for no collar trials, mean SD for NC = ± 1.0. Main effect trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P < 0.001$).

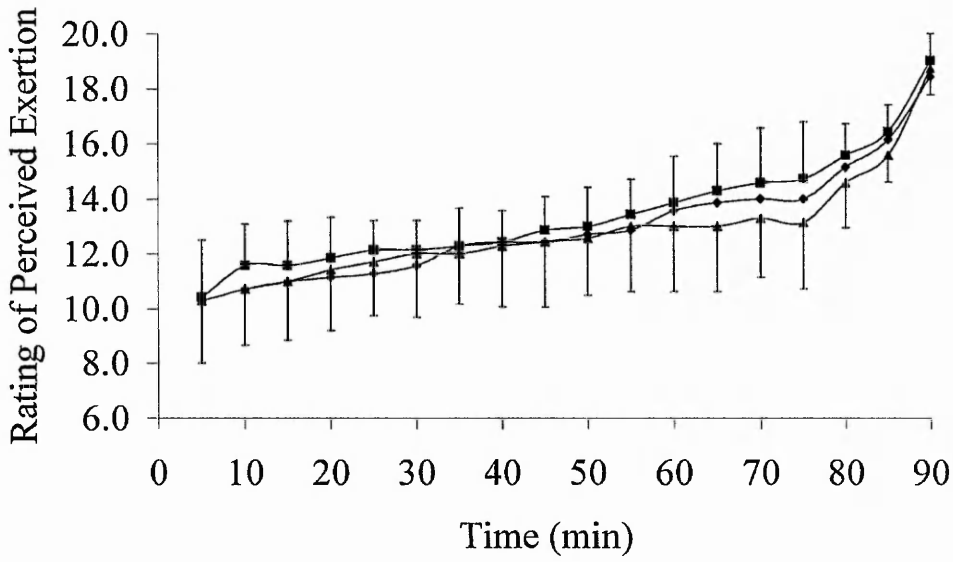

Figure 8.7. The mean (± 1 SD) rating of perceived exertion reported during the 90 min preloaded time-trial. ♦ = no collar (NC); ■ = cold collar (CC); ▲ = cold collar replaced (CC_{rep}). For clarity SD is not shown for no collar trials, mean SD = ± 2.4. Main effect time ($P < 0.001$).

8.3.5. Body fluid balance

There were no significant differences in the volume of water voluntary consumed ($P = 0.152$) or in the volume of sweat lost ($P = 0.187$) between trials. Participants consumed 0.850 ± 0.496L; 0.904 ± 0.500L and 0.683 ± 0.235L of water and lost 2.01 ± 0.39L; 2.18 ± 0.31L and 1.88 ± 0.19L of sweat during the NC, CC and CC_{rep} trials. There was a trend for participants to drink less and lose less body mass in CC_{rep} trials however due to large individual differences there were no significant differences observed. The mean plasma volume changes observed in the NC, CC and CC_{rep} were -2.4 ± 8.27, 0.4 ± 6.4 and -5.8 ± 8.2% ($P = 0.583$).

8.3.6. Blood data

Whole blood lactate and glucose concentrations are shown in table 8.2. Whole blood lactate and glucose concentrations increased over time ($P < 0.001$), although there were no significant main effect differences between trials for whole blood lactate ($P = 0.215$) or glucose ($P = 0.866$). There was no significant increase in cortisol levels over time ($P = 0.139$) or significant differences between trials ($P = 0.269$) (Figure 8.7). Plasma concentrations of serotonin and dopamine are displayed in table 8.2. Concentrations of both increased over time (main effect time; $P < 0.001$) and were significantly higher at 90 min compared to all other time points ($P = 0.003$ and $P < 0.001$); however, there was no difference between trials for either neurotransmitter ($P = 0.462$ and $P = 0.790$).

Table 8.2. Cortisol, serotonin, dopamine, lactate and glucose concentrations

	0 min	10 min	40 min	70 min	90 min
Cortisol ($nmol \cdot l^{-1}$)					
NC	370.5 ± 85.1	338.7 ± 81.9	308.2 ± 120.9	332.4 ± 170.3	387.3 ± 195.4
CC	347.8 ± 83.6	360.5 ± 79.1	332.2 ± 105.8	394.9 ± 114.4	496.3 ± 144.7
CC_{rep}	373.7 ± 95.7	409.1 ± 70.8	371.0 ± 82.6	352.5 ± 131.4	446.7 ± 175.6
Serotonin ($nmol \cdot l^{-1}$)					
NC	80.7 ± 48.5	80.7 ± 73.8	68.2 ± 42.4	73.1 ± 52.9	173.9 ± 93.8*
CC	48.0 ± 32.3	103.6 ± 118.6	134.4 ± 172.2	115.2 ± 115.3	210.2 ± 86.5*
CC_{rep}	81.8 ± 71.2	76.5 ± 57.2	57.2 ± 24.9	57.5 ± 34.3	89.2 ± 69.8*
Dopamine ($nmol \cdot l^{-1}$)					
NC	0.69 ± 0.29	0.78 ± 0.50	0.85 ± 0.26	1.02 ± 0.26	1.80 ± 0.52**
CC	0.67 ± 0.14	0.83 ± 0.23	0.93 ± 0.22	0.93 ± 0.14	1.62 ± 0.31**
CC_{rep}	0.56 ± 0.19	0.58 ± 0.15	0.76 ± 0.12	1.02 ± 0.26	1.51 ± 0.26**
Lactate ($mmol \cdot l^{-1}$)					
NC	0.94 + 0.26	1.54 + 0.63	1.56 + 0.75	1.70 + 0.79	6.19 + 1.43**
CC	1.01 ± 0.32	1.47 ± 0.43	1.71 ± 1.18	1.45 ± 0.66	6.77 ± 1.31**
CC_{rep}	0.93 ± 0.43	1.35 ± 0.38	1.37 ± 0.48	1.36 ± 0.47	5.88 ± 1.43**
Glucose ($mmol \cdot l^{-1}$)					
NC	4.17 ± 0.29	3.88 ± 0.30	4.13 ± 0.29	3.83 ± 1.12	4.71 ± 1.07*
CC	3.97 ± 0.42	3.82 ± 0.23	3.99 ± 0.44	4.01 ± 0.30	4.96 ± 0.96*
CC_{rep}	4.00 ± 0.39	3.67 ± 0.40	4.09 ± 0.31	4.11 ± 0.23	4.58 ± 0.89*

NC = no collar, CC = cold collar, CC_{rep} = cold collar replaced. ** = $P < 0.05$; ** = $P < 0.001$ difference from other time-points.

8.4. Discussion

As previously reported in Chapter 5 cooling the neck region via a practical neck cooling collar significantly improves 15 min time-trial performance in the heat when preceded by a 75 min submaximal preload phase. The data from the present study confirms this observation with an improvement of ~7.3% in the collar cold trials compared to the non-collar control trials. The main finding of the current study is that replacing the collar at regular intervals also improves time-trial performance in the heat but offers no cumulative or additional benefit and improves performance by 6.9%.

Previous literature that has investigated the effect of a cooling device during exercise on exercise performance or capacity, or on the physiological response to the bout has shown that beneficial effects are dependent on a sufficient level of cooling provided and/or thermal strain experienced (Castle *et al.*, 2006; Nunneley *et al.*, 1971). Palmer *et al.* (2001) reported that sustained cooling of the head region during a bout of rest and subsequent exercise via a water-perfused cooling hood improved 15 min treadmill performance in a hot environment (33°C; 55% rh) by ~2.5% compared to cooling at rest alone. No data was provided comparing cooling at rest to no cooling however the data provided regarding cooling at rest versus cooling at rest and during exercise offers tentative support for the notion of a cumulative benefit of sustained cooling. Such a benefit was not observed in the current study.

Palmer *et al.* (2001) reported a reduction in rectal temperature with the sustained head-cooling and due to the inverse relationship observed between exercise performance and capacity, and the levels of hyperthermia (Gonzalez-Alonso *et al.*, 1999) the improvement in performance observed could be, in part, due to this reduction. In the previous experimental studies investigating the practical cooling collar used in this study (presented in Chapters 5 and 7) the collar was shown to have no effect on the physiological or hormonal response to the exercise bout. The current study replicated these findings and it was established that replacing the collar at 30 min intervals also had no effect on the rectal temperature or heart rate response to the 90 min preloaded time-trial and that the hormonal response remained unaffected. Reductions in core temperature caused by cooling interventions appear dependent on the cooling of peripheral blood which circulates to the core (Kay *et al.*, 1999); however, the neck only forms ~10 – 12% of the body's surface area and therefore has limited potential to reduce core temperature. Previously it was proposed that peripheral hormonal concentrations are important markers of exercise stress (e.g. Brisson *et al.*, 1989); however, more recently it has been suggested that central, rather than peripheral, levels are key in regulating exercise performance in hot environments (Meeusen *et al.*, 2006; Roelands *et al.*, 2008a; Roelands *et al.*, 2008b; Roelands *et al.*, 2008c). Brisson and co-workers (1989) suggested that the effects of a cooling intervention on the peripheral hormonal concentrations was magnitude dependent but the data from the current study and that reported in Chapter 5 shows that cooling the neck has no effect on such concentrations even when the cooling is sustained and pronounced.

The improvements in performance were not matched with significant alterations in thermal sensation or ratings of perceived exertion and these results are different from those reported in the study presented in Chapter 5. In the current study participants were required to differentiate the levels of thermal comfort they experienced at the neck from the rest of their body and this was an additional measurement adopted in the present study. Thermal sensation of the neck was significantly reduced via the application of the cooling collar compared to the non collar trial and by the replacement of the collar compared to the non collar and cold collar trials. It has been proposed that the neck is an optimal site for cooling due to its proximity to the thermoregulatory centre (Shvartz, 1976); however, the extent to which cooling the neck affects perceived thermal states compared to cooling elsewhere has not been investigated. It has been established that the face is a site of high alliesthesial thermosensitivity and that cooling the face region results in a 2 – 5-fold greater suppression in thermal discomfort than cooling areas of the trunk and limbs (Cotter & Taylor, 2005) and so the neck may have similar qualities. It is possible that the lack of difference in thermal comfort reported in this study is explained by the addition of the thermal sensation specific to the neck region and previous data reporting a combined thermal comfort.

The performance benefit observed in the previous experimental study investigating the modified cooling collar (Chapter 5) was attributed to an upregulation in the pace selected due to an alteration in the level of perceived thermal comfort. In the present study there were no significant differences in the pacing strategy adopted, however Figure 8.2 demonstrates that participants initially adopted a faster pace in the cold collar replaced trials however were then unable to increase the pace selected as progressively as in the cold collar trial. This data suggests that the collar replacement may have provided a false signal which resulted in the adoption of an initial pace in excess of what was sustainable while the single application of the collar allowed for a progressive increase in pace during the performance test. The idea that the pacing strategy could be influenced by cooling the neck is due to the association between hyperthermia and the down-regulation of self-selected pace (Marino, 2004). During self-paced exercise the intensity is regulated by a complex network of feedback and feed-forward systems regarding the physiological state of the body to allow for the completion of the task within homeostatic limits (Marino, 2004; Tucker *et al.*, 2004). The data from the current study and from the one reported in Chapter 5 suggests that cooling the neck enhances preloaded time-trial performance in a hot environment by masking the extent of the thermal strain, allowing a faster pace to be selected, however there is a limit to the benefit to exercise performance that can be gained. The critical core temperature and central governor theories are the two main theories

proposed to explain the impairment in sporting performance observed in hot temperatures and both models propose that there are mechanisms in place to prevent the onset of a dangerously high internal temperature (Gonzalez-Alonso *et al.*, 1999; Marino, 2004; Tucker *et al.*, 2004; Walters *et al.*, 2000). Acute cooling of the neck can improve exercise performance in a hot environment however there appears to be a limit to the gain that can be achieved and to the extent to which the mechanisms which regulate exercise in the heat can be deceived.

Another possible reason for the lack of cumulative benefit is that there is a limit to the improvement which can be observed in a 15 min time-trial performance in a homogenous population. The fixed duration of the time-trial dramatically improves the reliability of the test (Jeukendrup *et al.*, 1996), although there is also a limited potential for performance differences and therefore it is possible that the replaced collar may have a cumulative performance benefit in longer duration tests.

8.5. Conclusion

Cooling the neck can improve time-trial performance in a hot environment, although maintaining the neck at a reduced temperature via the replacement of a practical cooling collar offers no additional benefit. Cooling the neck region does not alter the physiological or hormonal response to running exercise performed in high ambient temperatures. However, it does improve the subjective rating of thermal comfort and this improvement in thermal comfort may improve performance by masking the thermal strain of the body, allowing for the selection of a faster pacing strategy.

Chapter 9: The effect of neck cooling on running capacity in the heat

9.1. Introduction

Data from previous studies have shown that time-trial performance is impaired by ~10% in a hot environment but that cooling the neck region via a cooling collar can attenuate this reduction and improve exercise performance by 6 – 7%. Although the beneficial effect of a cooling collar on running performance was established, the finite nature of the test limited the extent to which the improvement could be explained. Exercise capacity is also impaired in hot, compared to moderate, conditions (Galloway & Maughan, 1997); however, the reason for the reduced ability to perform or maintain exercise in elevated ambient temperatures is unclear. The impairment is often attributed to the development of hyperthermia because in laboratory-based investigations exercise is consistently and voluntarily terminated at core temperatures of ~40°C regardless of the initial temperature, acclimation status or hydration levels (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993). The reason for the voluntary termination of exercise is unknown, although it has been proposed that it serves as a protective cut-off to prevent the body reaching temperatures that would lead to the onset of potentially fatal heat illness (MacDougall *et al.*, 1974; Nielsen *et al.*, 1993). Although there is consistency in the core temperatures observed at voluntary exercise termination in laboratory tests, core temperatures of up to 41.1°C have been recorded following athletic competition (Pugh *et al.*, 1967) demonstrating that the mechanisms that limit exercise in a hot environment can be overridden and that temperatures in excess of 40°C can be tolerated during exercise in sufficiently motivated athletes.

Cooling the body prior to exercise has regularly been shown to improve exercise capacity and performance in hot conditions (Kay *et al.*, 1999; Lee & Haymes, 1995). The reason for the improvement is often attributed to a reduction in core temperature at any comparative time-point during the exercise bout but improvements have occurred following reductions in skin temperature in the absence of such reductions (Hessemer *et al.*, 1984; Kay *et al.*, 1999). Cooling the skin improves thermal comfort levels (Cotter & Taylor, 2005) and this suggests that cooling-induced benefits may occur due to a reduction in the perceived, rather than actual, thermal stress or an as yet unknown cooling-induced effect. The head and face regions are regions of high alliesthesial thermosensitivity while cooling the neck region has been shown to be more effective in the alleviation of heat strain than cooling elsewhere (Shvartz, 1976) and it has been proposed that the neck may be an optimal site to cool due to its close proximity to the thermoregulation centre (Gordon *et al.*, 1990; Shvartz, 1976). The thermoregulation centre is located at the base of the brain and receives

afferent signals regarding the thermal state of the body from many deep and peripheral thermoreceptors (Hammel *et al.*, 1963). The data from the 2nd and 5th experimental study (presented in Chapters 5 and 8) demonstrate that cooling the neck region via a practical neck cooling collar can significantly enhance exercise performance in hot conditions without altering the physiological or peripheral neuroendocrinological response to the exercise bout. It has been suggested that self-paced exercise in a hot environment is regulated by integrated feedback from many physiological and perceptual systems (Marino *et al.*, 2004; Marino, 2004) and because the improvement in time-trial performance was associated with improvements in the participants' perceived level of exertion it appears that cooling this region may enhance exercise performance via the provision of a 'false' signal regarding the body's thermal state which allows for the selection of a faster, 'un-natural', pace. Maintaining the neck at a reduced temperature throughout the 90 min protocol by replacing the collar at 30 min intervals did not benefit running performance to a greater extent than the single application of the collar and therefore it appears that there is a limit to the extent that the cues limiting exercise can be deceived. If cooling the neck region does provide a false signal allowing for exercise performance to be improved it would also be expected to increase exercise capacity in the heat by masking the extent of the thermal strain experienced allowing the participant to tolerate a higher core temperature and level of thermal strain by overriding the thermal signals to terminate exercise.

As discussed in Chapter 4 exercise capacity tests are inherently more unreliable than performance tests due to the open-ended nature of the assessment; however, performance tests are unable to investigate the physiological response to steady-state exercise or the physiological variables at the point of voluntary exercise termination. To combat the natural variation associated with capacity tests the current study was designed to assess the variation of the test as well as the effect of cooling the neck on exercise capacity in the heat.

The aim of the current study was two-fold; to establish the variability of time to exhaustion in the heat and to investigate the effect of cooling the neck via the application of a practical neck cooling collar on exercise capacity in the heat. It was hypothesised that exercise capacity in a hot environment would be improved by the application of the collar and that the time to exhaustion would be extended due to an improvement in the levels of perceived strain experienced.

9.2. Methods

9.2.1. Participants

Eleven healthy, endurance trained males volunteered for the study but due to illness three volunteers failed to complete all trials and therefore their data has been omitted. The mean (± 1 SD) age, height, body mass (BM) and maximal oxygen uptake ($\dot{V}O_{2max}$) of the eight participating volunteers was 26 ± 2 y, 1.79 ± 0.04 m, 77.0 ± 6.2 kg and 56.2 ± 9.2 $ml \cdot kg^{-1} \cdot min^{-1}$. The participants were fully informed of any risks and discomforts associated with the study before giving their informed written consent to participate and completing a health screen. The health screening procedure was repeated prior to each laboratory visit to ensure the health status of the participants. The study was approved by the institution's Ethical Advisory Committee.

9.2.2. Experimental procedures

Prior to the main trials, participants completed an incremental motorised treadmill test to determine $\dot{V}O_{2max}$ as per Jones and Doust (1996). Following the preliminary test participants visited the laboratory on four occasions, 7-days apart, at the same time of the day on each occasion ± 30 min. All trials were conducted in hot ambient conditions (32.2 ± 0.2°C; 53 ± 2% rh). During all four visits to the laboratory participants completed an 8 min standardised warm-up followed by a treadmill test to exhaustion at a speed set to elicit 70% of $\dot{V}O_{2max}$. The 8 min standardised warm-up consisted of 2 min at 50% $\dot{V}O_{2max}$ (8.6 ± 1.3$km \cdot h^{-1}$); 1 min at 60% $\dot{V}O_{2max}$ (10.1 ± 1.6$km \cdot h^{-1}$); 1 min at 70% $\dot{V}O_{2max}$ (11.9 ± 1.8$km \cdot h^{-1}$); 1 min at 50% $\dot{V}O_{2max}$ and 3 min of stretching. The first two trials were both non-collar trials and formed two habituation trials (FAM1 and FAM2). Participants ran wearing a neck cooling collar (CC) (see section 3.6.2.6. for specification) during one of the other two visits to the laboratory (visit 3 or 4) and ran without in the other (NC). The collar was applied after the 8 min warm-up period. The order of the neck cooling collar trial and third non-collar trial was randomised and counter-balanced. Cooled-water was allowed *ad libitum* throughout the trials. The data from the three trials conducted without the cooling collar intervention (FAM1, FAM2 and NC) were used to assess the individual and mean variability in capacity time.

Participants abstained from alcohol and caffeine and completed a food record for the 24h prior to the initial experimental trial. They adopted the same diet and abstained from strenuous exercise for 24h, prior to each main trial. Participants arrived at the laboratory ~30 min before the commencement of the trial and ≥ 4h post-prandial. On arrival, nude body mass was recorded. A rectal probe (Grant Instruments (Cambridge) Ltd, England, U.K.) was self-inserted ~10cm past the anal sphincter and a heart rate (HR) monitor (Polar Electro Oy, Kempele, Finland) was attached prior to the participant entering the environmental chamber (Design Environmental WIR52-20HS, Design Environmental Ltd., Gwent, Wales, U.K.). During the final two visits (NC and CC trials) four skin thermistors (Grant Instruments (Cambridge) Ltd, England, U.K.) were attached evenly across the posterior aspect of the neck. Heart rate, rectal temperature, mean neck temperature, rating of perceived exertion (Borg, 1982), thermal sensation (TS) and thermal sensation of the neck region (TS_{neck}) were recorded at 5 min intervals and at the point of exercise termination. Mean neck temperature was calculated as the mean temperature of the four skin thermistors. All thermistors were attached via a transparent dressing (Tagaderm, 3M Health Care, USA) and water-proof tape (Transpore, 3M Health Care, USA). TS was rated with an eight-point scale, ranging from 0 (unbearably cold) to 8 (unbearably hot) with 4 as comfortable (neutral) (Young *et al.*, 1987)- participants were asked to differentiate between whole-body and neck thermal sensation during the final two trials. A finger-prick blood sample was taken during the stretching phase of the warm-up period and immediately at the point of exercise termination for the determination of whole blood lactate and glucose (Yellow Springs Instrument 2300 STAT plus, Yellow Springs Instruments Inc., Ohio, USA). Time to exhaustion was recorded in all four trials. Following the completion of each trial participants towel-dried and recorded a post-exercise nude body mass from which sweat loss and percentage body mass loss was calculated taking into account voluntary fluid consumption.

9.2.3. Statistical analysis

Data are presented as mean ± 1 standard deviation. Data for capacity, variability, sweat loss, fluid consumption and variables at exhaustion were analysed using paired t-tests. Two-way ANOVA with repeated-measures were conducted on paired physiological and perceptual data points. Following a significant F value, Tukey's HSD *post hoc* tests were conducted to identify pair-wise differences. Time to exhaustion data from the three non-collar trials were used for the calculation of the test to exhaustion's reliability. Individual coefficient of variations were calculated for each participant between pairs of trials (*i.e.*

between non-collar trials 1 and 2 and between non-collar trials 2 and 3) from which a mean CV for FAM1 and FAM 2 and FAM2 and NC trial pairings was calculated. Significance was set at the $P < 0.05$ level.

9.3. Results

9.3.1. Exercise capacity variability

The coefficient of variation between trial pairings was 8.7% between FAM1 and FAM2 and 8.0% between FAM2 and NC (Figure 9.1). There was no significant difference between the coefficient of variation for the two trial pairings ($P = 0.768$).

9.3.2. Exercise capacity

Participants ran significantly longer in the cold collar trial compared to the no collar trial (38:12 ± 11:42 v 43:09 ± 12:49 min; $P < 0.001$) (Figure 9.1.). The exercise capacity was improved for all participants with the individual percentage improvement observed ranging from 11.1 to 24.4% (Figure 9.2). The mean percentage improvement observed in the cold collar trial compared to the experimental non-collar trial was 13.5 ± 3.8% (Figure 9.1).

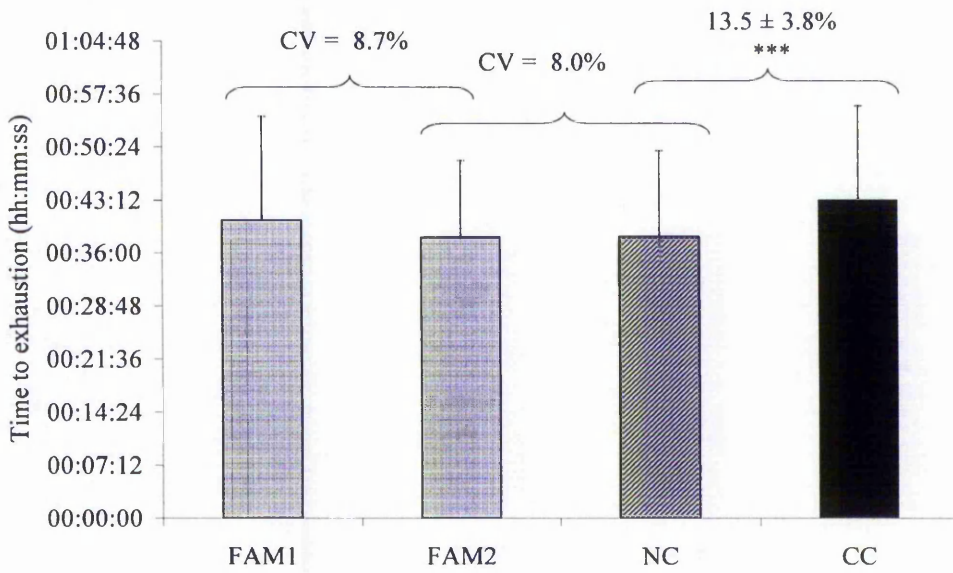

Figure 9.1. The mean (± 1 SD) exercise capacity times for the reliability (FAM1, FAM2 and NC) and experimental (NC and CC) trials. CV = coefficient of variation. *** = $P <$ 0.001 between trials.

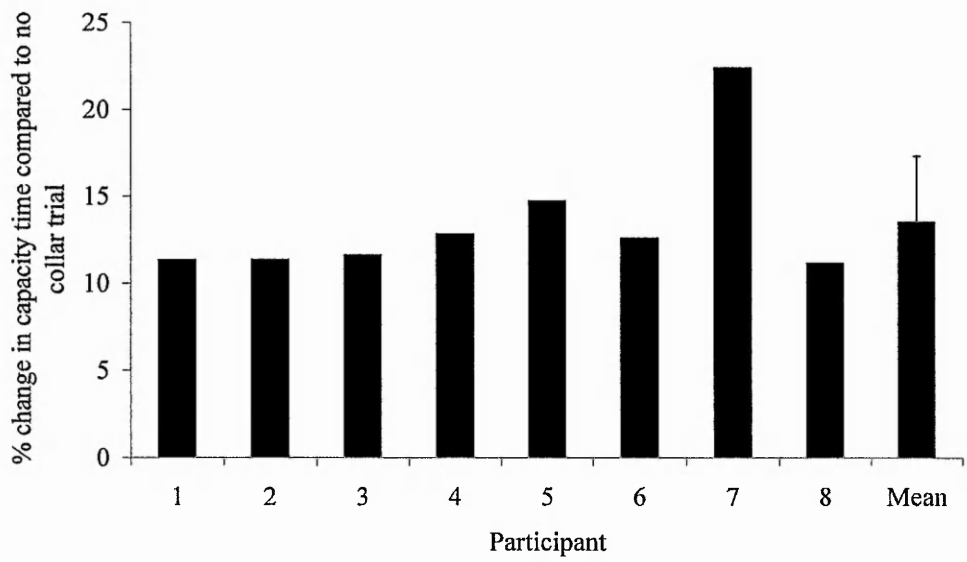

Figure 9.2. Individual and mean (± 1 SD) change in exercise capacity with the cooling collar.

9.3.3. Neck temperature

The application of the neck cooling collar resulted in a significantly lower mean neck temperature in the cold collar trial ($P < 0.001$) (Figure 9.3). The mean difference in neck temperature was greatest after 5 min (-17.91 ± 3.95°C) and lowest at exercise termination (-8.11 ± 4.59°C); however, despite the decreased cooling effect of the collar during the course of the trial, the neck temperature remained significantly lower throughout the test ($P < 0.001$).

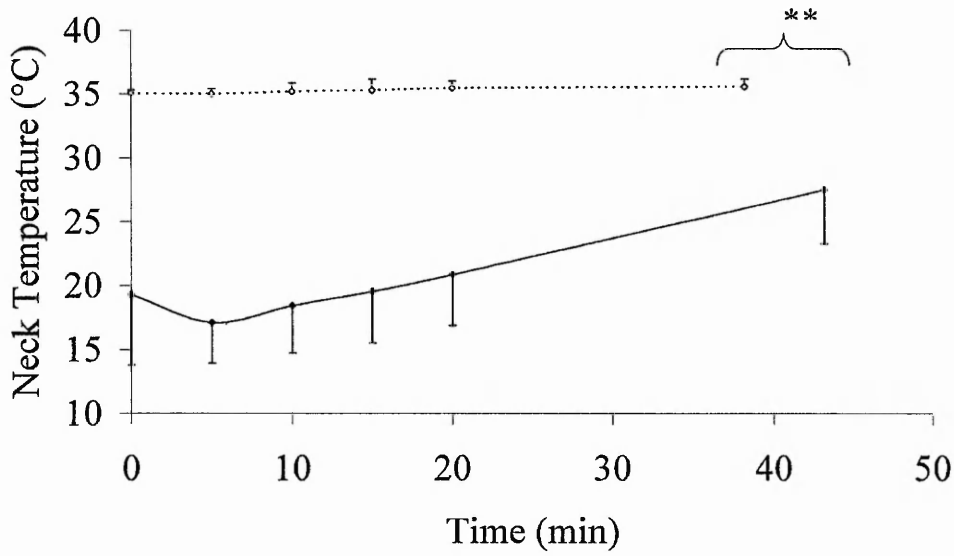

Figure 9.3. The mean (± 1 SD) neck temperature observed in the cold collar and no collar trials. Open with dashed line = no collar (NC); filled with solid line = cold collar (CC). ** = $P < 0.01$ between trials. Main effect trial ($P < 0.001$), time ($P < 0.001$) and interaction ($P < 0.001$).

9.3.4. Heart rate and rectal temperature

Wearing the cooling collar resulted in a significantly elevated rectal temperature and heart rate ($P = 0.003$ and $P = 0.023$ respectively) (Figure 9.4 and 9.5). Participants voluntarily terminated exercise at a significantly higher rectal temperature (39.18 ± 0.7°C v 39.61 ± 0.45°C; $P = 0.015$) and heart rate (181 ± 6 v 178 ± 9 b·min^{-1}; $P = 0.03$) in the cold collar trial compared to the no collar trial.

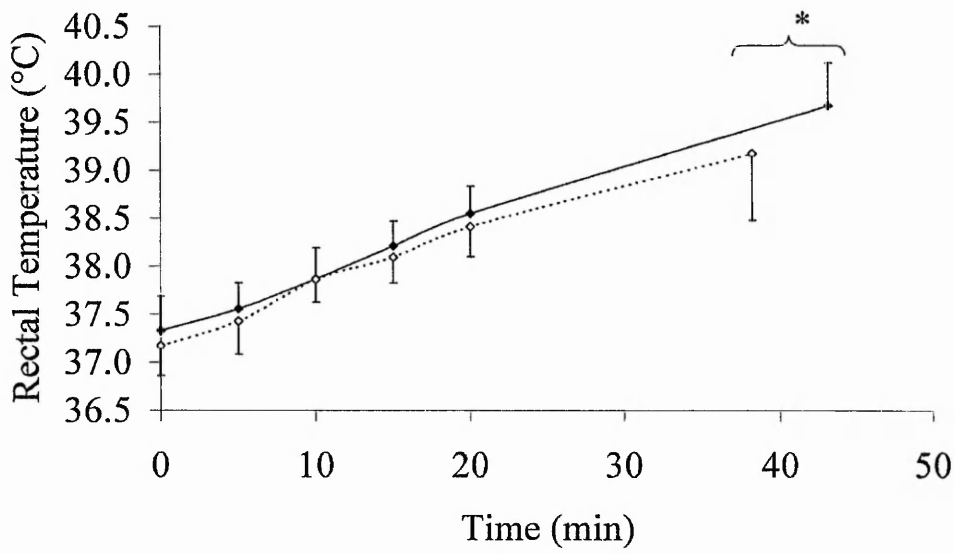

Figure 9.4. The mean (± 1 SD) rectal temperature observed in the cold collar and no collar trials. Open with dashed line = no collar; filled with solid line = cold collar. * = $P < 0.05$ between trials. Main effect trial ($P = 0.003$), time ($P < 0.001$) and interaction ($P = 0.016$).

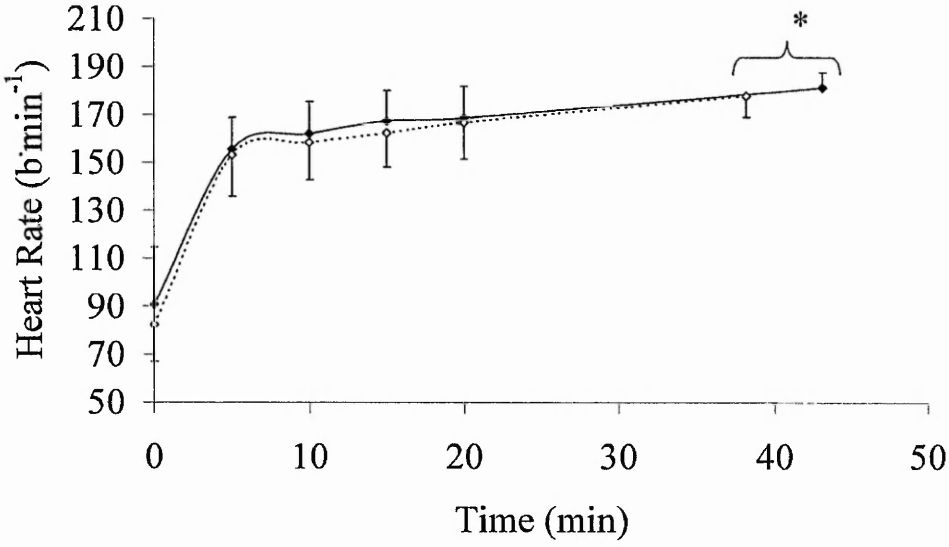

Figure 9.5. The mean (± 1 SD) heart rate observed in the cold collar and no collar trials. Open with dashed line = no collar; filled with solid line = cold collar. * = $P < 0.05$ between trials. Main effect trial ($P = 0.023$) and time ($P < 0.001$).

9.3.5. Perceptual measurements

The data for thermal sensation of the body and neck and the rating of perceived exertion are shown in Figures 9.6, 9.7 and 9.8 respectively. The application of a cooling collar significantly enhanced the perceived levels of whole-body and neck thermal comfort (main effect trial: $P = 0.007$ and $P = 0.001$ respectively) although it had no effect on the rating of perceived exertion (main effect trial: $P = 0.691$). There was no difference in the whole-body thermal sensation or ratings of perceived exertion at the termination of exercise between trials ($P = 0.142$ and $P = 1.00$ respectively); however, thermal sensation of the neck was significantly lower in the cold collar trial at the end of the trial ($P = 0.006$).

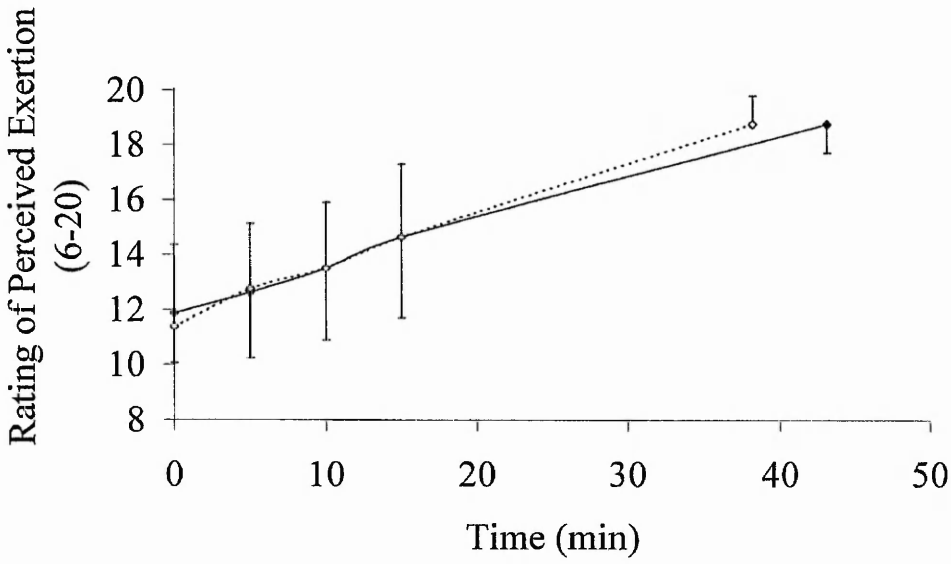

Figure 9.6. The mean (± 1 SD) rating of perceived exertion observed in the cold collar and no collar trials. Open with dashed line = no collar; filled with solid line = cold collar. Main effect time ($P < 0.001$).

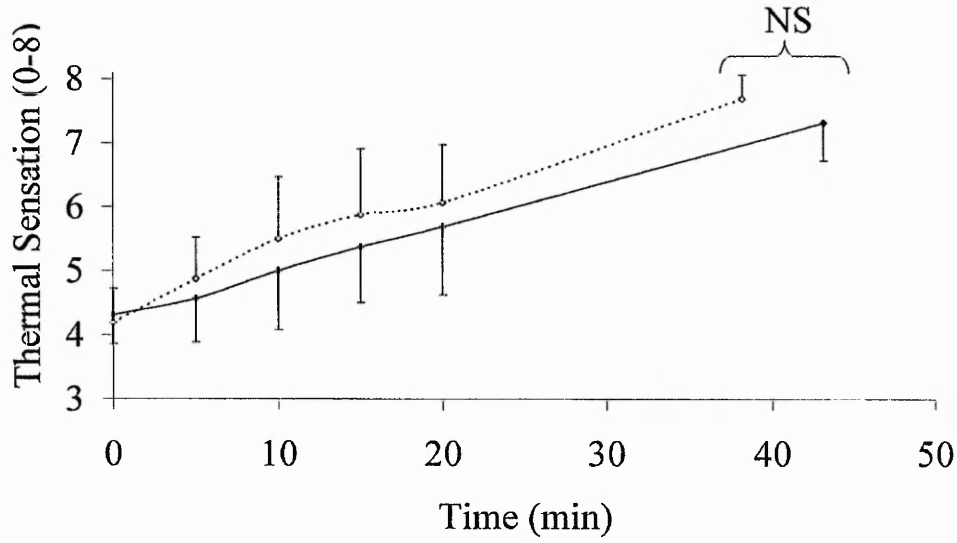

Figure 9.7. The mean (± 1 SD) thermal sensation observed in the cold collar and no collar trials. Open with dashed line = no collar; filled with solid line = cold collar. Main effect trial ($P = 0.007$) and time ($P < 0.001$).

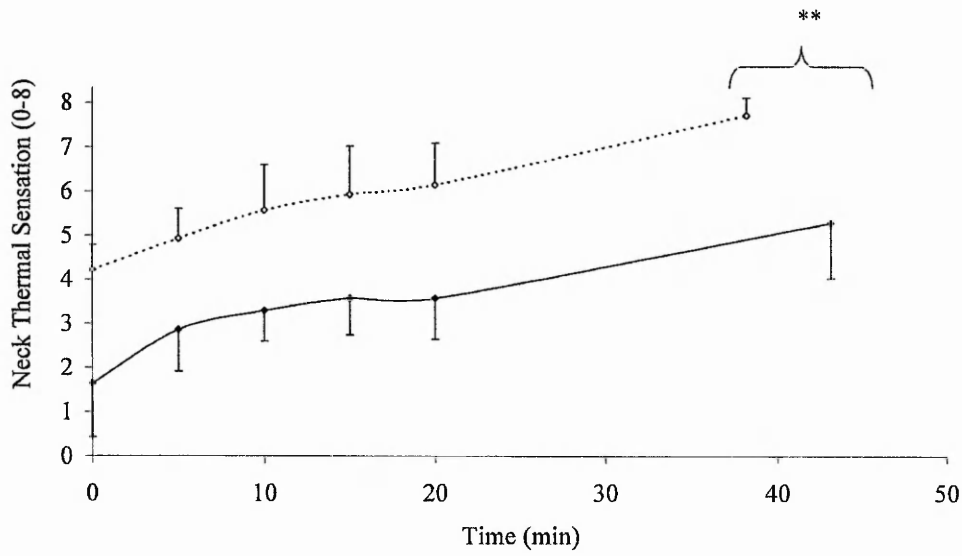

Figure 9.8. The mean (± 1 SD) thermal sensation for the neck observed in the collar cold and non-collar trials. Open with dashed line = non-collar; filled with solid line = collar cold. ** = $P < 0.01$ between trials. Main effect trial ($P = 0.001$) and time ($P < 0.001$).

9.3.6. Body fluid balance

There was no significant difference in the amount of chilled water voluntarily consumed in non-collar and cold collar trials (452 ± 220 v 427 ± 253 ml respectively; $P = 0.501$). There was also no significant difference in the total sweat lost in non-collar (1.51 ± 0.83 L) and cold collar (1.55 ± 0.53 L) trials ($P = 0.825$) resulting in participants losing ~1.5% of their body mass during each trial (1.46 ± 1.29 v 1.46 ± 0.74%; $P = 1.00$).

9.3.7. Blood data

Concentrations of whole blood lactate and glucose were significantly higher at the end of exercise compared to the beginning ($P = 0.010$ and $P = 0.003$) but there were no significant differences observed for whole blood lactate ($P = 0.279$) or glucose ($P = 0.584$) between cold collar and non collar trials. The final lactate concentrations observed in the non collar and cold collar trials were 2.66 ± 1.81 $mmol \cdot L^{-1}$ and 2.77 ± 1.96 $mmol \cdot L^{-1}$.

9.4. Discussion

As previously reported in the experimental studies presented in Chapters 5, 7 and 8, cooling the neck region via the application of a cooling collar enhances time-trial performance in the heat when the thermal strain is of a sufficient magnitude. These findings led to the hypothesis that, when the body is under a sufficient thermal strain, cooling the neck region provides a false representation of the thermal state of the body allowing for the selection of a faster pace than would be selected without the intervention. The main findings from the current study support this hypothesis. Exercise capacity was improved when wearing the cold collar and participants voluntarily terminated exercise at identical perceptions of exertion and thermal stress despite higher rectal temperatures and heart rates in the cold collar trials.

As demonstrated in the study presented in Chapter 4 exercise performance is impaired in hot compared to moderate environmental conditions and during such self-paced exercise performance tests this impairment is attributed to a centrally regulated decrease in the pace selected (Marino *et al.*, 2004; Marino, 2004). The mechanisms governing this decrease are unknown, although it has been proposed that feedback from a variety of peripherally and centrally located receptors (*e.g.* thermoreceptors, chemoreceptors, baroreceptors) provides information regarding the state of the body and results in the selection of a pacing strategy which enables the task to be completed within homeostatic limits (Marino *et al.*, 2004; Marino, 2004; Morrison *et al.*, 2004; Tucker *et al.*, 2004). This 'central governor' theory was developed in opposition to the critical core temperature hypothesis, which proposes that exercise in the heat is limited due to the obtainment of a dangerously high core temperature (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993); however, the models have very similar themes and are therefore not as contrasting as previously suggested. The central governor theory is a theory that can only be applied to self-paced performance tests whereas the critical core temperature theory can only be applied to fixed-intensity capacity tests and this fundamental methodological difference explains much of the conflict. The data presented in Chapters 5 and 8 shows that performance can be enhanced in a hot environment with the application of a cold collar without any alterations in the physiological response to the exercise bout. It seems prudent to suggest that the application of the cooling collar might have provided a false signal regarding the body's thermal strain which allowed a faster pace to be adopted. The concept of the false signal was difficult to fully elucidate using the performance test model.

Exercise capacity is also impaired in a hot environment (Galloway & Maughan, 1997) and in such exercise tests, when the intensity is fixed, the point of voluntary exercise termination regularly occurs at core temperatures of approximately 40°C in a laboratory setting regardless of the initial rectal temperature or acclimation status (Gonzalez-Alonso *et al.*, 1999; Nielsen *et al.*, 1993). Core temperatures in excess of 40°C have been reported following competitive marathon races and this demonstrates that high internal temperatures can be tolerated, and that the mechanisms limiting exercise can be overridden, albeit in conditions where there is a sufficient incentive and a high level of motivation (Pugh *et al.*, 1967). Data from the current study shows that exercise capacity is enhanced, and that exercise is terminated at a higher rectal temperature, when the neck region is cooled via the application of a neck cooling collar. Participants voluntarily terminated exercise at a rectal temperature of 39.18 ± 0.70°C in the no collar trials but they did not cease exercising until reaching temperatures approximately 0.43°C higher (39.61 ± 0.45°C) in the cold collar trials. The heart rate at the termination of exercise was also significantly higher in the cold collar trial but the subjective perceptual measurements revealed that at the point of termination there was no significant difference in the level of thermal comfort or rating of perceived exertion. This data shows that participants reached voluntary exhaustion at the same perceived levels of thermal and physical stress and discomfort despite being under significantly less thermal and cardiovascular stain in the no collar trial. Similar findings were presented in two recent pharmacological investigations in which cerebral concentrations of dopamine and noradrenaline were manipulated (Roelands *et al.*, 2008a; Watson *et al.*, 2005c). Watson *et al.* (2005a) and Roelands *et al.* (2008c) both reported that higher levels of thermal and cardiovascular strain could be tolerated when cerebral dopamine concentrations were elevated and that the perceptual response to the level of strain was dampened. Peripheral concentrations of dopamine are not elevated by the application of the cooling collar (Chapter 5 and 8); however, peripheral concentrations of neurotransmitters may not accurately represent cerebral levels (Fernstrom & Fernstrom, 2006). The excitability of cerebral dopaminergic neurons is temperature-dependent (Guatteo *et al.*, 2005) and so it is possible that cerebral dopamine concentrations may be elevated by the application of the cooling collar.

Pre-cooling studies often report an improvement in subsequent exercise performance and capacity and attribute the improvement to a reduction in the rectal temperature at any given comparative point however improvements in a subsequent exercise bout have been observed without reductions in core temperature (Hessemer *et al.*, 1984). This suggests that the benefit observed may not be dependent on a reduction in core temperature and that

the benefit may be due to an alternative cooling-induced alteration in the actual or perceived state of the body. It was hypothesised that the cooling collar enhanced the time-trial performance in Chapters 5 and 8 via the provision of a false signal regarding the body's thermal state. The data from Chapter 8 suggests that there is a limit to the extent to which the body can be deceived, or that any related adaptations are slow in their progression; however, the ability to offer a false signal resulting in an improved exercise performance has been demonstrated in situations when there is a sufficient thermal strain to require masking. The data from the current study demonstrates that, during fixed intensity exercise, cooling the neck region can dampen the perceived levels of thermal sensation and rating of perceived exertion because participants reported feeling the same despite higher core temperature and heart rate. In previous studies the collar had no effect on the core temperature or heart rate response to the exercise bout but in these studies the participants were exercising in cooler temperatures (30 v 32°C) and at a lower intensity for the majority of the test (60% v 70% $\dot{V}O_{2max}$). The effectiveness of cooling the head region has been shown to be dependent on the level of thermal strain experienced (Nunneley *et al.*, 1971) and this may explain the differences observed. The elevation in core temperature despite the fixed work intensity suggests that cooling the neck region may alter the thermoregulatory drive and suppress the heat loss mechanisms. There were no differences in the sweat loss observed but sweat loss estimation via nude body mass weighing may have lacked the sensitivity to detect such changes. Despite the higher physiological strain participants reported no differences in their perceptual scales. A similar dampening of perceived variables at comparative time-points during exercise has been reported in investigations that have pharmacologically elevated cerebral levels of dopamine (Roelands *et al.*, 2008a; Watson *et al.*, 2005c). The effects of cooling the neck, or elsewhere, on cerebral concentrations of dopamine warrants further investigation.

9.5. Conclusion

Cooling the neck region via a practical neck cooling device improves exercise capacity in the heat. Cooling the neck region allows participants to tolerate a higher rectal temperature and heart rate before they voluntarily terminate exercise at identical levels of perceived thermal comfort and ratings of perceived exertion. This data suggests that cooling this region masks the true state of the body, delaying the point at which the voluntary termination of exercise occurs, possibly due to cooling-induced alterations in cerebral dopamine concentrations.

Chapter 10: General Discussion

10.1. Introduction and key findings

The investigations presented in the six preceding experimental Chapters examined the effects of a cooling collar on running performance and capacity in a hot environment and on the physiological, perceptual and neuroendocrinological responses to the cooling intervention. The key results are summarised below and presented in table 10.1. In table 10.1 they are presented alongside data from the other two investigations that have researched the effect of a practical cooling device on exercise in a hot environment (Bulbulian *et al.*, 1999; Gordon *et al.*, 1990) to allow for comparison between results.

1. Data presented in Chapter 4 demonstrates that a 90 min preloaded treadmill time-trial (as used in the experimental Chapters 5, 7 and 8) was a reliable test of performance in hot and moderate environments. It showed that 15 min time-trial performance was impaired by ~10% in hot compared to moderate temperatures and that trials in hot conditions can be conducted seven-days apart without heat acclimation occurring.

2. Cooling the neck region, via the application of a practical cooling collar, improved treadmill running performance and capacity in a hot environment. Performance of a 15 min time-trial was improved by ~6 – 7% when the exercise performance test was preceded by a 75 min preload phase (Chapters 5 and 8); however, it was not improved when preceded by a 5 min warm-up (Chapter 7). Maintaining a cool neck throughout the 90 min exercise bout offered no additional benefit to cooling applied only at the start of the trial (Chapter 8).

3. The application of a practical neck cooling collar significantly reduced the temperature of the site that it is intended to cool but had no effect on rectal temperature, heart rate, or the sweat and peripheral neuroendocrinological responses to prolonged sub-maximal exercise in a hot environment (Chapters 5 – 8). During more demanding exercise cooling the neck can elevate rectal temperature and heart rate (Chapter 9).

4. The collar had no effect on serum S100β concentrations during exercise in a hot environment but the effect of cooling the neck on the maintenance of blood-brain barrier integrity and neuroprotection remains an interesting health-related research topic.

5. The cooling collar had no effect on the perception of effort, as measured via the rating of perceived exertion (Chapters 5 – 9) but improved the level of thermal comfort (either whole-body or neck-region specific). When the body was under sufficient thermal strain cooling the neck region dampened the perceptual response to exercise at comparative time points. Despite running further (Chapter 5 and 8) or for longer (Chapter 9) participants did not report feeling hotter or worse while wearing the collar device. The dampening of perceptual feedback allowed individuals to tolerate higher levels of thermal and cardiovascular strain (Chapter 9).

The following discussion will summarise the data with reference to pertinent existing literature and evaluate the potential mechanism where-by the cooling of the neck, via the application of a practical cooling collar, can enhance exercise performance and capacity in a hot environment.

Table 10.1. Summary of investigations cooling the neck region during exercise via the application of a practical neck cooling collar

Ref	Participants	Protocol	Cooling intervention	Neck temp	Perf	Core temp	Heart rate	RPE	TS	Sweat rate
Bulbulian *et al.*, (1999)	N = 20 (10 males; 10 females)	30-min CE @ 60%VO_{2max} T_{amb} = 30°C; 25% rh	Head and neck via cooling collar	↓**	-	↔	↔	↔	↔	↔
Gordon *et al.*, (1990)	N = 10 male	45-min submaximal run T_{amb} = 21°C	Neck via cooling collar	-	-	End ↓*	↔	↔	↔	↓*
Chapter 5i	**N = 9 male**	**90-min TT_{pre} T_{amb} = 30°C; 50% rh**	**Modified black ice cooling collar worn for 90-min**	**↓***	**Improved ***	**↔**	**↔**	**↔**	**↓***	**↔**
Chapter 5ii	**N = 9 male**	**90-min TT_{pre} T_{amb} = 30°C; 50% rh**	**Modified black ice cooling collar uncooled worn for 90-min**	**↔**	**↔**	**↔**	**↔**	**↔**	**↑***	**↔**
Chapter 7	**N = 8 male**	**5-min WU followed by 15-min TT_{pre} T_{amb} = 30°C; 50% rh**	**Modified black-ice cooling collar worn for 15-min**	**↓***	**↔**	**↔**	**↔**	**↔**	**↓***	**↔**
Chapter 8i	**N = 7 male**	**90-min TT_{pre} T_{amb} = 30°C; 50% rh**	**Modified black ice cooling collar worn for 90-min**	**↓***	**Improved ** **	**↔**	**↔**	**↔**	**↔**	**↔**
Chapter 8ii	**N = 7 male**	**90-min TT_{pre} T_{amb} = 30°C; 50% rh**	**Modified black ice cooling collar worn for 90-min (replaced at 30 and 60-min)**	**↓***	**Improved ** **	**↔**	**↔**	**↔**	**↔**	**↔**
Chapter 9	**N = 8 male**	**8-min WU followed by T_{exh} @ 70% VO_{2max} T_{amb} = 32°C; 50% rh**	**Modified black-ice cooling collar worn during T_{exh}**	**↓***	**Improved *** **	**End ↑***	**End ↑***	**End ↔**	**End ↔**	**↔**

↑ = increase; ↓ = decrease; ↔ = no change; - = not measured; * P = <0.05; **P = <0.01; ***P < 0.001; T_{exh} = test to exhaustion; TT_{pre} = preloaded time-trial; T_{amb} = ambient temperature; Perf = performance, temp = temperature, RPE = rating of perceived exertion, TS = thermal sensation; ROR = rate of rise; END = at exercise termination

10.2. The effect of a cooling collar on running performance and capacity, and on the physiological, perceptual and neuroendocrinological response to exercise in a hot environment

The results presented in Chapters 5, 8 and 9 within this thesis demonstrate that cooling the neck region, via the application of a practical cooling collar, improves preloaded time-trial performance and exercise capacity in a hot environment. The effects of practical cooling devices on exercise performance or capacity have not been previously investigated and therefore these are the first studies to show that such an intervention can benefit these parameters in a hot environment. Similarly, to the author's knowledge there have been no other studies investigating the effect of cooling the neck region on exercise capacity or performance in the heat. Although they did not focus on cooling the neck, two studies have investigated cooling the face and head. Cooling the face via the combination of a fine water-mist and facial fanning improved cycling capacity in a hot environment (29°C; 50% rh) by ~51% (Ansley *et al.*, 2007); while cooling the head via a water-perfused garment during a period of rest and subsequent treadmill exercise improved the distance covered in a 15 min time-trial by 3.3 ± 3.4% in the heat (33°C) (Palmer *et al.*, 2001). The collar that was investigated in the current thesis improved 90 min preloaded time-trial performance by ~6 – 7% (Chapters 5 and 8) and improved capacity by ~14% (Chapter 9) in temperatures similar to those investigated by Ansley *et al.* (2007) and Palmer *et al.* (2001). The application of the collar had no effect; however, on 15 min time-trial performance when the performance bout was preceded by a 5 min warm-up.

The reduction observed in exercise performance and capacity in hot environments is often attributed to the obtainment of a high internal temperature (Gonzalez-Alonso *et al.*, 1999; Tucker *et al.*, 2004). It has been suggested that it is the obtainment of a high brain, rather than trunk, temperature that ultimately limits exercise (Caputa *et al.*, 1986); however, in an exercise setting, core temperature (measured at the rectum or oesophagus) is used as a surrogate measurement. Many cooling studies that have reported performance and capacity benefits have attributed the enhanced exercise ability to reductions in core temperatures at comparative time-points (*e.g.* Booth *et al.*, 1997), although others have shown that reductions in core temperatures are not required to improve performance (Hessemer *et al.*, 1984; Kay *et al.*, 1999). Hessemer *et al.* (1984) reported that participants achieved a 6.8% greater (172W v 161W) mean work rate following pre-cooling compared to the control trial in a 60 min cycle ergometer time-trial performed in moderate temperatures (18°C) despite no significant main effect between conditions for oesophageal temperature. Skin

temperature was significantly lowered by the pre-cooling intervention and a reduction in skin temperature in the absence of a reduced core temperature can result in improved time-trial performance (Kay *et al.*, 1999; Hessemer *et al.*, 1984). Due to technical difficulties, skin temperature was not measured in the current thesis; however, time-trial performance improvements were observed without alterations in rectal temperature while capacity was improved despite increases observed.

In addition to having no effect on rectal temperature during the preloaded time-trial, cooling the neck via the practical cooling collar had no effect on the heart rate or peripheral neuroendocrinological responses to the exercise bout (Chapter 5 and 8). Cooling the neck also had no effect on the ratings of perceived exertion (Chapters 5, 7, 8 and 9) but reduced the thermal sensation reported (Chapters 5 and 7). Whole-body thermal sensation was not attenuated with the neck-cooling in the final preloaded time-trial investigation (Chapter 8) and this may have been because participants were asked to differentiate between the thermal sensation of the whole-body and of the neck region in this study but were not in the previous studies. Participants were asked to differentiate between the two sites in the capacity study and reported that the collar significantly attenuated both whole-body and neck thermal sensation. The effectiveness of a cooling device appears heavily dependent on the difference between the level of thermal strain experienced and the magnitude of cooling provided (Nunneley *et al.*, 1971) and the differences in the strain experienced due to the differing intensities (60% v 70% $\dot{V}O_{2max}$) and environmental temperatures (30 v 32°C) investigated.

It appears that the performance and capacity benefit gained by the neck-cooling device occurs as a result of a false signal regarding the physiological state of the body during the exercise bout. The central governor model has been proposed to explain the reduced ability to exercise in a hot environment during self-paced testing models (Marino, 2004). During fixed-intensity testing, the attainment of a high core temperature results in the termination of exercise (Gonzalez-Alonso *et al.*, 1999; Walters *et al.*, 2000), yet, in self-paced activity, work-rate is down-regulated well before the attainment of a very high core temperature (Marino *et al.*, 2004; Tatterson *et al.*, 2000). The central governor theory proposes that this down-regulation in self-selected work-rate allows for the task to be completed within homeostatic limits. It seems prudent to suggest that the reduction in self-selected work-rate occurs as a result of an integrated feed-back and feed-forward system from a variety of physiological systems. The thermoregulatory system has been implemented in the regulation of pacing strategies during performance tests and it has been proposed that

pacing strategies are altered due to feedback regarding the rate of heat storage occurring (Marino *et al.*, 2004; Marino, 2004). The head, neck and face are regions of high alliesthesial thermosensitivity (Cotter & Taylor, 2005; Shvartz, 1976) and due to the dampening of the perceptual response to exercise it appears that the increased performance and capacity observed as a result of neck-cooling (Chapters 5, 8 and 9) occur due to the manipulation of the feedback which governs the termination of exercise. Below a certain level of thermal strain there appears to be no need for a masking signal and as mentioned earlier, the effectiveness of a cooling intervention is heavily dependent on the magnitude of cooling provided and the level of thermal stain experienced (Nunneley *et al.*, 1971). Time-trial performance was not enhanced in the experimental study presented in Chapter 7; however, this study differed in design to the other time-trial investigations in that the 75 min preload was replaced by a 5 min warm-up and so the participant undertook 70 min less exercise in this study. Due to the difference in the protocol participants commenced the time-trial performance test with rectal temperatures ~1.35°C cooler (Figure 10.1).

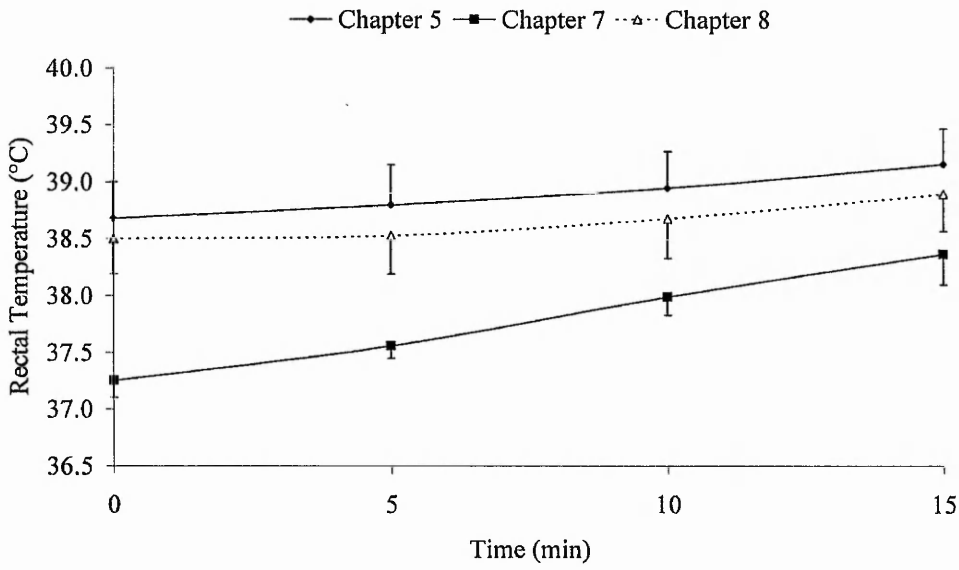

Figure 10.1. The mean (± 1 SD) rectal temperatures observed during the 15 min time-trial phase in the three experimental studies which investigated cooling the neck using this performance test. [data from Chapters 5, 7 and 8]

The data presented in Figure 10.1 shows the lower rectal temperatures experienced during the experimental study presented in Chapter 7 compared to the longer duration trials (Chapter 5 and 8) and it is hypothesised that a performance gain was not observed in this

trial due to the insufficient thermal strain negating the need for the extent of the thermal strain to be masked. Cooling the neck region improves exercise performance and prolongs exercise capacity in a hot environment when the participant is subjected to a sufficient level of thermal strain; however, the data from Chapter 8 suggests that there is a limit to extent to which the body can be deceived. Maintaining the neck at a reduced temperature via the replacement of the collar at 30 min intervals had no additional benefit on exercise performance compared to wearing the collar from the start of exercise alone. The reasons for this are not clear, although if, as proposed by Caputa (2004), numerous mechanisms have evolved to prevent the development of a dangerously high temperature it is unsurprising that there is a limit to the extent to which the mechanisms can be suppressed.

Cooling the neck region dampens the perception of subjective variables and a similar dampening of perceived variables at comparative time-points during exercise has recently been reported in investigations that have pharmacologically elevated cerebral levels of dopamine (Roelands *et al.*, 2008a; Watson *et al.*, 2005c). Both studies demonstrated that the administration of either a dual dopamine/noradrenaline reuptake inhibitor (Watson *et al.*, 2005c) or a specific dopamine reuptake inhibitor (Roelands *et al.*, 2008a) enhanced time-trial performance in the heat by allowing participants to tolerate higher levels of cardiovascular and thermal strain. Peripheral concentrations of dopamine were not influenced by the application of a cooling collar (Chapters 5 and 8) but it is likely that cerebral, rather than peripheral, concentrations of the neurotransmitter that are important (Nybo, 2008). Dopamine is related to feelings of motivation and arousal (Chaouloff, 1989) and therefore has been proposed as a major central component regulating exercise performance in hot environments. There are a number of temperature sensitive dopamine neurons within the human brain (Guatteo *et al.*, 2005). It is possible that the positive effect of the neck-cooling on performance, capacity and the perceptual variables, in the absence of physiological alterations, observed in a hot environment may be due to increased activity of these temperature-sensitive neurons. This topic requires further investigation and is included in the directions for future research.

10.4. Directions for future research

- *Examine the potential role of cerebral dopamine concentrations in the improved ability to perform exercise in a hot environment with the application of a practical neck cooling device.* Recent data has shown that exercise performance in hot conditions is enhanced when cerebral dopamine concentrations are elevated via

pharmacological interventions (Roelands *et al.*, 2008a; Roelands *et al.*, 2008c; Watson *et al.*, 2005c). The brain contains many temperature sensitive dopamine neurons (Guatteo *et al.*, 2005) and therefore it is possible that the improved performance and capacity reported in Chapters 5, 7,8 and 9 may have been due to cooling-induced activation of these temperature sensitive neurons. Conducting a study combining the collar and the pharmacological manipulation of cerebral dopamine concentrations would help to test this theory.

- *Establish the thermal threshold above which practical cooling offers a performance enhancement and the magnitude of cooling that is required to yield this gain.* It has been demonstrated that the extent of the benefit that cooling the head and neck can have is largely dependent upon the difference between the magnitude of the cooling provided and the thermal stress experienced (Nunneley *et al.*, 1971). In the current thesis one collar was investigated after a number of pilot trials but there is scope for further research on the optimal design of the collar (*e.g.* magnitude and duration of cooling provided). In addition, data from Chapters 5, 7 and 8 suggest that there is a threshold of thermal strain that must be breached to benefit from the cooling. Further research is required to verify and establish this threshold to optimise the effectiveness of the cooling intervention.

- *Investigate if cooling the neck region has any direct effect on carotid blood temperature or on the temperature of the human brain during hyperthermia.* Much of the data regarding the effectiveness of cooling the neck on the reduction in brain/carotid blood temperature has been calculated via mathematical modelling approaches (Sukstanskii & Yablonskiy, 2004; Sukstanskii & Yablonskiy, 2007b; Zhu, 2000; Zhu & Diao, 2001). The data suggests that cooling of the carotid blood on route to the brain is possible via an external cooling device; however, direct measurement in either a human or an animal model is required to support or refute this proposal.

- *Examine the effect of the cooling collar post-acclimation. Does the cooling collar offer an additional benefit to performance?* It has been well demonstrated that exercise performance and capacity can be improved in hot environments following the completion of an acclimation programme (*e.g.* Sunderland *et al.*, 2008). Chapters 5 and 8 showed that cooling the neck via the application of a collar can attenuate much of the reduction in time-trial performance observed in Chapter 4.

However, the cooling collar did not fully attenuate the reduction observed. It would be of interest to investigate whether a combined collar and acclimation intervention resulted in a further improvement in exercise performed in a hot environment.

10.5. Practical advice

The research presented within this thesis demonstrates that cooling the neck region via the application of a practical cooling collar can improve both exercise performance and capacity. The neck is a highly thermosensitive region (Cotter & Taylor, 2005) and more efficient in the alleviation of heat strain than cooling the same surface of the body elsewhere (Shvartz, 1976) and the data shows that cooling the neck via practical neck cooling collar offers an athlete, who is training or competing in a hot environment, a performance enhancement. Wearing a collar may not be possible in competition due to the rules and legislation of the particular governing body and so its practical application may be limited to training, warm-up and non-competitive usage. Chapter 9 showed that wearing the collar enables athletes to tolerate a higher core temperature. It has been shown that heat acclimation is accelerated by the obtainment of a high core temperature (Sunderland *et al.*, 2008) so cooling the neck may enhance the adaptations that can be made to a hot environment by allowing athletes to work harder in such conditions. The ability to tolerate a higher core temperature during exercise has an obvious potential for heat injury to occur and so caution is required (see 10.6. Health, safety and ethical considerations)

10.6. Health, safety and ethical considerations

When adopting any intervention that may allow the participant to perform past a natural 'stop-point' it is important that the ethical and heath issues are considered. In sport, the coach, physiologist or trainer has an obligation to ensure the long-term heath and safety of the performers in their care. Cooling the neck has a limited effect on physiological parameters but it can dampen the perceptual feedback of the body and therefore caution is required to ensure that the athlete does not get too hot as a result of the collar masking the true extent of the thermal strain experienced. The capacity data presented in Chapter 9 demonstrated that higher heart rates and rectal temperatures could be tolerated when wearing the collar; however, the participants still terminated exercise below the attainment of a dangerously high core temperature. Highly motivated athletes can achieve and tolerate higher temperatures with sufficient levels of motivation (Pugh *et al.*, 1967) and so they may be at a greater risk of heat illness if using the collar without appropriate monitoring. If

the collar was to be worn by a highly motivated athlete performing in a hot environment careful monitoring should be conducted to ensure that the athlete is not at a greater risk of developing a serious heat illness. The monitoring of core temperature may not be feasible (although ingestible pills are available for field assessment of body temperature) and so close monitoring of heart rate, perceptual responses and fluid balanced should be conducted.

10.7. Conclusion

The data from the experimental Chapters reported in this thesis show that cooling the neck region, via the application of a practical cooling-collar, improves time-trial performance and extends exercise capacity during running in a hot environment, when the body is under a sufficient level of thermal strain. Enhanced performance and capacity do not occur as a result of alterations in physiological or peripheral neuroendocrinological variables and appears to occur due to a dampening of the perceived levels of exertion and thermal strain. This dampening offers a false signal regarding the physiological state of the body and allows a faster pace to be selected and a higher level of physiological strain to be tolerated, although there is a limit to the extent to which the physiological strain can be masked.

References

Abraha HD, Butterworth RJ, Bath PM, Wassif WS, Garthwaite J, & Sherwood RA (1997). Serum S-100 protein, relationship to clinical outcome in acute stroke. *Ann Clin Biochem*, **34 (Pt 5)**, 546-550.

Adams H, Adams R, Del ZG, & Goldstein LB (2005). Guidelines for the early management of patients with ischemic stroke: 2005 guidelines update a scientific statement from the Stroke Council of the American Heart Association/American Stroke Association. *Stroke*, **36**, 916-923.

Alberty M, Sidney M, Huot-Marchand F, Dekerle J, Bosquet L, Gorce P, & Lensel G (2006). Reproducibility of performance in three types of training test in swimming. *Int J Sports Med*, **27**, 623-628.

Ali MS, Harmer M, & Vaughan R (2000). Serum S100 protein as a marker of cerebral damage during cardiac surgery. *Br J Anaesth*, **85**, 287-298.

Altareki A, Drust B, Cable NT, Atkinson G, & Gregson WA (2006). The effects of an environmental heat stress (35°C) on simulated 4 km cycling time-trial performance. *Med Sci Sports Exerc*, **38**, S354.

Altman DI, Powers WJ, Perlman JM, Herscovitch P, Volpe SL, & Volpe JJ (1988). Cerebral blood flow requirement for brain viability in newborn infants is lower than in adults. *Ann Neurol*, **24**, 218-226.

Anderson RE, Hansson LO, Nilsson O, jlai-Merzoug R, & Settergren G (2001). High serum S100B levels for trauma patients without head injuries. *Neurosurgery*, **48**, 1255-1258.

Ansley L, Marvin G, Sharma A, Kendall MJ, Jones DA, & Bridge MV (2007). The effects of head cooling on endurance and neuroendocrine responses to exercise in warm conditions. *Physiol Res* Epub ahead of print.

Armada-da-Silva PA, Woods J, & Jones DA (2004). The effect of passive heating and face cooling on perceived exertion during exercise in the heat. *Eur J Appl Physiol*, **91**, 563-571.

Arngrimsson SA, Petitt DS, Stueck MG, Jorgensen DK, & Cureton KJ (2004). Cooling vest worn during active warm-up improves 5-km run performance in the heat. *J Appl Physiol*, **96**, 1867-1874.

Astrand P-O, Rodahl K, Dahl HA, & Stromme SB (2003). *Textbook of work physiology*, 4 ed. Human Kinetics, Champaign.

Atkinson G & Nevill AM (1998). Statistical methods for assessing measurement error (reliability) in variables relevant to sports medicine. *Sports Med*, **26**, 217-238.

Atkinson G & Nevill AM (2001). Selected issues in the design and analysis of sport performance research. *J Sports Sci*, **19**, 811-827.

Bailey SP, Davis JM, & Ahlborn EN (1993). Neuroendocrine and substrate responses to altered brain 5-HT activity during prolonged exercise to fatigue. *J Appl Physiol*, **74**, 3006-3012.

Baker MA & Hayward JN (1968). Intracranial heat exchange and regulation of brain temperature in sheep. *Life Sci*, **7**, 349-357.

Baker MA (1972). Influence of the carotid rete on brain temperature in cats exposed to hot environments. *J Physiol*, **220**, 711-728.

Baker MA, Chapman LW, & Nathanson M (1974). Control of brain temperature in dogs: effects of tracheostomy. *Respir Physiol*, **22**, 325-333.

Baker MA (1979). A brain-cooling system in mammals. *Sci Am*, **240**, 130-139.

Baker MA (1982). Brain cooling in endotherms in heat and exercise. *Annu Rev Physiol*, **44**, 85-96.

Baker MA & Nijland MJ (1993). Selective brain cooling in goats: effects of exercise and dehydration. *J Physiol*, **471**, 679-692.

Barnett A & Maughan RJ (1993). Response of unacclimatized males to repeated weekly bouts of exercise in the heat. *Br J Sports Med*, **27**, 39-44.

Barone FC, Feuerstein GZ, & White RF (1997). Brain cooling during transient focal ischemia provides complete neuroprotection. *Neurosci Biobehav Rev*, **21**, 31-44.

Bassett DR, Jr., Nagle FJ, Mookerjee S, Darr KC, Ng AV, Voss SG, & Napp JP (1987). Thermoregulatory responses to skin wetting during prolonged treadmill running. *Med Sci Sports Exerc*, **19**, 28-32.

Belehradek J (1957). Physiological aspects of heat and cold. *Annu Rev Physiol*, **19**, 59-82.

Benzinger TH (1969). Clinical temperature. New physiological basis. *JAMA*, **209**, 1200-1206.

Bernstein MH, Curtis MB, & Hudson DM (1979). Independence of brain and body temperatures in flying American kestrels, Falco sparverius. *Am J Physiol*, **237**, R58-R62.

Billat V, Renoux JC, Pinoteau J, Petit B, & Koralsztein JP (1994). Reproducibility of running time to exhaustion at VO2max in subelite runners. *Med Sci Sports Exerc*, **26**, 254-257.

Bishop D (1997). Reliability of a 1-h endurance performance test in trained female cyclists. *Med Sci Sports Exerc*, **29**, 554-559.

Bland JM & Altman DG (1999). Measuring agreement in method comparison studies. *Stat Methods Med Res*, **8**, 135-160.

Blomstrand E, Perrett D, Parry-Billings M, & Newsholme EA (1989). Effect of sustained exercise on plasma amino acid concentrations and on 5-hydroxytryptamine metabolism in six different brain regions in the rat. *Acta Physiol Scand*, **136**, 473-481.

Blomstrand E (2001). Amino acids and central fatigue. *Amino Acids*, **20**, 25-34.

Bolster DR, Trappe SW, Short KR, Scheffield-Moore M, Parcell AC, Schulze KM, & Costill DL (1999). Effects of precooling on thermoregulation during subsequent exercise. *Med Sci Sports Exerc*, **31**, 251-257.

Booth J, Marino FE, & Ward JJ (1997). Improved running performance in hot humid conditions following whole body precooling. *Med Sci Sports Exerc*, **29**, 943-949.

Booth J, Wilsmore BR, Macdonald AD, Zeyl A, Storlien LH, & Taylor NA (2004). Intramuscular temperatures during exercise in the heat following pre-cooling and pre-heating. *J Therm Biol*, **29**, 709-715.

Borg G (1982). Psychophysical bases of perceived exertion. *Med Sci Sports Exerc*, **14**, 377-382.

Brengelmann GL (1993). Specialized brain cooling in humans? *FASEB J*, **7**, 1148-1152.

Bridge MW, Weller AS, Rayson M, & Jones DA (2003a). Ambient temperature and the pituitary hormone responses to exercise in humans. *Exp Physiol*, **88**, 627-635.

Bridge MW, Weller AS, Rayson M, & Jones DA (2003b). Responses to exercise in the heat related to measures of hypothalamic serotonergic and dopaminergic function. *Eur J Appl Physiol*, **89**, 451-459.

Brisson GR, Audet A, Ledoux M, Matton P, Pellerin-Massicotte J, & Peronnet F (1986a). Exercise-induced blood prolactin variations in trained adult males: a thermic stress more than an osmotic stress. *Horm Res*, **23**, 200-206.

Brisson GR, Peronnet F, Ledoux M, Pellerin-Massicotte J, Matton P, Garceau F, & Boisvert P, Jr. (1986b). Temperature-induced hyperprolactinemia during exercise. *Horm Metab Res*, **18**, 283-284.

Brisson GR, Bouchard J, Peronnet F, Boisvert P, & Garceau F (1987). Evidence for an interference of selective face ventilation on hyperprolactinemia induced by hyperthermic treadmill running. *Int J Sports Med*, **8**, 387-391.

Brisson GR, Boisvert P, Peronnet F, Quirion A, & Senecal L (1989). Face cooling-induced reduction of plasma prolactin response to exercise as part of an integrated response to thermal stress. *Eur J Appl Physiol Occup Physiol*, **58**, 816-820.

Brisson GR, Peronnet F, Perrault H, Boisvert P, Massicotte D, & Gareau R (1991). Prolactinotrophic effect of endogenous and exogenous heat loads in human male adults. *J Appl Physiol*, **70**, 1351-1355.

Brown GA & Williams GM (1982). The effect of head cooling on deep body temperature and thermal comfort in man. *Aviat Space Environ Med*, **53**, 583-586.

Bulbulian R, Shapiro R, Murphy M, & Levenhagen D (1999). Effectiveness of a commercial head-neck cooling device. *J Strength Cond Res*, **13**, 198-205.

Butterworth RJ, Sherwood RA, & Bath PM (1998). Serum S-100 protein in acute stroke. *Stroke*, **29**, 730.

Buttner T, Weyers S, Postert T, Sprengelmeyer R, & Kuhn W (1997). S-100 protein: serum marker of focal brain damage after ischemic territorial MCA infarction. *Stroke*, **28**, 1961-1965.

Cabanac M & Caputa M (1979). Natural selective cooling of the human brain: evidence of its occurrence and magnitude. *J Physiol*, **286**, 255-264.

Cabanac M & Brinnel H (1985). Blood flow in the emissary veins of the human head during hyperthermia. *Eur J Appl Physiol Occup Physiol*, **54**, 172-176.

Cabanac M (1986). Keeping a cool head. *News Physiol Sci*, **1**, 41-44.

Cabanac M (1993). Selective brain cooling in humans: "fancy" or fact? *FASEB J*, **7**, 1143-1146.

Cabanac M (1998). Selective brain cooling and thermoregulatory set-point. *J Basic Clin Physiol Pharmacol*, **9**, 3-13.

Caputa M, Feistkorn G, & Jessen C (1986). Effects of brain and trunk temperatures on exercise performance in goats. *Pflugers Arch*, **406**, 184-189.

Caputa M (2004). Selective brain cooling: a multiple regulatory mechanism. *J Therm Biol*, **29**, 691-702.

Carithers RW & Seagrave RC (1976). Canine hyperthermia with cerebral protection. *J Appl Physiol*, **40**, 543-548.

Castle PC, Macdonald AL, Philp A, Webborn A, Watt PW, & Maxwell NS (2006). Precooling leg muscle improves intermittent sprint exercise performance in hot, humid conditions. *J Appl Physiol*, **100**, 1377-1384.

Chandler JV & Blair SN (1980). The effect of amphetamines on selected physiological components related to athletic success. *Med Sci Sports Exerc*, **12**, 65-69.

Chaouloff F, Elghozi JL, Guezennec Y, & Laude D (1985). Effects of conditioned running on plasma, liver and brain tryptophan and on brain 5-hydroxytryptamine metabolism of the rat. *Br J Pharmacol*, **86**, 33-41.

Chaouloff F, Kennett GA, Serrurrier B, Merino D, & Curzon G (1986). Amino acid analysis demonstrates that increased plasma free tryptophan causes the increase of brain tryptophan during exercise in the rat. *J Neurochem*, **46**, 1647-1650.

Chaouloff F, Laude D, Merino D, Serrurrier B, Guezennec Y, & Elghozi JL (1987). Amphetamine and alpha-methyl-p-tyrosine affect the exercise-induced imbalance between the availability of tryptophan and synthesis of serotonin in the brain of the rat. *Neuropharmacology*, **26**, 1099-1106.

Chaouloff F (1989). Physical exercise and brain monoamines: a review. *Acta Physiol Scand*, **137**, 1-13.

Cheung SS & McLellan TM (1998). Heat acclimation, aerobic fitness, and hydration effects on tolerance during uncompensable heat stress. *J Appl Physiol*, **84**, 1731-1739.

Cheung SS & Sleivert GG (2004a). Lowering of skin temperature decreases isokinetic maximal force production independent of core temperature. *Eur J Appl Physiol*, **91**, 723-728.

Cheung SS & Sleivert GG (2004b). Multiple triggers for hyperthermic fatigue and exhaustion. *Exerc Sport Sci Rev*, **32**, 100-106.

Cheung SS & Robinson AM (2004). The influence of upper-body pre-cooling on repeated sprint performance in moderate ambient temperatures. *J Sports Sci*, **22**, 605-612.

Cheung SS (2007). Hyperthermia and voluntary exhaustion: integrating models and future challenges. *Appl Physiol Nutr Metab*, **32**, 808-817.

Cheuvront SN, Chinevere TD, Ely BR, Kenefick RW, Goodman DA, McClung JP, & Sawka MN (2008). Serum S-100beta Response to Exercise-Heat Strain before and after Acclimation. *Med Sci Sports Exerc*, **40**, 1477-1482.

Cohen J (1988). *Statistical power analysis for the behavioral sciences*, 2 ed. Lawrence Erlbaum, New Jersey.

Corbett RJ, Laptook AR, Tollefsbol G, & Kim B (1995). Validation of a noninvasive method to measure brain temperature in vivo using 1H NMR spectroscopy. *J Neurochem*, **64**, 1224-1230.

Cotter JD & Taylor NA (2005). The distribution of cutaneous sudomotor and alliesthesial thermosensitivity in mildly heat-stressed humans: an open-loop approach. *J Physiol*, **565**, 335-345.

Crewe H, Tucker R, & Noakes TD (2008). The rate of increase in rating of perceived exertion predicts the duration of exercise to fatigue at a fixed power output in different environmental conditions. *Eur J Appl Physiol*, **103**, 569-577.

Cunningham RT, Watt M, Winder J, McKinstry S, Lawson JT, Johnston CF, Hawkins SA, & Buchanan KD (1996). Serum neurone-specific enolase as an indicator of stroke volume. *Eur J Clin Invest*, **26**, 298-303.

Currell K, Jentjens RL, & Jeukendrup AE (2006). Reliability of a cycling time trial in a glycogen-depleted state. *Eur J Appl Physiol*, **98**, 583-589.

Curzon G, Friedel J, Katamaneni BD, Greenwood MH, & Lader MH (1974). Unesterified fatty acids and the binding of tryptophan in human plasma. *Clin Sci Mol Med*, **47**, 415-424.

Daniel PM (1966). The blood supply of the hypothalamus and pituitary gland. *Br Med Bull*, **22**, 202-208.

Davis JM, Bailey SP, Jackson DA, Stansner AB, & Morehouse SL (1993). Effects of a serotonin (5-HT) agonist during prolonged exercise to fatigue in humans. *Med Sci Sports Exerc*, **25**, S78.

Davis JM & Bailey SP (1997). Possible mechanisms of central nervous system fatigue during exercise. *Med Sci Sports Exerc*, **29**, 45-57.

Desruelle AV & Candas V (2000). Thermoregulatory effects of three different types of head cooling in humans during a mild hyperthermia. *Eur J Appl Physiol*, **81**, 33-39.

Dethy S, Laute MA, Luxen A, Hildebrand J, & Goldman S (1995). Effect of pergolide on endogenous and exogenous L-DOPA metabolism in the rat striatum: a microdialysis study. *J Neural Transm Gen Sect*, **101**, 1-11.

Dietrich MO, Tort AB, Schaf DV, Farina M, Goncalves CA, Souza DO, & Portela LV (2003). Increase in serum S100B protein level after a swimming race. *Can J Appl Physiol*, **28**, 710-716.

Dill DB & Costill DL (1974). Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. *J Appl Physiol*, **37**, 247-248.

Doyle JA & Martinez AL (1998). Reliability of a protocol for testing endurance performance in runners and cyclists. *Res Q Exerc Sport*, **69**, 304-307.

Drust B, Cable NT, & Reilly T (2000). Investigation of the effects of the pre-cooling on the physiological responses to soccer-specific intermittent exercise. *Eur J Appl Physiol*, **81**, 11-17.

Drust B, Rasmussen P, Mohr M, Nielsen B, & Nybo L (2005). Elevations in core and muscle temperature impairs repeated sprint performance. *Acta Physiol Scand*, **183**, 181-190.

Duffield R, Dawson B, Bishop D, Fitzsimons M, & Lawrence S (2003). Effect of wearing an ice cooling jacket on repeat sprint performance in warm/humid conditions. *Br J Sports Med*, **37**, 164-169.

Duffield R & Marino FE (2007). Effects of pre-cooling procedures on intermittent-sprint exercise performance in warm conditions. *Eur J Appl Physiol*, **100**, 727-735.

Febbraio MA, Snow RJ, Stathis CG, Hargreaves M, & Carey MF (1994). Effect of heat stress on muscle energy metabolism during exercise. *J Appl Physiol*, **77**, 2827-2831.

Fernstrom JD (1983). Role of precursor availability in control of monoamine biosynthesis in brain. *Physiol Rev*, **63**, 484-546.

Fernstrom JD & Fernstrom MH (2006). Exercise, serum free tryptophan, and central fatigue. *J Nutr*, **136**, 553S-559S.

Fernstrom MH & Fernstrom JD (1993). Large changes in serum free tryptophan levels do not alter brain tryptophan levels: studies in streptozotocin-diabetic rats. *Life Sci*, **52**, 907-916.

Freeman ME, Kanyicska B, Lerant A, & Nagy G (2000). Prolactin: structure, function, and regulation of secretion. *Physiol Rev*, **80**, 1523-1631.

Frewin DB, Frantz AG, & Downey JA (1976). The effect of ambient temperature on the growth hormone and prolactin response to exercise. *Aust J Exp Biol Med Sci*, **54**, 97-101.

Fruth JM & Gisolfi CV (1983). Work-heat tolerance in endurance-trained rats. *J Appl Physiol*, **54**, 249-253.

Fuller A, Carter RN, & Mitchell D (1998). Brain and abdominal temperatures at fatigue in rats exercising in the heat. *J Appl Physiol*, **84**, 877-883.

Galloway SDR & Maughan RJ (1997). Effects of ambient temperature on the capacity to perform prolonged exercise in man. *Med Sci Sports Exerc*, **29**, 1240-1249.

Gandevia SC (2001). Spinal and supraspinal factors in human muscle fatigue. *Physiol Rev*, **81**, 1725-1789.

Gerald MC (1978). Effects of (+)-amphetamine on the treadmill endurance performance of rats. *Neuropharmacology*, **17**, 703-704.

Gonzalez RR, Berglund LG, & Gagge AP (1978). Indices of thermoregulatory strain for moderate exercise in the heat. *J Appl Physiol*, **44**, 889-899.

Gonzalez-Alonso J, Teller C, Andersen SL, Jensen FB, Hyldig T, & Nielsen B (1999). Influence of body temperature on the development of fatigue during prolonged exercise in the heat. *J Appl Physiol*, **86**, 1032-1039.

Gonzalez-Alonso J & Calbet JA (2003). Reductions in systemic and skeletal muscle blood flow and oxygen delivery limit maximal aerobic capacity in humans. *Circulation*, **107**, 824-830.

Gordon NF, Bogdanffy GM, & Wilkinson J (1990). Effect of a practical neck cooling device on core temperature during exercise. *Med Sci Sports Exerc*, **22**, 245-249.

Graham KS & McLellan TM (1989). Variability of time to exhaustion and oxygen deficit in supramaximal exercise. *Australian Journal of Science and Medicine in Sport*, **21**, 11-14.

Gray H (1918). *Anatomy of the human body*, 20 ed. Lea & Febiger, Philadelphia.

Greenleaf JE, Van BW, Brock PJ, Montgomery LD, Morse JT, Shvartz E, & Kravik S (1980). Fluid-electrolyte shifts and thermoregulation: Rest and work in heat with head cooling. *Aviat Space Environ Med*, **51**, 747-753.

Guatteo E, Chung KK, Bowala TK, Bernardi G, Mercuri NB, & Lipski J (2005). Temperature sensitivity of dopaminergic neurons of the substantia nigra pars compacta: involvement of transient receptor potential channels. *J Neurophysiol*, **94**, 3069-3080.

Hamada S, Torii M, Szygula Z, & Adachi K (2006). Effect of partial body cooling on thermophysiological responses during cycling work in a hot environment. *J Therm Biol*, **31**, 194-207.

Hammel HT, Jackson D, Stolwijk JA, Hardy JD, & Stromme SB (1963). Temperature regulation by hypothalamic proportional control with an adjustable set point. *J Appl Physiol*, **18**, 1146-1154.

Hasegawa H, Yazawa T, Yasumatsu M, Otokawa M, & Aihara Y (2000). Alteration in dopamine metabolism in the thermoregulatory center of exercising rats. *Neurosci Lett*, **289**, 161-164.

Hasegawa H, Takatori T, Komura T, & Yamasaki M (2005). Wearing a cooling jacket during exercise reduces thermal strain and improves endurance exercise performance in a warm environment. *J Strength Cond Res*, **19**, 122-128.

Hasegawa H, Takatori T, Komura T, & Yamasaki M (2006). Combined effects of pre-cooling and water ingestion on thermoregulation and physical capacity during exercise in a hot environment. *J Sports Sci*, **24**, 3-9.

Hasselblatt M, Mooren FC, von AN, Keyvani K, Fromme A, Schwarze-Eicker K, Senner V, & Paulus W (2004). Serum S100beta increases in marathon runners reflect extracranial release rather than glial damage. *Neurology*, **62**, 1634-1636.

Haymaker W (1969). Blood supply of the human hypothalamus. In *The Hypothalamus*, eds. Haymaker W, Anderson E, & Nauta WJH, pp. 210-218. Charles C. Thomas, Springfield.

Hayward JN (1968). Brain temperature regulation during sleep and arousal in the dog. *Exp Neurol*, **21**, 201-212.

Hayward JN & Baker MA (1969). A comparative study of the role of the cerebral arterial blood in the regulation of brain temperature in five mammals. *Brain Res*, **16**, 417-440.

Hessemer V, Langusch D, Bruck LK, Bodeker RH, & Breidenbach T (1984). Effect of slightly lowered body temperatures on endurance performance in humans. *J Appl Physiol*, **57**, 1731-1737.

Hickey MS, Costill DL, McConell GK, Widrick JJ, & Tanaka H (1992). Day to day variation in time trial cycling performance. *Int J Sports Med*, **13**, 467-470.

Hopkins WG, Hawley JA, & Burke LM (1999). Design and analysis of research on sport performance enhancement. *Med Sci Sports Exerc*, **31**, 472-485.

Hopkins WG (2000). Measures of reliability in sports medicine and science. *Sports Med*, **30**, 1-15.

Hopkins WG, Schabort EJ, & Hawley JA (2001). Reliability of power in physical performance tests. *Sports Med*, **31**, 211-234.

Horn M, Schlote W, & Henrich HA (1991). Global cerebral ischemia and subsequent selective hypothermia. A neuropathological and morphometrical study on ischemic neuronal damage in cat. *Acta Neuropathol*, **81**, 443-449.

Hornery DJ, Papalia S, Mujika I, & Hahn A (2005). Physiological and performance benefits of halftime cooling. *J Sci Med Sport*, **8**, 15-25.

Hunter I, Tegeder AR, & Martini E (2006). Core body temperature during cross country racing with the Nike ice-vest. *Med Sci Sports Exerc*, **38**, S58.

Ishiwata T, Saito T, Hasegawa H, Yazawa T, Otokawa M, & Aihara Y (2004). Changes of body temperature and extracellular serotonin level in the preoptic area and anterior hypothalamus after thermal or serotonergic pharmacological stimulation of freely moving rats. *Life Sci*, **75**, 2665-2675.

Isobe T, Ishioka N, Masuda T, Takahashi Y, Ganno S, & Okuyama T (1983). A rapid separation of S100 subunits by high performance liquid chromatography: the subunit compositions of S100 proteins. *Biochem Int*, **6**, 419-426.

IUPS Thermal Commission (1987). Glossary of terms for thermal physiology. Second edition. Revised by The Commission for Thermal Physiology of the International Union of Physiological Sciences (IUPS Thermal Commission). *Pflugers Arch*, **410**, 567-587.

Jackson A, Der WK, Schick R, & Sanchez R (1990). An analysis of the validity of the three-mile run as a field test of aerobic capacity in college males. *Res Q Exerc Sport*, **61**, 233-237.

Jensen R, Marshak DR, Anderson C, Lukas TJ, & Watterson DM (1985). Characterization of human brain S100 protein fraction: amino acid sequence of S100 beta. *J Neurochem*, **45**, 700-705.

Jeukendrup AE, Saris WHM, Bronus F, & Arnold DM (1996). A new validated endurance performance test. *Med Sci Sports Exerc*, **28**, 266-270.

Johnsen HK, Blix AS, Merker J, & Bolz KD (1987). Selective cooling of the brain in reindeer. *Am J Physiol*, **253**, 848-853.

Jones AM & Doust J (1996). A comparison of three protocols for the determination of maximal aerobic power in runners. *J Sports Sci*, **14**, S89.

Judelson DA, Maresh CM, Yamamoto LM, Farrell MJ, Armstrong LE, Kraemer WJ, Volek JS, Spiering BA, Casa DJ, & Anderson JM (2008). Effect of hydration state on resistance exercise-induced endocrine markers of anabolism, catabolism, and metabolism. *J Appl Physiol*, **105**, 816-824.

Kapural M, Krizanac-Bengez L, Barnett G, Perl J, Masaryk T, Apollo D, Rasmussen P, Mayberg MR, & Janigro D (2002). Serum S-100beta as a possible marker of blood-brain barrier disruption. *Brain Res*, **940**, 102-104.

Kay D, Taaffe DR, & Marino FE (1999). Whole-body pre-cooling and heat storage during self-paced cycling performance in warm humid conditions. *J Sports Sci*, **17**, 937-944.

Kayser B (2003). Exercise starts and ends in the brain. *Eur J Appl Physiol*, **90**, 411-419.

Kissen AT, Hall JF, Jr., & Klemm FK (1971). Physiological responses to cooling the head and neck versus the trunk and leg areas in severe hyperthermic exposure. *Aerosp Med*, **42**, 882-888.

Kiyatkin EA (2007). Brain temperature fluctuations during physiological and pathological conditions. *Eur J Appl Physiol*, **101**, 3-17.

Kotchen TA, Hartley LH, Rice TW, Mougey EH, Jones LG, & Mason JW (1971). Renin, norepinephrine, and epinephrine responses to graded exercise. *J Appl Physiol*, **31**, 178-184.

Kratzing CC & Cross RB (1984). Effects of facial cooling during exercise at high temperature. *Eur J Appl Physiol Occup Physiol*, **53**, 118-120.

Krebs PS & Powers SK (1989). Reliability of endurance tests. *Med Sci Sports Exerc*, **21**, S10.

Lambert EV, St Clair GA, & Noakes TD (2005). Complex systems model of fatigue: integrative homoeostatic control of peripheral physiological systems during exercise in humans. *Br J Sports Med*, **39**, 52-62.

Laursen P, Francis G, Abbiss C, & Newton M (2006). Comparison of the reliability of open- versus closed-loop treadmill running tests in well-trained runners. *Proceedings from the 11th Annual Congress of the European College of Sport Science* 214.

Lee DT & Haymes EM (1995). Exercise duration and thermoregulatory responses after whole body precooling. *J Appl Physiol*, **79**, 1971-1976.

Linnane DM, Bracken RM, Brooks S, Cox VM, & Ball D (2004). Effects of hyperthermia on the metabolic responses to repeated high-intensity exercise. *Eur J Appl Physiol*, **93**, 159-166.

Lipton JM & Clark WG (1986). Neurotransmitters in temperature control. *Annu Rev Physiol*, **48**, 613-623.

Lopez RM, Cleary MA, Jones LC, & Zuri RE (2008). Thermoregulatory influence of a cooling vest on hyperthermic athletes. *J Athl Train*, **43**, 55-61.

Low D, Cable T, & Purvis A (2005). Exercise thermoregulation and hyperprolactinaemia. *Ergonomics*, **48**, 1547-1557.

MacDougall JD, Reddan WG, Layton CR, & Dempsey JA (1974). Effects of metabolic hyperthermia on performance during heavy prolonged exercise. *J Appl Physiol*, **36**, 538-544.

Marchi N, Rasmussen P, Kapural M, Fazio V, Kight K, Mayberg MR, Kanner A, Ayumar B, Albensi B, Cavaglia M, & Janigro D (2003). Peripheral markers of brain damage and blood-brain barrier dysfunction. *Restor Neurol Neurosci*, **21**, 109-121.

Marino FE & Booth J (1998). Whole body cooling by immersion in water at moderate temperatures. *J Sci Med Sport*, **1**, 72-81.

Marino FE (2002). Methods, advantages, and limitations of body cooling for exercise performance. *Br J Sports Med*, **36**, 89-94.

Marino FE, Kay D, Cannon J, Serwach N, & Hilder M (2002). A reproducible and variable intensity cycling performance protocol for warm conditions. *J Sci Med Sport*, **5**, 95-107.

Marino FE (2004). Anticipatory regulation and avoidance of catastrophe during exercise-induced hyperthermia. *Comp Biochem Physiol A Mol Integr Physiol*, **139**, 561-569.

Marino FE, Lambert MI, & Noakes TD (2004). Superior performance of African runners in warm humid but not in cool environmental conditions. *J Appl Physiol*, **96**, 124-130.

Marsh D & Sleivert G (1999). Effect of precooling on high intensity cycling performance. *Br J Sports Med*, **33**, 393-397.

Martin DE & Gynn RWH (2000). *The Olympic marathon* Human Kinetics, Champaign, IL.

Martin DT, Hahn A, Ryan-Tanner R, Yates K, Lee H, & Smith JA (1998). Ice jackets are cool. *Sport Science*, **2**, sportsci.org/jour/9804/dtm.html.

McCaffrey TV, Geis GS, Chung JM, & Wurster RD (1975). Effect of isolated head heating and cooling on sweating in man. *Aviat Space Environ Med*, **46**, 1353-1357.

McConaghy FF, Hales JR, Rose RJ, & Hodgson DR (1995). Selective brain cooling in the horse during exercise and environmental heat stress. *J Appl Physiol*, **79**, 1849-1854.

McLellan TM, Cheung SS, & Jacobs I (1995). Variability of time to exhaustion during submaximal exercise. *Can J Appl Physiol*, **20**, 39-51.

McMenamy RH, Lund CC, & Oncley J (1957). Unbound amino acid concentrations in human blood plasmas. *J Clin Invest*, **36**, 1672-1679.

Meeusen R, Roeykens J, Magnus L, Keizer H, & De MK (1997). Endurance performance in humans: the effect of a dopamine precursor or a specific serotonin (5-HT2A/2C) antagonist. *Int J Sports Med*, **18**, 571-577.

Meeusen R, Watson P, Hasegawa H, Roelands B, & Piacentini MF (2006). Central fatigue: the serotonin hypothesis and beyond. *Sports Med*, **36**, 881-909.

Millan N, Murdock LL, Bleier R, & Siegel FL (1979). Effects of acute hyperthermia on polyribosomes, in vivo protein synthesis and ornithine decarboxylase activity in the neonatal rat brain. *J Neurochem*, **32**, 311-317.

Mills DE & Robertshaw D (1981). Response of plasma prolactin to changes in ambient temperature and humidity in man. *J Clin Endocrinol Metab*, **52**, 279-283.

Mitchell JB, McFarlin BK, & Dugas JP (2003). The effect of pre-exercise cooling on high intensity running performance in the heat. *Int J Sports Med*, **24**, 118-124.

Morrison S, Sleivert GG, & Cheung SS (2004). Passive hyperthermia reduces voluntary activation and isometric force production. *Eur J Appl Physiol*, **91**, 729-736.

Mundel T, Hooper P, Bunn S, & Jones D (2005). The effects of face-cooling on the perception of exertion and neuroendocrine response to hyperthermic exercise. *J Appl Physiol*, **565P**, C31.

Mundel T, Hooper PL, Bunn SJ, & Jones DA (2006). The effects of face cooling on the prolactin response and subjective comfort during moderate passive heating in humans. *Exp Physiol*, **91**, 1007-1014.

Mundel T, Bunn SJ, Hooper PL, & Jones DA (2007). The effects of face cooling during hyperthermic exercise in man: evidence for an integrated thermal, neuroendocrine and behavioural response. *Exp Physiol*, **92**, 187-195.

Mustafa S & Thulesius O (2002). Cooling-induced carotid artery dilation: an experimental study in isolated vessels. *Stroke*, **33**, 256-260.

Nadel ER & Stolwijk JA (1971). Physiologic control of rate of local sweat secretion in man. *J Physiol (Paris)*, **63**, 353-355.

Nadel ER, Bullard RW, & Stolwijk JA (1971a). Importance of skin temperature in the regulation of sweating. *J Appl Physiol*, **31**, 80-87.

Nadel ER, Mitchell JW, Saltin B, & Stolwijk JA (1971b). Peripheral modifications to the central drive for sweating. *J Appl Physiol*, **31**, 828-833.

Newsholme EA & Blomstrand E (1995). Tryptophan, 5-hydroxytryptamine and a possible explanation for central fatigue. *Adv Exp Med Biol*, **384**, 315-320.

Newsholme EA & Blomstrand E (2006). Branched-chain amino acids and central fatigue. *J Nutr*, **136**, 274S-276S.

Nielsen B, Hales JR, Strange S, Christensen NJ, Warberg J, & Saltin B (1993). Human circulatory and thermoregulatory adaptations with heat acclimation and exercise in a hot, dry environment. *J Physiol*, **460**, 467-485.

Nielsen B, Hyldig T, Bidstrup F, Gonzalez-Alonso J, & Christoffersen GR (2001). Brain activity and fatigue during prolonged exercise in the heat. *Pflugers Arch*, **442**, 41-48.

Noakes TD & St Clair GA (2004). Logical limitations to the "catastrophe" models of fatigue during exercise in humans. *Br J Sports Med*, **38**, 648-649.

Nunneley SA, Troutman SJ, Jr., & Webb P (1971). Head cooling in work and heat stress. *Aerosp Med*, **42**, 64-68.

Nunneley SA & Nelson DA (1994). Limitations on arteriovenous cooling of the blood supply to the human brain. *Eur J Appl Physiol Occup Physiol*, **69**, 474-479.

Nybo L & Nielsen B (2001a). Hyperthermia and central fatigue during prolonged exercise in humans. *J Appl Physiol*, **91**, 1055-1060.

Nybo L & Nielsen B (2001b). Perceived exertion is associated with an altered brain activity during exercise with progressive hyperthermia. *J Appl Physiol*, **91**, 2017-2023.

Nybo L & Nielsen B (2001c). Perceived exertion is associated with an altered brain activity during exercise with progressive hyperthermia. *J Appl Physiol*, **91**, 2017-2023.

Nybo L & Nielsen B (2001d). Middle cerebral artery blood velocity is reduced with hyperthermia during prolonged exercise in humans. *J Physiol*, **534**, 279-286.

Nybo L, Secher NH, & Nielsen B (2002a). Inadequate heat release from the human brain during prolonged exercise with hyperthermia. *J Physiol*, **545**, 697-704.

Nybo L, Moller K, Volianitis S, Nielsen B, & Secher NH (2002b). Effects of hyperthermia on cerebral blood flow and metabolism during prolonged exercise in humans. *J Appl Physiol*, **93**, 58-64.

Nybo L (2007). Exercise and heat stress: cerebral challenges and consequences. *Prog Brain Res*, **162**, 29-43.

Nybo L (2008). Hyperthermia and fatigue. *J Appl Physiol*, **104**, 871-878.

Olschewski H & Bruck K (1988). Thermoregulatory, cardiovascular, and muscular factors related to exercise after precooling. *J Appl Physiol*, **64**, 803-811.

Otto M, Holthusen S, Bahn E, Sohnchen N, Wiltfang J, Geese R, Fischer A, & Reimers CD (2000). Boxing and running lead to a rise in serum levels of S-100B protein. *Int J Sports Med*, **21**, 551-555.

Palmer CD, Sleivert G, & Cotter JD (2001). The effects of head and neck cooling on thermoregulation, pace selection and performance. *Proceedings from the Australian Physiological and Pharmacological Society*, **32**, 122P.

Palmer GS, Dennis SC, Noakes TD, & Hawley JA (1996). Assessment of the reproducibility of performance testing on an air-braked cycle ergometer. *Int J Sports Med*, **17**, 293-298.

Pandolf KB, Cadarette BS, Sawka MN, Young AJ, Francesconi RP, & Gonzalez RR (1988). Thermoregulatory responses of middle-aged and young men during dry-heat acclimation. *J Appl Physiol*, **65**, 65-71.

Pannier JL, Bouckaert JJ, & Lefebvre RA (1995). The antiserotonin agent pizotifen does not increase endurance performance in humans. *Eur J Appl Physiol Occup Physiol*, **72**, 175-178.

Pugh LG, Corbett JL, & Johnson RH (1967). Rectal temperatures, weight losses, and sweat rates in marathon running. *J Appl Physiol*, **23**, 347-352.

Quirion A, Boisvert P, Brisson GR, DeCarufel D, Laurencelle L, Dulac S, Vogelaere P, & Therminarias A (1989). Effects of selective cooling of the facial area on physiological and metabolic output during graded maximal or prolonged submaximal exercise. *Int J Biometeorol*, **33**, 82-84.

Quod MJ, Martin DT, & Laursen PB (2006). Cooling athletes before competition in the heat: comparison of techniques and practical considerations. *Sports Med*, **36**, 671-682.

Radomski MW, Cross M, & Buguet A (1998). Exercise-induced hyperthermia and hormonal responses to exercise. *Can J Physiol Pharmacol*, **76**, 547-552.

Ramanathan NL (1964). A new weighting system for mean surface temperature of the human body. *J Appl Physiol*, **19**, 531-533.

Rasch W, Samson P, Cote J, & Cabanac M (1991). Heat loss from the human head during exercise. *J Appl Physiol*, **71**, 590-595.

Reynolds MA, Kirchick HJ, Dahlen JR, Anderberg JM, McPherson PH, Nakamura KK, Laskowitz DT, Valkirs GE, & Buechler KF (2003). Early biomarkers of stroke. *Clin Chem*, **49**, 1733-1739.

Riggs CE, Jr., Johnson DJ, Konopka BJ, & Kilgour RD (1981). Exercise heart rate response to facial cooling. *Eur J Appl Physiol Occup Physiol*, **47**, 323-330.

Riggs CE, Jr., Johnson DJ, Kilgour RD, & Konopka BJ (1983). Metabolic effects of facial cooling in exercise. *Aviat Space Environ Med*, **54**, 22-26.

Roelands B, Hasegawa H, Watson P, Piacentini MF, Buyse L, De SG, & Meeusen RR (2008a). The effects of acute dopamine reuptake inhibition on performance. *Med Sci Sports Exerc*, **40**, 879-885.

Roelands B, Hasegawa H, Watson P, Piacentini MF, Buyse L, De SG, & Meeusen R (2008b). Performance and thermoregulatory effects of chronic bupropion administration in the heat. *Eur J Appl Physiol*, **105**, 493-498.

Roelands B, Goekint M, Heyman E, Piacentini MF, Watson P, Hasegawa H, Buyse L, Pauwels F, De SG, & Meeusen R (2008c). Acute norepinephrine reuptake inhibition decreases performance in normal and high ambient temperature. *J Appl Physiol*, **105**, 206-212.

Romet TT (1988). Mechanism of afterdrop after cold water immersion. *J Appl Physiol*, **65**, 1535-1538.

Rubenstein E, Meub D, & Eldridge FA (1960). Common carotid blood temperature. *J Appl Physiol*, **15**, 603-604.

Russell RD, Redman SM, Ravussin E, Hunter GR, & Larson-Meyer DE (2004). Reproducibility of endurance performance on a treadmill using a pre-loaded time trial. *Med Sci Sports Exerc*, **36**, 717-724.

Saboisky J, Marino FE, Kay D, & Cannon J (2003). Exercise heat stress does not reduce central activation to non-exercised human skeletal muscle. *Exp Physiol*, **88**, 783-790.

Saltin B, Gagge AP, & Stolwijk JA (1970). Body temperatures and sweating during thermal transients caused by exercise. *J Appl Physiol*, **28**, 318-327.

Sawka MN & Wenger CB (1988). Physiological responses to acute exercise-heat stress. In *Human Performance Physiology and Environmental Medicine at Terrestrial Extremes*, ed. Gonzalez RR, pp. 97-151. Cooper Publishing Group, Carmel.

Schabort EJ, Hopkins WG, & Hawley JA (1998). Reproducibility of self-paced treadmill performance of trained endurance runners. *Int J Sports Med*, **19**, 48-51.

Schabort EJ, Hawley JA, Hopkins WG, & Blum H (1999). High reliability of performance of well-trained rowers on a rowing ergometer. *J Sports Sci*, **17**, 627-632.

Schmidt V & Bruck K (1981). Effect of a precooling maneuver on body temperature and exercise performance. *J Appl Physiol*, **50**, 772-778.

Schulpis KH, Moukas M, Parthimos T, Tsakiris T, Parthimos N, & Tsakiris S (2007). The effect of alpha-Tocopherol supplementation on training-induced elevation of S100B protein in sera of basketball players. *Clin Biochem*, **40**, 900-906.

Sharma HS, Cervos-Navarro J, & Dey PK (1991). Increased blood-brain barrier permeability following acute short-term swimming exercise in conscious normotensive young rats. *Neurosci Res*, **10**, 211-221.

Shiraki K, Sagawa S, Tajima F, Yokota A, Hashimoto M, & Brengelmann GL (1988). Independence of brain and tympanic temperatures in an unanesthetized human. *J Appl Physiol*, **65**, 482-486.

Shvartz E (1970). Effect of a cooling hood on physiological responses to work in a hot environment. *J Appl Physiol*, **29**, 36-39.

Shvartz E (1976). Effect of neck versus chest cooling on responses to work in heat. *J Appl Physiol*, **40**, 668-672.

Shvartz E, Ben-Mordechai Y, Aldjem M, & Magazanik A (1976). Neck and back cooling in a hot environment. *Isr J Med Sci*, **12**, 796-799.

Simmons SE, Mundel T, & Jones DA (2008). The effects of passive heating and head-cooling on perception of exercise in the heat. *Eur J Appl Physiol* 271-280.

Sleivert GG, Cotter JD, Roberts WS, & Febbraio MA (2001). The influence of whole-body vs. torso pre-cooling on physiological strain and performance of high-intensity exercise in the heat. *Comp Biochem Physiol A Mol Integr Physiol*, **128**, 657-666.

Smiles KA, Elizondo RS, & Barney CC (1976). Sweating responses during changes of hypothalamic temperature in the rhesus monkey. *J Appl Physiol*, **40**, 653-657.

Smith MF, Davison RC, Balmer J, & Bird SR (2001). Reliability of mean power recorded during indoor and outdoor self-paced 40 km cycling time-trials. *Int J Sports Med*, **22**, 270-274.

Stalnacke BM, Tegner Y, & Sojka P (2003). Playing ice hockey and basketball increases serum levels of S-100B in elite players: a pilot study. *Clin J Sport Med*, **13**, 292-302.

Stalnacke BM, Ohlsson A, Tegner Y, & Sojka P (2006). Serum concentrations of two biochemical markers of brain tissue damage S-100B and neurone specific enolase are increased in elite female soccer players after a competitive game. *Br J Sports Med*, **40**, 313-316.

Strachan AT, Leiper JB, & Maughan RJ (2004). Paroxetine administration failed [corrected] to influence human exercise capacity, perceived effort or hormone responses during prolonged exercise in a warm environment. *Exp Physiol*, **89**, 657-664.

Strachan AT, Leiper JB, & Maughan RJ (2005). Serotonin2C receptor blockade and thermoregulation during exercise in the heat. *Med Sci Sports Exerc*, **37**, 389-394.

Stroud MA (1991). Effects on energy expenditure of facial cooling during exercise. *Eur J Appl Physiol Occup Physiol*, **63**, 376-380.

Sukstanskii AL & Yablonskiy DA (2004). An analytical model of temperature regulation in human head. *J Therm Biol*, **29**, 583-587.

Sukstanskii AL & Yablonskiy DA (2007a). Theoretical limits on brain cooling by external head cooling devices. *Eur J Appl Physiol*, **101**, 41-49.

Sukstanskii AL & Yablonskiy DA (2007b). Theoretical limits on brain cooling by external head cooling devices. *Eur J Appl Physiol*, **101**, 41-49.

Sunderland C, Morris JG, & Nevill ME (2008). A heat acclimation protocol for team sports. *Br J Sports Med*, **42**, 327-333.

Tatterson AJ, Hahn AG, Martin DT, & Febbraio MA (2000). Effects of heat stress on physiological responses and exercise performance in elite cyclists. *J Sci Med Sport*, **3**, 186-193.

Taylor CR (1970). Dehydration and heat: effects on temperature regulation of East African ungulates. *Am J Physiol*, **219**, 1136-1139.

Thomas MM, Cheung SS, Elder GC, & Sleivert GG (2006). Voluntary muscle activation is impaired by core temperature rather than local muscle temperature. *J Appl Physiol*, **100**, 1361-1369.

Towfighi J, Housman C, Heitjan DF, Vannucci RC, & Yager JY (1994). The effect of focal cerebral cooling on perinatal hypoxic-ischemic brain damage. *Acta Neuropathol*, **87**, 598-604.

Tucker R, Rauch L, Harley YX, & Noakes TD (2004). Impaired exercise performance in the heat is associated with an anticipatory reduction in skeletal muscle recruitment. *Pflugers Arch*, **448**, 422-430.

Uckert S & Joch W (2007). Effects of warm-up and precooling on endurance performance in the heat. *Br J Sports Med*, **41**, 380-384.

Veghte JH & Webb P (1961). Body cooling and response to heat. *J Appl Physiol*, **16**, 235-238.

Walters TJ, Ryan KL, Tate LM, & Mason PA (2000). Exercise in the heat is limited by a critical internal temperature. *J Appl Physiol*, **89**, 799-806.

Watanuki S (1993). Effects of head cooling on cardiovascular and body temperature responses during submaximal exercise. *Ann Physiol Anthropol*, **12**, 327-333.

Watson P, Shirreffs SM, & Maughan RJ (2005a). Blood-brain barrier integrity may be threatened by exercise in a warm environment. *Am J Physiol Regul Integr Comp Physiol*, **288**, R1689-R1694.

Watson P, Shirreffs SM, & Maughan RJ (2005b). The effect of passive elevation of core temperature on serum S-100B, a peripheral marker of blood-brain barrier permeability. *Med Sci Sports Exerc*, **37**, 1231.

Watson P, Hasegawa H, Roelands B, Piacentini MF, Looverie R, & Meeusen R (2005c). Acute dopamine/noradrenaline reuptake inhibition enhances human exercise performance in warm, but not temperate conditions. *J Physiol*, **565**, 873-883.

Watson P, Black KE, Clark SC, & Maughan RJ (2006). Exercise in the heat: effect of fluid ingestion on blood-brain barrier permeability. *Med Sci Sports Exerc*, **38**, 2118-2124.

Watson P, Love TD, Maughan RJ, & Shirreffs SM (2008). A comparison of the effects of milk and a carbohydrate-electrolyte drink on the restoration of fluid balance and exercise capacity in a hot, humid environment. *Eur J Appl Physiol*, **104**, 633-642.

Webster J, Holland EJ, Sleiverts G, Laing RM, & Niven BE (2005). A light-weight cooling vest enhances performance of athletes in the heat. *Ergonomics*, **48**, 821-837.

Williams PA & Kilgour RD (1993). Cardiovascular responses to facial cooling during low and moderate intensity exercise. *Eur J Appl Physiol Occup Physiol*, **67**, 53-58.

Wilson TE, Johnson SC, Petajan JH, Davis SL, Gappmaier E, Luetkemeier MJ, & White AT (2002). Thermal regulatory responses to submaximal cycling following lower-body cooling in humans. *Eur J Appl Physiol*, **88**, 67-75.

Wilson WM & Maughan RJ (1992). Evidence for a possible role of 5-hydroxytryptamine in the genesis of fatigue in man: administration of paroxetine, a 5-HT re-uptake inhibitor, reduces the capacity to perform prolonged exercise. *Exp Physiol*, **77**, 921-924.

Yamazaki F & Sone R (2000). Modulation of arterial baroreflex control of heart rate by skin cooling and heating in humans. *J Appl Physiol*, **88**, 393-400.

Young AJ, Sawka MN, Epstein Y, Decristofano B, & Pandolf KB (1987). Cooling different body surfaces during upper and lower body exercise. *J Appl Physiol*, **63**, 1218-1223.

Zetterberg H, Jonsson M, Rasulzada A, Popa C, Styrud E, Hietala MA, Rosengren L, Wallin A, & Blennow K (2007). No neurochemical evidence for brain injury caused by heading in soccer. *Br J Sports Med*, **41**, 574-577.

Zhu L (2000). Theoretical evaluation of contributions of heat conduction and countercurrent heat exchange in selective brain cooling in humans. *Ann Biomed Eng*, **28**, 269-277.

Zhu L & Diao C (2001). Theoretical simulation of temperature distribution in the brain during mild hypothermia treatment for brain injury. *Med Biol Eng Comput*, **39**, 681-687.

Zhu M, Ackerman JJ, Sukstanskii AL, & Yablonskiy DA (2006). How the body controls brain temperature: the temperature shielding effect of cerebral blood flow. *J Appl Physiol*, **101**, 1481-1488.

Zimmer DB & Landar A (1995). Analysis of S100A1 expression during skeletal muscle and neuronal cell differentiation. *J Neurochem*, **64**, 2727-2736.

Zimmer DB, Cornwall EH, Landar A, & Song W (1995). The S100 protein family: history, function, and expression. *Brain Res Bull*, **37**, 417-429.

"The more I see, the more I know…

…the more I know, the less I understand"

The Changingman, Paul Weller

Appendices

Appendix A1

NOTTINGHAM TRENT UNIVERSITY

PARTICIPANT INFORMATION SHEET

The effect of a cooling collar on exercise capacity in the heat (2)

You are being invited to take part in a research study. Before you decide it is important for you to understand what it will involve. Please take time to read the following information carefully and discuss it with others if you wish. Ask us if there is anything that is not clear or if you would like more information. Take time to decide whether or not you wish to take part.

Protocol and requirements

We will arrange for you to attend the laboratory on five separate occasions, each lasting approximately 60-minutes. The first visit will consist of a maximal oxygen uptake test where you will be asked to complete a submaximal incremental treadmill test followed by a maximal test to determine your maximum oxygen uptake (VO2max). The following four visits will constitute the main study and will treadmill running test at ~70% VO2max until volitional fatigue in hot conditions (30°C; 50%RH) on each occasion. The first trial will be a habituation session that will give you the opportunity to become accustomed with the laboratory environment and the exercise protocol used in this study. Prior to the start of the first trial you will be asked to abstain from caffeine and alcohol and complete a 24-hour food record sheet- you will also be required to repeat this diet on the day before each of the remaining trials. You will also be asked to abstain from vigorous exercise for 24-hours prior to each main trial.

Main Trials

Following the preliminary maximum oxygen uptake session you will carry out four main trials in a randomised order with ~7days between each main trial.

During the sessions you will be asked to run with a cooling collar, a cooling collar replaced to maintain a reduced neck temperature or with no collar.

You will arrive at the laboratory at least 2 hours after eating. Following self-assessed nude body weight. Resting core temperature, skin temperature, heart rate, perceived comfort and blood samples (via finger-prick) will be taken while at rest in the test conditions. After baseline measurements have been taken you will be asked to run at a predetermined intensity until you can run no further ("volitional fatigue"). Water will be allowed ad libitum throughout the trial. Core temperature, heart rate, thermal sensation, rating of perceived exertion will be taken at regular intervals throughout the trial. Nude body weight will be taken at the commencement of exercise.

Benefits of taking part

From the information obtained I will also be able to give you individual feedback on your performance and your individual physiological responses to the exercise sessions.

Risks of taking part
The exercise protocol used in this study (i.e. exhaustive exercise) is demanding but the stress that accompanies this exercise soon disappears once the exercise has stopped.

Core temperature will be measured via rectal thermometer self-inserted ~10cm past the anal sphincter. It will be constantly assessed and the trial will be terminated if it exceeds 40.5°C.

It is up to you to decide whether or not to take part. If you do decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still free to withdraw at any time and without giving a reason.

General and specific requirements
- You must be a male non-smoker aged 18-45 years.
- You must not have recently gained or lost a significant amount of weight.
- You will be required to record your diet for one day prior to the main period of testing and repeat this prior to each trial.
- You should not engage in any demanding exercise or physical activity during the 48 hours before each main trial.
- You will be required to refrain from alcohol consumption during the 48 hours before each main trial.
- You will be required to refrain from caffeine consumption during the 24 hours before each main trial.
- You will be required to arrive at the laboratory at least two hours after eating.

The results of this study will be submitted for publication, but no personal information will be included in any such publication. Blood samples will be stored until analysis is complete.

Investigator contact information:
If you would like any further information about this study, or related studies, please contact:

Mr Christopher Tyler *	Tel: 0115 8486601	christopher.tyler@ntu.ac.uk
Dr Caroline Sunderland	Tel: 0115 8486379	caroline.sunderland@ntu.ac.uk

* Primary contact

Appendix A2

SPORT AND EXERCISE SCIENCE
NOTTINGHAM TRENT UNIVERSITY

HEALTH SCREEN QUESTIONNAIRE

Name: _____ Date: _____

It is important that volunteers participating in research studies are currently in good health and have had no significant medical problems in the past. This is to ensure (i) their own continuing well-being and (ii) to avoid the possibility of individual health issues confounding study outcomes.

Please complete this brief questionnaire to confirm fitness to participate.

1. **At present**, do you have any health problem for which you are:
   (a) On medication, prescribed or otherwise ..... Yes ☐ No ☐
   (b) Attending your general practitioner ..... Yes ☐ No ☐
   (c) On a hospital waiting list ..... Yes ☐ No ☐

2. **In the past two years**, have you had any illness which requires you to:
   (a) Consult your GP ..... Yes ☐ No ☐
   (b) Attend a hospital outpatient department ..... Yes ☐ No ☐
   (c) Be admitted to hospital ..... Yes ☐ No ☐

3. **Do you, or have you ever had,** any of the following:
   (a) Convulsion/epilepsy ..... Yes ☐ No ☐
   (b) Asthma ..... Yes ☐ No ☐
   (c) Diabetes ..... Yes ☐ No ☐
   (d) Any blood disorder ..... Yes ☐ No ☐
   (e) Head injury ..... Yes ☐ No ☐
   (f) Digestive problems ..... Yes ☐ No ☐
   (g) Heart problems ..... Yes ☐ No ☐
   (h) Problems with bones or joints ..... Yes ☐ No ☐
   (i) Disturbance of balance/co-ordination ..... Yes ☐ No ☐
   (j) Ear/hearing problems ..... Yes ☐ No ☐
   (k) Thyroid problems ..... Yes ☐ No ☐
   (l) Kidney or liver problems ..... Yes ☐ No ☐
   (j) Problems with your diet ..... Yes ☐ No ☐

If **YES** to any question, please describe briefly if you wish (e.g. to confirm whether the problem was/is short-lived, insignificant or well-controlled).

.....

.....

.....

Additional questions for **female** participants
(a) Are your periods normal/regular? ..... Yes ☐ No ☐
(b) Are you on 'the pill'? ..... Yes ☐ No ☐
(c) Could you be pregnant? ..... Yes ☐ No ☐
(d) Are you on hormone-replacement therapy? ..... Yes ☐ No ☐

Signature _____

Date _____

BELOW: To be completed on subsequent visits to the laboratory, when these visits are more than one week after the form was first completed.

Please read the form again and sign and date the boxes below to confirm that your situation has not changed since your first visit to the lab when you completed this form.

	Visit 2	Visit 3	Visit 4	Visit 5
Signature Date				
	Visit 6	Visit 7	Visit 8	Visit 9
Signature Date				

If anything has changed since your first visit, please use the space below to briefly provide information:

.....

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Appendix A3

ETHICAL COMMITTEE
APPLICATION FOR CATEGORY B HUMAN BIOLOGICAL INVESTIGATIONS

(i) Applicants:

Mr Christopher Tyler (Primary researcher)
Dr Caroline Sunderland (Primary supervisor)
Prof Mark Darlison (Secondary supervisor)

(ii) Project Title:

The effects of a neck cooling device on exercise capacity in the heat

(iii) Aims and Outline of the Project:

Background:

During exercise the human body is mechanically inefficient which results in the production of large quantities of heat causing the core temperature to rise. Fatigue is multi-factorial however it has been suggested that during exercise in elevated temperatures hyperthermia is the single greatest reason for the termination of physical activity. With this in mind there has been substantial interest in cooling interventions however this has primarily focused on whole-body interventions which lack practical application. It has been suggested that cooling the neck during hyperthermia may be more efficacious in reducing heat strain than cooling areas of equal surface area elsewhere and such interventions have been shown to attenuate the rate at which core temperature and heart rate rises as well as decreasing sweat rate and perceptions of task difficulty and thermal comfort in some, but not all, studies. Although the benefit of cooling has been widely shown the depth at which the investigations were conducted has failed to conclusively identify what the benefits can be attributed to.

Aims of the project:

The aim of this project is to investigate the effects of a neck cooling device on prolonged treadmill running in elevated ambient conditions with special focus on exercise capacity and the physiological responses to such exercise.

(iv) Names and status of investigators:

Mr Christopher Tyler (Primary researcher)
Dr Caroline Sunderland (Primary supervisor)
Prof Mark Darlison (Secondary supervisor)

(v) Subjects:

Subjects will all be male and under forty years of age. They will be recruited from local running clubs and from within the university, via word of mouth, website advertisements and a poster campaign. All will be taking part voluntarily and will be aware of their ability to leave the study at any time. Due to the demanding nature of the study all the subjects will be well-trained and of good health and will be required to complete a health screening questionnaire prior to each laboratory session.

(vi) Location (any special facilities to be used):

Sports Physiology Laboratory
Human Performance Laboratory
Environmental walk-in Chamber

(all located within the Erasmus Darwin Building at Nottingham Trent University, Clifton Campus, Clifton Lane, Nottingham, NG11 8NS)

(vii) Duration (including demand on subject's time):

Each subject will be required to attend the laboratory on five occasions. The initial visit will last approximately one hour while the other four visits will all last approximately one hour resulting in a total of five hours of laboratory time per subject.

The duration of the study depends on the final number of subjects recruited (aim = 10) but should last no longer than six months from the date of ethical approval.

(viii) Reasons for undertaking the study (e.g. contract, student research):

PhD student research

(ix) Methodology (a brief outline of research design):

On their first visit to the laboratory subjects will perform an incremental maximal oxygen uptake test on a motorised treadmill in ambient conditions. The subsequent four visits will be comprised of a capacity treadmill test in which subjects will run at 70-80% maximal oxygen uptake until volitional fatigue. The four main laboratory visits will be conducted in hot ambient conditions [~30°C, 50% relative humidity] in a randomised trial order. The first of the four visits will act as a habituation during which the subject will be made familiar with the testing protocol and equipment used. During the other three main trials subjects will complete one trial without a cooling collar, one with a cooling collar in a cooled state and one with a collar in a cooled state replaced when required to maintain neck temperature cooled. Trial order will be randomised for the three experimental trials.

For the experimental trials subjects will be at least 2 hours post-prandial.

(iv) Procedures and measurements

Expired gas samples: Expired gas samples will be collected during the habituation trial at 20min intervals using the Douglas bag technique.
Blood Samples: Venepuncture samples will be taken pre and post exercise by a trained and experienced phlebotomist. Blood samples will analysed for haematocrit, haemoglobin, glucose and lactate with later analysis for prolactin, cortisol and catecholamines.
Nude body weight: Nude body weight will be recorded before and after each trial. This will be conducted by the subject themselves in the strictest privacy.
Core Temperature: Core temperature will be measured using a fully calibrated rectal thermometer inserted ~10cm past the anal sphincter.
Skin temperature: Skin temperature will be measured at four sites by calibrated skin thermisters and mean skin temperature calculated via weighted formula.
Subjective report: Thermal sensation and rating of perceived exertion will be recorded at regular intervals using fully validated scales.
Heart Rate: Heart rate will be monitored throughout.

(xi) Possible risks, discomforts and/or distress:

Heat illness such as heat stroke (however all investigators have experience with exercise in the heat and so are well qualified to identify the onset of associated symptoms and stop subjects prior to their development). The core temperature of each subject will constantly be monitored and trials will be terminated if the temperature exceeds 40.5°C. Heart rate (a good indicator of cardiovascular strain) will also be constantly monitored as will subject well-being in the form of verbal communication.

(xii) Procedures for taking measurements and for chaperoning and supervision of subjects during investigations:

Nude body weight will be recorded before and after each trial. This will be conducted by the subject themselves in the strictest privacy.

(xii) Names of investigators and personal experience of proposed procedures and/or methodologies:

Mr Chris Tyler, Dr Caroline Sunderland

Both have extensive experience of VO2max testing, core temperature assessment and gas and blood sampling and analysis during undergraduate and postgraduate study (Dr Sunderland's PhD thesis: "Effect of heat acclimation on field hockey skill performance" and Chris Tyler's previous PhD studies) in ambient and hot conditions with elite and sub-elite individuals.

(xiv) Details of any payments to be made to the subjects

None

(xv) Do any investigators stand to gain from a particular conclusion of the research project:

No

(xvi) Whether the University's Insurers have indicated that they are content for the University's Public Liability Policy to apply to the proposed investigation (Committee use only):

(xvii) Whether the insurance cover additional to (xvi) has been arranged by the Investigator:

(xviii) In the case of studies involving new drugs or radioisotopes, written approval for the study must be obtained from the appropriate national body and submitted with the protocol. State if applicable:

(xix) Declaration

I have read the University's Ethical Constitution and Protocol Document and completed this application.

Signature of applicant: .....

Signature of Head of Department: .....

Date: .....

Appendix B1

Raw data for the calculation of the coefficient of variation for whole blood lactate and glucose measurements

Whole blood Lactate ($mmol \cdot l^{-1}$)

Gender		**M**	**M**	**M**	**M**	**M**	**F Post Ex**	**M Post Ex**	**F Post Ex**
Condition		**Rest**	**Rest**	**Rest**	**Rest**	**Rest**			
0-5 mins	1	1.16	0.905	1.37	1.07	1.25	4.43	7.94	5.94
post-sample	2	1.15	0.925	1.32	0.99	1.32	4.40	8.26	6.23
	3	1.19	0.921	1.35	1.02	1.29	4.41	8.13	6.09
5-10 mins	1	1.14	0.825	1.53	1.14	1.25	4.33	7.86	6.24
	2	1.16	0.845	1.44	1.09	1.34	4.39	8.07	6.01
	3	1.18	0.895	1.48	1.14	1.28	4.42	8.77	6.15
1 SD		0.02	0.04	0.05	0.03	0.05	0.05	0.48	0.12
Mean		1.16	0.89	1.42	1.07	1.29	4.40	8.17	6.11
CV		1.72	4.07	3.19	2.69	3.56	1.04	5.83	1.90

Summary: 0 - 10 min sample

1 SD	0.10
Mean	3.06
CV %	**3.00**

Whole blood glucose ($mmol \cdot l^{-1}$)

Gender		**M**	**M**	**M**	**M**	**M**	**F Post Ex**	**M Post Ex**	**F Post Ex**
Condition		**Rest**	**Rest**	**Rest**	**Rest**	**Rest**			
0-5 mins	1	4.94	5.04	3.70	4.85	4.02	4.03	4.93	4.2
post-sample	2	4.92	5.01	3.68	4.24	4.25	4.10	4.95	4.32
	3	4.90	5.02	3.66	4.62	4.15	4.09	4.91	4.27
5-10 mins	1	4.92	4.56	3.45	5.25	3.98	4.01	3.92	4.37
	2	4.97	4.44	3.29	4.94	4.2	4.05	4.15	4.21
	3	4.98	4.42	3.31	5.01	3.95	4.06	4.12	4.22
1 SD		0.03	0.08	0.09	0.16	0.14	0.03	0.13	0.09
Mean		4.94	4.75	3.52	4.82	4.09	4.06	4.50	4.27
CV		0.65	1.59	2.48	3.37	3.34	0.65	2.78	2.10

Summary: 0 – 10 min sample

1 SD	0.09
Mean	4.37
CV %	**2.12**