RESPONSES OF BOYS AND MEN EXERCISING IN THE HEAT

BOYS AND MEN EXERCISING IN THE HEAT: THERMOREGULATION, DEHYDRATION AND PERFORMANCE, AND SUBSTRATE UTILIZATION

By

GABRIELA TOMEDI LEITES, MSc Human Movement Sciences

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AUTHOR:	Gabriela Tomedi Leites, M.Sc
SUPERVISOR:	Dr. Brian W. Timmons
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LAY ABSTRACT

Children may not adapt to temperature extremes as effectively as adults. Thermoregulatory differences may be due to variable exercise intensities prescribed in available child-adult comparisons. Dehydration affects aerobic performance, but its effects on strength are not well understood. Sports drinks are often used to offset dehydration and improve performance; however, the body may have a harder time using sugar drinks when exercising in the heat. The purpose of this thesis was to compare bodily responses of boys and men during exercise in the heat. Our findings have important practical implications for boys and men who exercise and play sports in hot temperatures. Specifically, exercise in the heat is equally safe for boys and men. The effects of hypohydration and a sports drink on muscle strength performance following exercise in the heat are minimal. Finally, although exercise in the heat may affect the use of a sugar drink, boys and men may use this drink to improve aerobic performance, while maintaining hydration levels.

ABSTRACT

There is a common belief that children compared with adults are at a greater risk for exercise-induced heat illness. However, a limitation of previous studies involves different exercise intensities used in the comparison between children and adults. Dehydration impairs aerobic performance, but its effects on strength are not well understood. Sports drinks are often used as ergogenic aids. There is some evidence to suggest children have a greater reliance on exogenous carbohydrate compared to adults, which could require special considerations when children exercise in the heat. The overall purpose of this thesis was to compare physiological and metabolic responses during exercise in the heat between boys and men. Three studies were conducted to 1) compare the thermoregulatory responses of boys and men exercising in the heat at a fixed absolute metabolic heat production or a fixed metabolic heat production per unit of body mass; 2) compare the effects of 2% hypohydration or, euhydration with and without carbohydrate ingestion during exercise in the heat on subsequent strength in boys and men; and 3) examine the effects of exogenous carbohydrate on endogenous metabolism in boys and men exercising in the heat. Age and body size differences between boys and men did not influence thermoregulatory responses at a fixed metabolic heat production per unit of body mass. No differences in strength were observed among trials. When exercising in the heat, the relative contribution of ingested carbohydrate and endogenous substrate to total energy yield were not

different between groups. These findings together have practical implications for boys and men who exercise in the heat. Specifically, moderate exercise in the heat is equally safe for boys and men. The effects of hypohydration and a carbohydrate drink compared to water on muscle strength performance following exercise in the heat are minimal. An important practical application is that carbohydrate intake spared endogenous fuels during exercise in the heat in both groups.

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LIST OF ABBREVIATIONS

Abbreviations	Definition of terms	Units
ANOVA	Analysis of variance	ND
BSA	Du Bois body surface area	m^{-2}
BSA_r	Effective radiating area of the body	m^{-2}
BM	Body mass	kg
C	Convective heat loss per unit area	$W{\cdot}m^{-2}$
CARB	Carbohydrate trial	ND
CHO	Carbohydrate	ND
CHO_{exo}	Exogenous carbohydrate oxidation	$g{\cdot}min^{-1}$
CHO_{endo}	Endogenous carbohydrate oxidation	$g{\cdot}min^{-1}$
CHO_{total}	Rate of total carbohydrate oxidation	$g{\cdot}min^{-1}$
CONT	Control trial	ND
C_{res}	Respiratory losses through convection	$W{\cdot}m^{-2}$
DXA	Dual-energy X-ray absorptiometry	Kg
E	Evaporation	$W{\cdot}m^{-2}$
E_{max}	Maximal evaporative potential per unit area	$W{\cdot}m^{-2}$
E_{req}	Required evaporative loss per unit area for heat balance	W, $W{\cdot}m^{-2}$
E_{res}	Evaporative loss from respiration	$W{\cdot}m^{-2}$
E_{sk}	Total evaporative heat loss from the skin	$W{\cdot}m^{-2}$
e_c	Caloric equivalent per liter of oxygen for the oxidation of carbohydrate	kJ
e_f	Caloric equivalent per liter of oxygen for the oxidation of fat	kJ
EU-CHO	Euhydration with carbohydrate solution trial	ND
EU-W	Euhydration with water trial	ND
Ƒ	Partial pressure of water vapor in air	mmHg

Fat_{total}	Total fat oxidation	$g{\cdot}min^{-1}$
H	Combined heat transfer coefficient	$W{\cdot}m^{2}{\cdot}K^{-1}$
h_c	Convective heat transfer coefficient	$W{\cdot}m^{2}{\cdot}K^{-1}$
h_e	Evaporative heat transfer coefficient	$W{\cdot}m^{2}{\cdot}KPa^{-1}$
h_r	Radiative heat transfer coefficient	$W{\cdot}m^{2}{\cdot}K^{-1}$
HR	Heart rate	Bpm
$\dot{H}_p$	Metabolic heat production	W, $W{\cdot}kg^{-1}$
HY	Hypohydration trial	ND
K	Conduction	$W{\cdot}m^{2}$
LBM	Lean body mass	Kg
LLMM	Lower limb muscle mass	Kg
M	Metabolic energy expenditure	$W{\cdot}m^{2}$
MEN_{REL}	Men exercising at fixed metabolic heat production per unit total body mass	$W{\cdot}kg^{-1}$
MEN_{ABS}	Men exercising at fixed absolute metabolic heat production	W
MVCs	Maximal voluntary contractions	$Nm{\cdot}LMM^{-1}$
R	Radiative heat loss per unit area	$W{\cdot}m^{2}$
RER	Respiratory exchange ratio	ND
RH	Relative humidity	%
RPE	Rate of perceived exertion	ND
S	Heat storage	W, $W{\cdot}m^{-2}$
SD	Standard deviation	ND
t_a	Ambient temperature	^{o}C
t_b	Mean body temperature	^{o}C
T_{re}	Mean rectal temperature	^{o}C
T_{sk}	Mean skin temperature	^{o}C
USG	Urine specific gravity	ND

ULMM	Upper limb muscle mass	kg
V	Air velocity (air speed)	$m \cdot s^{-1}$
VO_{2peak}	Peak oxygen uptake	$ml \cdot kg^{-1} \cdot min^{-1}$
w	Skin wittedness	ND
W	External mechanical work per unit area	$W \cdot m^{-2}$
W_{max}	Maximal workload	W

FORMAT AND ORGANIZATION OF THIS THESIS

This thesis was prepared in the “sandwich thesis” format outlined in the McMaster University School of Graduate Studies Guide for the Preparation of Master’s and Doctoral Theses, published in November 2014.

Chapter 1 is an introduction and literature review, which sets the context for the complete body of research. Chapters 2, 3, and 4 consist of three manuscripts, which are either published or under peer-review. These chapters are formatted in accordance with the requirements of the journal in which they are published or submitted for publication. Chapter 5 is a concluding chapter which summarizes and discusses overall findings of this thesis and includes future research directions. Finally, appendices are also included to supplement chapters.

CONTRIBUTIONS TO MULTI-AUTHORED PAPERS

CHAPTER 2.

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CHAPTER 1. INTRODUCTION

Children often perform physical activity in an outdoor environment and there is a commonly held belief that children do not adapt to extremes of temperature as effectively as adults due to differences in their body size, physiology, and metabolism (Bar-Or 1989a; Bergeron 2011). Indeed, thermoregulatory responses to exercise in the heat may be influenced by unique physical and physiological changes that occur as a part of growth and biological maturation. Among these are morphological and physiological characteristics such as a lower sweating rate, a higher body surface area to body mass ratio, a slow acclimatization to the heat, and a reduced cardiac output at a given metabolic rate in children compared to adults (Bar-Or and Rowland 2004; Falk et al. 1992a; Inoue et al. 2004; Meyer et al. 1992). These observations have led to the widely held notion that physically active youth have an increased risk of heat-illness and exercise intolerance in hot environments. However, it is important to note that early studies often lacked direct child-adult comparisons and failed to prescribe an exercise intensity to elicit a fixed metabolic heat production, which may have biased study findings. In adults, recent reports suggest that the use of an inappropriate exercise intensity for heterogeneous groups (e.g. differences in aerobic fitness, body mass and body surface area) may affect comparisons of thermoregulatory responses (Cramer and Jay 2014; Jay et al. 2011). Thus, selection of exercise intensity is a critical consideration for appropriate comparisons of thermoregulatory responses to exercise in the heat in children and adults.

During prolonged periods of exercise in the heat, children usually do not replace all fluid losses, resulting in dehydration (Bar-Or et al. 1980a). Dehydration due to sweat loss and insufficient fluid replacement can impair physiological function. Abundant research documents the detrimental effects of hypohydration on endurance performance (Cheuvront et al. 2003; Kavouras et al. 2012; Sawka et al. 2001; Shirreffs 2005). However, there are a small number of studies with conflicting results that investigated the impact of hypohydration on strength, power, and high-intensity endurance (Judelson et al. 2007a). The effect of hydration status on these variables remains unclear. The most widely accepted theory postulates that hypohydration impairs the ability of the central nervous system to stimulate the musculature or inhibits muscle membrane excitability (Judelson et al. 2007a). A child's physical performance could be impaired by hypohydration, at shorter and more intense exercise efforts than adults. Indeed, previous studies showed that even low levels of hypohydration (1% and 2%) affect high-intensity endurance performance in 10- to-12 year old boys exercising the heat (Wilk et al. 2013). Dougherty et al. (2006) showed that in boys, suicide sprint times (high-intensity exercise) were higher in the 2% of hypohydration trial and lower in euhydration in the 6% CHO-electrolyte solution trial compared to the euhydration with water trial. Although this study showed that hypohydration can impair high-intensity endurance performance in children while consuming a carbohydrate-electrolyte solution can improve performance, more studies are needed involving different types of exercise such as muscle strength. Muscle strength is one of the single most important parameters for successful performance across a wide variety of sports. To the best of our knowledge, no study has compared the impact of hypohydration induced by exercise in the heat and the impact of carbohydrate intake during exercise in the heat on muscle strength between children and adults.

During exercise in a hot environment, sports drinks can offset dehydration and enhance physical performance (Jentjens et al. 2002a). In adults, prolonged exercise combined with heat stress can affect metabolism by decreasing exogenous carbohydrate oxidation and increasing the reliance on endogenous carbohydrate metabolism. Studies suggest this could be caused by a reduction in the absorptive capacity of the intestine during exercise in the heat, as well as an increase in muscle glycogen utilization without a compensatory change in muscle glucose uptake (Jentjens et al. 2002a). The potential reduction in exogenous carbohydrate metabolism in the heat compared to cool conditions may require special considerations for children, given the existing evidence that supports a greater reliance on exogenous carbohydrate during the exercise in youth compared with adults. More information is required to identify optimal carbohydrate intake before exercise in the heat for children to enhance performance, however not provide excessive carbohydrate consumption that may contribute to health complications over time. A study by Timmons et al. (2003) used ^{13}C isotope methodology to compare substrate utilization in pre- and early- pubertal boys and men. Boys demonstrated a greater reliance on exogenous carbohydrate as source of energy compared to men during 60 min of cycling at 70% VO_{2peak} (Timmons et al. 2003a). While the majority of studies have focused on hydration status and substrate utilization in thermoneutral environments, exercise in the heat may have detrimental effects on metabolism. Therefore, more studies are needed to understand the substrate utilization during exercise in the heat in order to avoid heat illness and enhance performance in children and adults.

Therefore, this thesis intends to investigate three major questions:

1. Do thermoregulatory responses in a hot environment differ between boys and men exercising at a fixed metabolic heat production per unit total body mass and a fixed absolute heat production?
2. Does hypohydration induced by an exercise in the heat protocol affect muscle strength performance of boys differently than men? Additionally, does exogenous carbohydrate during exercise in the heat affect muscle strength performance differently in boys and men?
3. Does carbohydrate intake during exercise in a hot environment affect endogenous substrate utilization differently in boys and men?

1.1 Thermoregulation and environment conditions

Temperature can be considered as the average kinetic energy of the molecules, which is the heat in a body. Humans are homeotherms (warm-blooded) and attempt to keep their core temperature at about 37°C. This means that, on average, heat production and heat transfer must be balanced by heat output from the body (Ozisick 1985). The thermoregulatory system is responsible for detecting deviations from normal body temperatures, interpreting their consequences, and taking action to restore homeostasis. These actions or responses may be behavioural (e.g. opening a window, changing clothes) or physiological (e.g. sweating, changes in blood flow).

The human body's temperature is greatly affected by the surrounding temperatures of solids and fluids, which influence heat transfer to and from the body. Heat transfer is driven by a temperature gradient (from hot to cold). This is called sensible heat transfer and takes place by conduction, convection and radiation (Gagge and Gonzalez 2011); definitions adapted from Parsons (2014)). *Conduction* is the heat transfer through solids surfaces by direct physical contact, and the rate of heat flow depends on the thermal conductivity of the material. Usually, heat transfer by conduction from the human body to the environment is small and often ignored. *Convection* is a more complex process than conduction because it entails the mass movement to transfer thermal energy from one site to another (Ozisick 1985). It involves transfer of heat from a solid surface (e.g., the body) to a moving gas or fluid (e.g. air or water), which then absorbs the heat by mixing and bulk movement resulting in an increase in body's temperature (Nelson and Eichna 1947). *Radiation* is part of the electromagnetic spectrum. When absorbed by the skin surface, radiation with wavelengths in the visible light to radio wave spectrum promotes heat exchange. Heat is gained if mean radiant temperature is greater than skin temperature, and lost if skin temperature exceeds mean radiant temperature.

E*vaporation,* defined as the change of state from liquid to vapor, is also involved in heat transfer. Evaporative heat loss is a practical physiological strain indicator of increases in environmental heat stress (Gagge and Gonzalez 2011). The process begins with the transition of sweat on the skin surface from liquid to vapor at a constant temperature (latent heat transfer). Then, the vapor diffuses away from the surface in a similar manner as heat diffusion in convection (Parsons 2014). Sweat evaporation from the skin has a cooling effect called evaporative cooling. *S*weat evaporation is the most effective mechanism of heat loss during the exercise and is the only way humans dissipate heat when skin temperature is greater than the ambient temperature.

Metabolic heat production represents the free energy produced by the transformation of chemical energy during exercise (mechanical energy) within an organism (Nishi 1981). The energy expenditure is the rate of free energy released from carbohydrate, fat and amino acids catabolism to adenosine triphosphate resynthesis. One of the by-products of this reaction is heat production (thermal energy). During physical activities, greater heat production is observed from the increase in oxygen uptake to enable active muscles to meet its energetic demands. Metabolic heat is produced in the cells of the body and is transferred to the surroundings by conduction (due to the temperature gradient) and by convection (due to the movement of extracellular fluid, such as blood) (Parsons 2014). During exercise if metabolic heat production and heat gain (inputs) are greater than heat losses (outputs), the body temperature will rise. Heat inputs and outputs can be calculated from equations of heat transfer. The conceptual equation for heat balance is (Gagge and Gonzalez 2011; Nishi 1981):

$$M - W = E + R + C + K + S$$

where: M is the metabolic heat production; W is the external mechanical workload; E is evaporation; R is radiation; C is convection; K is conduction; and S is heat storage.

Numerous models of the human thermoregulatory system have been proposed. It is well accepted that the hypothalamus is home to core temperature sensors, and acts as a central regulator of body temperature. There are two additional types of peripheral temperature sensors: the warm and cold receptors, which are free nerve endings that are widely distributed superficially and within the epidermis (Weddel and Miller, 1962). Signals from both core and peripheral sensors are integrated at the hypothalamus (Gagge et al. 1971). It has been suggested that there may be a complex whole-body integrated system, probably related to temperature and rate of temperature change, which has yet to be discovered (Parsons 2014). It has also been suggested that peripheral sensors are less sensitive to changes in body temperature compared to core sensors, but they may play an important anticipatory role for early adjustments (Kenney et al. 2012). When the hypothalamus detects a variation in temperature (e.g. an increase in body core temperature during prolonged exercise from the heat produced by muscle contraction), effectors are stimulated by the sympathetic nervous system thereby triggering compensatory cooling mechanisms. This primarily involves: 1) vasodilatation of skin's arterioles by the neurotransmitter norepinephrine, which then directs blood flow to the skin. For example, for each 1°C rise above thermoneutral body temperature, blood flow increases by 56 $g \cdot s^{-1} \cdot m^{-2}$; and 2) activation of eccrine glands by the neurotransmitter acetylcholine (sympathetic cholinergic stimulation), leading to an increase sweating rate (Kenney et al. 2012; Parsons 2014). The magnitude of convective heat loss is governed by the local skin-air temperature gradient as well as the adequacy of cutaneous blood flow. Heat loss by evaporation is directly tied to both sweat production and the skin-air vapor pressure gradient between environment and skin (Rowland 2008).

The thermal environment also influences thermal sensation and comfort (Fanger 1973). Little is known about the relationship between environmental conditions and physical and physiological factors in the thermoregulatory response. Thermal sensation is a sensory experience related to how a person feels (i.e. cold, neutral, warm, etc.). It is often used to adjust the temperature so as to provide comfortable environments (Gagge et al. 1969; Parsons 2014). To comprehensively assess human responses to thermal environments, it is necessary to quantify the environmental conditions and its effects in terms physiological and perceptual responses.Although principles for assessment of human thermal environment, and principles and recommendations for determining human thermoregulation and comfort may be appropriate for the general population, there are particular populations that may require special consideration, including the pediatric population.

1.2 Relationship among thermoregulatory responses, growth and maturation

Evidence suggests that children may be at higher risk of developing heat-illnesses compared to adults (Bar-Or and Rowland 2004), due to differences in their respective morphology, sweating rate, metabolism and cardiovascular responses during exercise (Bar-Or 1989b; Bar-Or et al. 1980b; Falk 1998; Falk and Dotan 2008). Heat illnesses occur secondary to prolonged exposure to the heat, and include heat cramps (severe, spreading and sustained painful muscle contractions that result from prolonged exercise, usually at high ambient temperatures), heat exhaustion (inability to continue exercise; presence of any combination of heavy sweating, dehydration, sodium loss, and energy depletion – may or may not affect body-core temperature), and heat stroke (elevated core temperature (>39^{o}C) accompanied by signs of organ system failure due to hyperthermia) (Binkley et al. 2002). The Centers for Disease Control and Prevention (Centers for Disease and Prevention 2011) in a document (*National Electronic Injury Surveillance System – All Injury Program)* reported that, between 2001 and 2009, heat illnesses caused by sports and recreation were more prevalent among males (72.5%) and among those aged 10–14 years (18.2%) and 15–19 years (35.6%). Interestingly, despite the impression that children are at high risk of heat-related illness, few empirical examples are available (Rowland 2008). Nevertheless, there remain unanswered questions surrounding safe exercise in the heat for children.

1.2.1 Morphological differences

The most obvious morphological differences between children and adults are body size and muscle mass, both of which are significantly smaller in children (Bar-Or 1989b; Falk 1998; Rowland 2005). Body surface area affects heat dissipation, as the process of sweat evaporation and convection eliminates body heat at the skin surface (Rowland 2008). Relative to body mass, an 8- to-9-year old child, for example, has a body surface area (heat radiator) that is ~40% greater than an adult (Bar-Or 1989b; Falk 1998; Falk and Dotan 2008; Inbar et al. 2004). In thermoneutral conditions, the greater body surface area related to body mass may be advantageous since children rely more on dry heat dissipation (radiation, convection and conduction) and less so on evaporative cooling (Falk and Dotan, 2008, Rowland, 2008). On the other hand, children absorb more heat from the environment in hot conditions compared to adults. Some authors (Havenith 2001; Naughton and Carlson 2008) have criticized this concept as being over simplistic, due to confounding factors such as body composition, fitness, sex and type and duration of exercise, which influence the link between body surface area per unit body mass and heat loss (Rowland 2008). In warm environments, children use evaporative cooling as the main route for body heat elimination during prolonged activities (Falk and Dotan, 2008, Maughan et al., 2007, Bar-Or and Rowland, 2004); however a child's sweat glands are described as smaller, immature and, consequently, less efficient compared to an adult.

1.2.2 Body core temperature

It was suggested that a child's exercise tolerance may be impaired when exercising in the heat (Bar-Or and Rowland 2004), which may be linked to maturational factors, sex, and differences in sweating rate. Indeed, one might expect prepubescent children to demonstrate a greater increase in body core temperature in hot environments compared to adults. In a classic study, Bar-Or et al. (Bar-Or et al. 1980a) found that the rate of change in rectal temperature during exercise was positively related to dehydration, and was twice as high in children as the rise observed in a separate study of adults (Bar-Or et al. 1976). This suggested that dehydration might impose greater health risks for children, particularly when exercising in the heat.

Conversely, other studies (Davies 1981; Falk et al. 1992b; Rowland et al. 2008; Wagner et al. 1972) found no age-related differences in the change in body core temperature during exercise in a hot environment. Armstrong and Maresh (1995) compiled results of different studies and reported the average rise in temperature during exercise in the heat was 1.24 ± 0.4 ^{o}C in children and 1.21 ± 0.39 ^{o}C in adults.

It is also important to note that a number of researchers (Drinkwater et al. 1977a; Falk et al. 1992b; Inbar et al. 2004; Shibasaki et al. 1997) have described age-related differences in thermoregulation despite similar increases in core temperature in children and adults exercising in the heat. Inbar et al. (2004) showed no difference in the change in rectal temperature between prepubertal boys and young men exercising at 50% VO_{2max}; however, they suggested that boys were more efficient thermoregulators due to differences in heat balance (i.e. estimated body mass-related heat-gain needed to raise T_{re} by 1 ^{o}C). Shibasaki et al. (1997) evaluated seven prepubertal boys and 11 young men who cycled for 45 minutes at ~40% VO_{2peak} in a warm environmental condition (30 ^{o}C and 45% relative humidity). They found no difference in the core temperature increase between boys and men during exercise; however, absolute final core temperature was greater in boys compared to men (37.4 vs. 37.0 ^{o}C respectively, $P < 0.05$). Based on these observations, physically active youth have been considered at greater risk for exercise-induced heat illness and exercise intolerance in the heat, despite the fact that these early studies often lacked direct child-adult comparisons and failed to prescribe an exercise intensity to elicit heat balance parameters, such as metabolic heat production.

A possible explanation for the aforementioned findings relates to differences in metabolic heat production between children and adults during exercise. It may be that thermoregulatory responses are not different between the groups when the exercise intensity elicits similar metabolic heat production, regardless of any physiological or anatomic feature unique to children. The similar change in core temperature reported in children and adults exercising at the same relative intensity (%VO_{2peak}) is likely not due to relative intensity, *per se*, but due to the same heat production in W per unit body mass. To the best of our knowledge, no study has directly compared children and adults exercising at the same metabolic heat production or same metabolic heat production per unit of body mass; therefore, studies are necessary to confirm this possible explanation.

1.2.3 Cardiovascular responses

At a similar absolute peak oxygen uptake (VO_{2peak}, $L \cdot min^{-1}$ or $mL \cdot min^{-1}$), children have lower cardiac output compared to adults, which may indicate lower cardiovascular efficiency in children (Bar-Or and Shepard 1971; Bar-Or et al. 1971; Falk and Dotan 2008; Rowland 2005). To maintain body core temperature homeostasis during exercise, blood flow to the skin is increased. Subsequently, heat is transported to the periphery, allowing for evaporative sweat loss and heat dissipation. The adequacy of the circulatory response is determined by body fluid content and blood volume (Rowland 2008).

Early studies suggested that younger children have lower blood volume relative to their body mass and body surface area compared with adolescents (Falk et al. 1992). To account for their greater body surface are per unit of body mass, children may exhibit an increase in cardiac output to ensure adequate perfusion in the periphery and subsequently maintain body core temperature (Falk 1998). In other words, a child's ability to transport heat to the periphery is likely affected by these differences to a greater degree than an adult during exercise in the heat. Furthermore, a greater proportion of cardiac output in children is diverted to the periphery under heat stress (Drinkwater et al. 1977b). Falk et al. (1992a) found a greater forearm blood flow in prepubertal compared to mid- and late- pubertal boys, while cycling at 50% VO_{2max} in the heat (Falk et al. 1992b). Shibasaki et al. (1997) evaluated seven prepubertal boys and 11 young men cycling for 45 minutes at ~40% of VO_{2max} in a warm environmental condition (30°C and 45% relative humidity). Cutaneous blood flow on the chest and back was greater in boys than in men ($P < 0.003$); but forearm blood flow and cutaneous vascular conductance were lower in boys compared to men ($P < 0.008$). The authors speculated that the age-related differences in cutaneous vascular conductance were due to lower mean arterial pressure in the boys during exercise. However, it has been argued that these differences are related to differences in the exercise intensity, as children did not exercise at the same absolute VO_2 ($mL \cdot min^{-1}$) as adults, inducing differences in metabolic heat production.

Even with differences in methodological design, such as environmental conditions, sex, and levels of dehydration, some studies showed no differences in cardiovascular responses between children and adults when exercise was performed at a percentage of VO_{2peak}. Rivera-Brown et al. (2006) compared cardiovascular responses and skin blood flow during an exercise tolerance protocol in pre-menarcheal girls and women. Participants cycled at 60% of VO_{2peak} in a hot and humid outdoor environment (33.4 ^{o}C and 55% relative humidity) and body fluid status was maintained by prescribed drinking. No differences in stroke index, heart rate or cardiac index were found between groups. Forearm skin blood flow increased during exercise but did not differ between girls and women. Rowland et al. (2008) found no difference between boys and men in cardiac responses during exercise to exhaustion at 65%VO_{2peak} in the heat (31 ^{o}C and 50% relative humidity), while hydration status was maintained by fluid intake. Stroke index, mean arterial pressure, and arterial venous oxygen differences remained stable in both groups. These results indicate that in hot environments, cardiovascular responses to exercise stress are effective in both children and adults. It is important to note that these studies avoided dehydration, as such the impact of hypohydration levels on these responses might differ.

In male adults, Sawka et al. (1985) observed that hypohydration level (3, 5, and 7 % body mass loss) could affect blood characteristics during aerobic exercise in the heat (49°C). Low-to-moderate hypohydration levels initially reduced plasma volume by ~3-4 % with little effect on plasma osmolality. Severe hypohydration caused no further reduction in plasma volume; however, an increase in plasma osmolality was observed (Sawka et al. 1985). The dehydration-induced decrease in plasma volume led to a decrease in stroke volume and an increase in heart rate to maintain cardiac output, while meeting the increasing demand for oxygen and nutrients during exercise. In addition, it is speculated that exercise-induced hypovolemia (decreased volume of circulating blood) may reduce blood flow to the brain leading to a greater rating of perceived exertion during exercise (Maughan et al. 2007).

1.2.4 Acclimatization

Acclimatization to heat is a gradual process that is achieved with regular exposure to naturally hot environments. The major physiological changes that progressively occur include: a decrease in heart rate and rectal and skin temperature at a given metabolic level, an increase in sweating rate and sensitivity, a reduction in electrolyte concentration in the sweat, a reduction in rating of perceived exertion, better maintenance of body core temperature and distribution of blood flow to the body and skin, and an increase in thermal comfort. Although children adapt physiologically to the heat, the process seems to be slower than in adults (Bar-Or 1989b). Thus, children may be more vulnerable to heat-related illnesses when high temperature seasons begin since they experience a longer acclimatization process (Bergeron et al. 2005; Godek et al. 2005). In addition, exercise training improves thermoregulation in the heat by eliciting an earlier onset of sweat secretion and by increasing the total amount of sweat that can be produced.

1.2.5 Sweating Responses

The number of sweat glands is determined in childhood, thus the density of glands at the skin surface decreases with growth (Falk, 1998; Sato et al., 1987). The number of sweat glands (eccrine and apocrine) varies, ranging from 1.6 to 4.0 million. Eccrine glands are responsible for thermoregulatory sweating (Shibasaki et al. 2006). Sweating is controlled in the hypothalamus, specifically where thermosensitive neurons are located and sweating is activated when skin temperature increases (Shibasaki et al. 2006). A number of factors can influence sweating rate, such as an individual's genetics, baseline level of acclimatization, fitness, and age/maturation status. In addition, exercise intensity, exercise modality, and environmental conditions (e.g., humidity, temperature, solar radiation, and clothing) can also affect sweating rate (Maughan et al. 2007; Meyer et al. 2012).

Sweating rate demonstrates great variability, as shown consistently in both children (Meyer et al. 2012) and adults (Maughan et al. 2007; Sawka et al. 2007). There is evidence to suggest that compared with adults, children experience a lower absolute sweating rate and lower sweating rate relative to body surface (Falk and Dotan 2008; Meyer et al. 1994). On average, a child's sweating rate is about 400-500 $mL{\cdot}m^{-2}{\cdot}h^{-1}$, while adults experience a sweating rate of 700-800 $mL{\cdot}m^{-2}{\cdot}h^{-1}$ (Armstrong 2007b). A study by Armstrong and Maresh (1995) showed that sweating rate was ~40% greater in men compared with pre-pubertal boys during exercise. Consequently, it is expected that boys depend more on cutaneous blood flow for heat loss compared to adults (Rowland 2005). On the other hand, there is some evidence to suggest no difference in sweating rates in pre-pubertal girls and women. Drinkwater et al. (1977) compared sweating rate in five pre-pubertal girls and five young female university students. They were required to run on a treadmill under thermoneutral (28°C and 45% RH), hot (35°C and 65% RH), and very hot (48°C and 10% RH) environmental conditions. They found that sweating rate was similar between groups (Drinkwater et al. 1977b). Rivera-Brown et al. (2006) compared the physiological responses to an exercise tolerance protocol of nine pre-pubertal girls and nine women cycling in the heat and also observed no differences in sweating rate between groups. These studies demonstrate that the difference in sweating rate between children and adults may be sex-dependent, and seems to be more apparent in boys than in girls during exercise in the heat.

However, it is also possible that the difference observed in sweating responses between boys and men may have been biased by the exercise intensity, which was set to a percentage of VO_{2peak}. In the adults, it is known that a higher absolute required evaporative loss for heat balance (E_{req}) induces a greater amount of absolute sweat rate (Gagnon et al. 2013). Therefore, when exercise is prescribed to achieve a similar percentage of VO_{2peak} instead of a similar E_{req}, it may induce significant differences in the amount of sweat in children and adults. No previous study has compared the sweating rate of boys and men exercising at an intensity prescribed to elicit a similar E_{req}. Studies are needed to test this hypothesis and confirm possible differences or similarities between boys and men when exercise is prescribed by the E_{req}.

1.2.6 Fluid balance and dehydration

Body hydration describes body water status, and can be classified as euhydration, hyperhydration and hypohydration (Armstrong 2007b). Euhydration refers to normal body water content. Under thermoneutral conditions, the body water content of a healthy active subject should change by about ± 0.25%, and in warm conditions body water deficit may increase by approximately 0.8%. Hyperhydration occurs when body water content is above normal, and, as with hypohydration, is undesirable. Although they are often used synonymously, exercise-induced dehydration and hypohydration refer to two different concepts. Dehydration is the active process of losing water, while hypohydration refers to a state of body water deficit. Both are caused by a mismatch of overall water loss and intake (Bergeron 2011; Mitchell et al. 1972). Inadvertent dehydration is a term used to describe the behaviour of insufficient fluid intake to replenish the losses, even when fluids are available to drink *ad libitum* (Bar-Or and Rowland 2004; Greenleaf 1992).

A hot and humid climate may impose a greater stress on various physiological systems compared to a thermoneutral condition. Thus, children who exercise in the heat are at risk of experiencing dehydration through fluid loss and insufficient fluid replacement. Hypohydration can be evaluated by comparing the quantity of body fluid lost to baseline body weight. This can be done by examining changes in body weight, which provide the simplest and most accurate index of hydration status in real time, when serial measurements are made in close proximity (Armstrong 2007a). Fluid loss which is less than 2-3% of baseline body weight is considered mild hypohydration, 4-5% is considered moderate, and greater than 6% is considered severe hypohydration (Casa et al. 2000; Meyer et al. 2012; Sawka et al. 2007). In adults, a 1-2% loss of body weight can increase body core temperature, a 3% loss can lead to a reduction in aerobic performance, while a 4-6% loss can cause cramps and thermal fatigue, and above 6% loss poses an increased risk to the heart (Greenleaf 1992; Sawka 1992). Hypohydration symptoms include red skin, fatigue, thirst, heat intolerance, and decreased and darker urine. There are more than 13 hydration assessment techniques that can be used in the laboratory and field to measure human hydration, including urinary parameters such as urine specific gravity and color.

It is suggested that from a hydration physiology point of view children are different from adults (Guelinckx et al. 2015). Compared to adults, children have a higher body water content relative to body mass, and may lose more water from the skin (relative to BSA) at rest and under thermoneutral conditions (Altman, 1961, Novak, 1989). Due to these differences, children may have higher water requirements per unit of body mass and require greater attention to avoid body fluid imbalances (Altman, 1961, Novak, 1989).

Interestingly, children commonly experience dehyhydration even when fluids are available *ad libitum (*inadvertent dehydration). This was shown in a study by Bar-Or et al. (1980a) with twelve, 10- to 12-year-old boys who were partially acclimatized. They cycled at 45% maximal aerobic capacity at 39ºC and 45% RH. During one session, children could drink *ad libitum* and in another, adequate fluid intake was ensured to replenish fluid losses. The authors concluded that exercising children progressively dehydrate when they were not forced to drink. On the other hand, in a study conducted by Rivera-Brown et al. (2008), 12 acclimatized girls cycled for 80 min at 60% VO_{2max} in the heat (31°C) in three exercise sessions with fluid available to drink ad libitum. Participants received different drinks at each visit (water, flavoured water or flavoured water plus 6% carbohydrate and ~18mml NaCl), and the authors reported that regardless of the drink, hypohydration was prevented (Rivera-Brown et al. 2008). Wilk et al. (2007) evaluated twelve, 9- to 12 year old girls who were not acclimatized to the heat. They participated in three exercise sessions in the heat (35°C), which were divided into four bouts of 20 minutes of cycling at 50% VO_{2max}. Over the three sessions, water or grape flavoured water plus carbohydrate and electrolytes were provided ad libitum and the girls remained hydrated (Wilk et al. 2007). This suggests that fluid availability is important for children during exercise in a hot environment. However, occurrence of dehydration is often described in young athletes practicing and competing in team sports, but the degree to which it affects specific performance skills is not clearly understood.

1.3 Hypohydration and Performance

Hypohydration impairs body core temperature regulation and may limit endurance performance (Bar-David et al. 2005; D'Anci et al. 2006; Rivera-Brown et al. 2006). Evidence shows that in adults, hypohydration accelerates the increase in core temperature and adds stress to the cardiovascular system during exercise, especially in the heat, by reducing total plasma volume, increasing submaximal heart rates and decreasing maximal cardiac output (Binkley et al. 2002). Hypohydration can impair physical performance, increase exhaustion, rate of perceived exertion and risk of heat-illness, and decrease motivation (Cheuvront et al. 2010; Dotan and Bar-Or 1980; Meyer et al. 2012). Hypohydration affects muscle metabolism by accelerating the rate of glycogen depletion, as well as central nervous system function by reducing motivation and effort (Hampson et al. 2001; Meyer et al. 2013).

Several studies show the detrimental effects of hypohydration on endurance performance (Cheuvront et al. 2003; Sawka et al. 2001; Shirreffs 2005). Conversely, there are a small number of studies with conflicting results on the impact of hypohydration on strength, power, and high-intensity endurance (Judelson et al., 2007). The most widely accepted theory is that hypohydration affects some neuromuscular component that impairs the ability of the central nervous system to stimulate the musculature or muscle membrane excitability (Judelson et al., 2007). Another theory is that body water loss affects water and electrolyte concentrations (Na^+, Cl^-, K^+ e Mg^{+2}) inside the cells, which in turn impair membrane excitability (Costill et al, 1976).

During sports practice and competitions, athletes become progressively hypohydrated when sweat loss exceeds fluid intake (Aragon-Vargas et al. 2013; Arnaoutis et al. 2014; Casa et al. 2000; Dougherty et al. 2006). It is believed that a child's physical performance could be impaired by hypohydration at shorter and more intense efforts than adults. Understanding whether and how hypohydration affects muscle performance in boys and men is important to provide recommendations for the appropriate hydration needs of these particular groups and guarantee their optimal performance.

Table 1.1 (see Appendix A) summarizes studies that assessed the impact of hypohydration on power and high intensity endurance performance in children and on muscle strength performance in adults. Wilk et al. (2013) examined the effect of hypohydration (0, 1 and 2%) induced by exercise in the heat on high-intensity endurance performance in 10- to 12-year-old boys. Mean time to exhaustion was 26.8% shorter at 2% hypohydration compared to euhydration, and mechanical work was 23.3% lower at 2% hypohydration compared to euhydration. Time to exhaustion at 1% hypohydration was 15.6% shorter compared to euhydration trial, and total work was 15.5% lower. These results suggest that in children even mild (~ 1%) to moderate (~ 2%) hypohydration reduces high-intensity cycling performance in the heat in a sample of healthy young boys.

Many young athletes are involved in sports such as soccer, basketball, handball, indoor soccer, rugby , tennis, hockey, and gymnastics, which are characterized by high-intensity, intermittent bouts of exercise that demand skilled performance over a long period of time (Meyer et al. 2012). Dougherty et al. (2006) examined the effects of exercise in the heat-induced 2% hypohydration, euhydration with 6% CHO-electrolyte solution, and euhydration with flavoured water on basketball skills in fifteen 12- to 15-year-old male players. Performance and component drills inherent to basketball were measured, such as combined shooting percentages (3-point, 15-foot, free-throw), sprints (suicides, court widths), lateral movement (zigzags, lane slides), defensive drills (combining lateral and front-to-back movement) times. Suicide sprint times were ~2 sec (5.4%) longer in 2% of dehydration trial and ~3 sec (7.5%) shorter in euhydration with a 6% CHO-electrolyte solution trial compared to the euhydration with flavoured water trial. Maximal vertical jump was similar among hypohydration, euhydration with 6% CHO-electrolyte solution and euhydration with flavoured water trials (68, 66 and 66 cm, respectively). Carvalho et al. (2011) evaluated twelve adolescent basketball athletes in three 90-min sessions in the following conditions: no fluid available (2.5 % hypohydration), water available to drink ad libitum (1.0 % hypohydration), and 8.0 % carbohydrate-electrolyte sports beverage available to drink ad libitum (0.65% hypohydration). No differences were observed in suicide sprint times between conditions (31.1, 30.9 and 30.9 seconds, respectively). Although some studies showed that hypohydration can impair high intensity endurance in children and that consuming a carbohydrate-electrolyte solution can improve high intensity and power performance, more studies are needed to clarify these results. In addition, it is necessary to investigate the effects of hypohydration and carbohydrate beverage specifically on muscle strength performance in children, as muscle strength is among the single most important physiological parameters for successful performance across a wide variety of sports (Andersen et al. 2010). The effects of hypophydration on muscle strength performance has, however, been addressed only in adults.

In a review about hydration and muscular performance in adults, Judelson et al. (2007a) suggested that differences in methodology exacerbate or attenuate the apparent effects of hypohydration, explaining the conflicting results across the body of literature. When these factors were taken into account (such as body temperature, caloric restriction, sauna and exercise protocol, exercise intensity, heat exposure), hypohydration resulted in reductions of ~ 2.0 % in strength, ~3.0 % in power and ~10.0 % in high intensity endurance. More recent studies (Hayes and Morse 2010; Rodrigues et al. 2014) suggested that in young adults moderate hypohydration (~ 2 %) induced by exercise in the heat can impair isometric knee extension a similar (Vallier et al. 2005) or greater extent (Kraemer et al. 2001) than previous studies. Rodrigues et al. (2014) also showed a 6.7 % reduction in isometric elbow extension with 2% hypohydration. Greenleaf et al. (1967) showed similar reductions (7.0 % and 9.5 % in left and right elbow respectively) with 3-4% hypohydration induced by exercise in the heat and control group (euhydration). On the other hand, Kraemer et al. (2001) induced hypohydration with a combination of techniques (one week caloric restriction, thermal stress and exercise the day before) in young male wrestlers and showed a lower impairment in elbow flexion (2.8 %) compared with previous studies (typically > 5 %). The variability of results presented most likely stems from the methodology employed to induce hypohydration (such as active heat exposure, passive heat, fluid restriction, diuretics, combined techniques), the main outcome measured (muscle strength isometric and isokinetic, high-intensity, power) and/or confounding factors (such as fatigue, body core temperature, caloric restriction, menstrual status, hydration status in baseline, experience in training, type of sport) (Bowtell et al. 2013; Judelson et al. 2007a). Although further studies are need to clarify the impact of hypohydration on muscle strength and elucidate the mechanisms, these studies suggest that hypohydration is an important factor to consider when attempting to measure maximal muscle performance, at least in young adults.

1.4 Oxidation rate of exogenous carbohydrates and substrate utilization: influence of environmental conditions and biological maturation

Carbohydrate sports beverages ingested during endurance exercise provide fluid to offset dehydration and are effective and commonly used ergogenic aids for endurance sports (Coyle 2004; Saunders 2007). During prolonged exercise, CHO availability is important to improve performance and postpone fatigue (Below et al. 1995). Possible mechanisms for the ergogenic effect of CHO ingestion are the maintenance of blood glucose levels and the increased ability to maintain high CHO oxidation over time (Jentjens et al. 2006; Jentjens et al. 2002b). Additionally, it has been suggested that CHO_{exo} may have a central effect in the brain that affects performance and perception of effort, possibly mediated through receptors in the mouth or intestinal tract for sweetness or specifically for CHO (Carter et al. 2005; Carter et al. 2004). Numerous factors may affect the oxidation rate of CHO_{exo} such as exercise intensity, modality, amount, type and timing of CHO ingestion, environment conditions, and biological maturation (Jentjens et al. 2002b; Jeukendrup and Jentjens 2000; Timmons et al. 2003a). Prolonged exercise and heat stress may affect metabolism in adults by increasing the reliance on CHO metabolism and reducing CHO_{exo} oxidation (Jentjens et al. 2002a). This could be caused by an increase in muscle glycogen utilization without a compensatory change in muscle glucose uptake, and a reduced absorptive capacity of the intestine during exercise in the heat (Jentjens et al. 2002a). The potential reduction in CHO_{exo} metabolism in the heat might be of particular concern for children given the existing evidence that supports a greater reliance on CHO_{exo} during the exercise in youth compared with adults. More information is required to identify optimal carbohydrate intake before exercise in the heat for children to enhance performance, however not provide excessive carbohydrate consumption that may contribute to health complications over time.

In adults, exercise in the heat increases muscle glycogen utilization, resulting in a shift in substrate utilization toward a greater reliance on CHO metabolism (Febbraio et al. 1994b; Hargreaves et al. 1996). Febbraio et al. (1994a) also showed that exercise in the heat alters muscle glycogenolysis, but heat acclimation can revert these changes. Fink et al. (1975) assessed the effects of environmental heat stress on muscle metabolism in six males cycling at 70-85% VO_{2peak} for 3×15 min bouts. Exercise was performed in the hot (41°C and 15 % RH) and cold (9°C and 55 % RH) settings. They showed that blood lactate concentration and muscle glycogen utilization was greater in hot conditions compared to the cold. Additionally, muscle triglyceride declined 23% during exercise in the cold and 11% in the heat. These findings may be a result of a decrease in cardiac output and the redistribution of blood flow to facilitate heat transfer to the skin during exercise in the heat. This redistribution may in turn lead to a reduction in the oxygen supply to the working muscle, thereby accelerating the rate of muscle glycogen depletion and lactate accumulation. Other early studies (Fink et al. 1975; Rowell et al. 1968; Williams et al. 1962) also assessed the metabolic response during exercise in hot conditions and reported similar results. In contrast, some studies failed to observe effects of exercise in the heat on muscle glycogenolysis (Nielsen et al. 1990; Yaspelkis et al. 1993). These contradictory findings may be related to the fact that participants were not provided with CHO_{exo}, as well as some methodological differences such as environment conditions (i.e.

immediately moving from thermoneutral to hot environment in the same session or small differences in temperature between thermoneutral and hot environments).

Few studies have assessed the effects of heat stress on CHO_{exo} oxidation during exercise in adults. In a thermoneutral environment, even when large amounts of CHO_{exo} (2.4-3.0 $g \cdot min^{-1}$) were ingested during prolonged exercise at light to moderate intensity (50-65% VO_{2peak}), the oxidation rate did not exceed 1.1 $g \cdot min^{-1}$ (Jeukendrup et al. 1999; Wagenmakers et al. 1993). In addition, even during longer exercises of 180 min, rates of CHO_{exo} oxidation are also limited to 1.0-1.1 $g \cdot min^{-1}$ (Bosch et al. 1994; Hawley et al. 1992).

While most studies examining CHO_{exo} oxidation during exercise have been performed in cool or thermoneutral environments, a few studies to date have evaluated CHO_{exo} oxidation and substrate utilization during exercise in the heat in adults, as outlined in Table 1.2 (see Appendix B). Jentjens et al. (2002b) compared nine trained male cyclists pedaling for 90 minutes at 55% VO_{2peak} in a hot (35.4 ^{o}C) and cool (16.4 ^{o}C) environment on separate occasions. They received 8% glucose solution enriched with [U-^{13}C] glucose. Initial T_{re} was similar between hot and cool conditions (37.1 vs. 36.9 ^{o}C, respectively), but final T_{re} was greater in the hot compared to cool environment (39.1 vs. 38.3 ^{o}C). Heat stress reduced the rate of CHO_{exo} oxidation (0.76 vs. 0.84 $g \cdot min^{-1}$) and increased the oxidation rate of total CHO (3.18 vs. 2.85 $g \cdot min^{-1}$) (Jentjens et al. 2002a). The authors suggested that the uptake and release of ingested glucose by the liver, glucose transport into the muscle, gastric emptying, and intestinal absorption are factors that contributed to these results. It is important to note that this study may be limited by the fact that hypohydration levels were greater during exercise in the heat, which may have reduced muscle blood flow. In theory, the exercise in the heat may have increased blood flow to the skin for evaporative cooling, resulting in a reduction of flow to other organs and limiting intestinal absorption of CHO_{exo} compared to the cool condition. However, another study showed that dehydration reduced leg blood glucose, but did not impair glucose delivery and glucose uptake (Gonzalez-Alonso et al. 1999).

Jentjens et al. (2006) evaluated eight trained, non-acclimated male cyclists and triathletes, aged ~28 years old, during three separate trials consisting of 120 min of cycling at 50%W_{max} in the heat (31.9 ^{o}C and 30% RH). The only difference between trials was the drink availability: 9% glucose + fructose (GLU + FRU), 9% glucose (GLU), or plain water. Oxidation rate of CHO_{exo} was greater in GLU+FRU compared to GLU trial (1.12 vs. 0.74 $g \cdot min^{-1}$, respectively) at the end of exercise. No differences were found in ΔT_{re} (~1.9 ^{o}C) and in T_{skin} (~34 ^{o}C). In this study, ingestion of GLU+FRU during exercise in the heat resulted in higher CHO_{exo} oxidation rate compared to GLU and reduced CHO_{endo} compared to water.

Carter et al. (2005) compared the ingestion of water, a sweet and an unsweetened 6.4% CHO solution on exercise capacity in hot environments in endurance-trained men cycling at 60% VO_{2peak} to exhaustion. They showed that compared to water, exercise capacity was ~16% greater with the sweet solution and ~12% greater with the unsweetened solution. CHO oxidation rate was greater in the sweet and unsweetened solutions compared to water. There was a trend for plasma glucose concentrations to be higher in both CHO solutions compared to water. These results showed that CHO during exercise in the heat prolongs time to fatigue compared to water. Dumke et al. (2013) showed that CHO oxidation was similar between a placebo and a 6% glucose beverage in males during exercise in the heat, but fat oxidation was 25% lower in the CHO trial compared to placebo. This study suggests that CHO availability during exercise can alter the metabolic response and perhaps result in increased reliance on fat stores when carbohydrate availability is low. Comparisons of studies are difficult due to differences in the purpose and methodological approach of each study. Nevertheless, it seems that exercise in the heat affects substrate utilization, resulting in a greater reliance on endogenous metabolism compared to cool and thermoneutral conditions. In adults, CHO_{exo} oxidation is lower in hot environments. The effect of CHO_{exo} on endogenous substrate utilization has not yet been evaluated during exercise in the heat in children.

Historically, differences in metabolism during exercise between children and adults have been described (Morse et al. 1949; Robinson 1938). Indeed, Robinson (1938) originally demonstrated lower respiratory exchange ratio (RER) during exercise in younger compared to older adolescents and adults, which was attributed to a reduced CHO reserve. However, he argued that prolonged fasting before exercise testing caused the observed endogenous CHO reduction in younger participants. Years later, Morse et al. (1949) confirmed lower RER in children compared to adults during exercise. The measurement of RER, an indicator of respiratory quotient, and provides information about substrate utilization. An RER of 1.0 indicates greater reliance on carbohydrate utilization, while 0.7 suggests fat utilization. Other studies also reported reduced RER in children compared to adults during prolonged, moderate intensity exercise (Boisseau and Delamarche 2000; Martinez and Haymes 1992), indicating that children utilize more fat and less carbohydrate for energy. Although RER does have its limitations for quantifying substrate utilization, this technique has been widely used to describe and compare substrate utilization in children and adults during exercise (Riddell 2008).

Several studies have examined the effect of maturation status on substrate utilization. Martinez and Haymes (1992) evaluated ten 8- to 10-year-old girls and ten 20- to 32-year-old women running on a treadmill for 30 min at 70% VO_{2peak} and at an absolute intensity of 7.2 $km \cdot h^{-1}$. Girls showed 43% higher fat oxidation levels and 19% lower CHO_{end} oxidation compared to women at the same relative intensities. No differences were described when groups were running at same absolute intensity. Rowland and Rimany (1995) evaluated eleven 9- to 13-year-old girls and thirteen 20- to 31-year-old women cycling for 40 min at 63% VO_{2peak}. No differences were observed in substrate utilization during exercise. Both groups demonstrated lower RER at the end of exercise (40 min) compared to the beginning (10 min) (fall from 0.95 to 0.85 in girls and 0.97 to 0.85 in women). Foricher et al. (2003) compared substrate utilization in fourteen prepubertal and thirteen adult male swimmers, cycling for 60 min at 40 to 60% maximal workload (W_{max}). They showed that at 40% W_{max} prepubertal swimmers oxidized less CHO and more fat compared to adults, similarly, children oxidized less CHO at 60% W_{max} compared to men.

Kaczor et al. (2005) aimed to examine the effects of age on the activity of enzymes involved in anaerobic and aerobic metabolism in human skeletal muscle from relatively sedentary children and adults. They compared samples of the *obliquus internus* and *abdominis* muscle of twenty 3- to 11-year-old children and twelve 29- to 54-year-old adults who underwent hernia surgery. Carnitine palmitoyl-transferase/2-oxoglutarate dehydrogenase (CPT/OGDH) ratio enzyme activity in skeletal muscle was 16% higher in children compared to adults, suggesting a preferential oxidation of fatty acids over other substrates in children. Stephens et al. (2006) also showed higher fat oxidation in prepubertal boys, based on their RER, compared to pubertal boys exercising between 40 and 70% of their VO_{2peak}. Riddell et al. (2008) tested the range of exercise intensities that elicit high fat oxidation rates in youth and the influence of pubertal status. They determined the peak fat oxidation rate during cycling for boys was 8 mg·kg$^{-1 lean\ body\ mass}$ at 60% VO_{2peak} and in 5 mg·kg^{-1} fat free mass at 40% VO_{2peak}. In addition, a 3-year longitudinal study in adolescent boys showed their relative fat oxidation rates decreased throughout puberty, and the exercise intensity required to elicited peak fat oxidation also decreased.

In summary, during submaximal exercise children's enhanced ability to oxidize lipids, and therefore spare glycogen, means that they are well-equipped for long-duration moderate intensity exercise. However, this phenomenon seems more apparent in boys. The mechanism for the higher rate of fat oxidation in children compared to adults is still unknown (Riddell 2008). Low muscle glycogen content is possibly associated with a reduced activity of glycolytic enzymes and high oxidative capacity, while lower levels of sympathoadrenal hormones are likely to favour lipid metabolism in children. Changes in metabolism may also be related to hormone differences, such as testosterone in boys and progesterone and estrogen in girls (Aucouturier et al. 2008). An additional explanation is higher percentage of type I muscle fiber in boys (Jansson 1996, Lexell et al. 1992) and possibly, higher utilization of type I muscle fibers (Dotan et al. 2012).

Substrate utilization can be manipulated by ingestion of CHO before or during exercise. The oxidation of CHO_{exo} can be measured by using isotope techniques (Costill et al. 1973; Jeukendrup and Jentjens 2000). Costill et al. (1973) were probably the first to investigate the oxidation of CHO_{exo} in adults. Seven adult males ingested a glucose drink tagged with uniformly labeled glucose ^{14}C glucose (unstable isotope) at rest and during prolonged exercise at 60 to 72% VO_{2peak}. They showed that only a small portion of CHO feeding was oxidized through the exercise. Currently, researchers suggested this finding may be due to methodological limitations since several studies have shown important contributions of ingested CHO to energy expenditure during exercise (Jeukendrup and Jentjens 2000).

Table 1.3 (see Appendix C) shows comparisons on the effects of exogenous carbohydrate on substrate utilization during exercise in thermoneutral conditions among different stages of biological maturation. Timmons et al. (2003a) used ^{13}C stable isotope methodology to compare substrate utilization between boys and men during exercise. Boys and men exercised on two occasions at 70% of their individual VO_{2peak} for two 30-min periods. In the CHO trial, participants ingested a 6% CHO-electrolyte solution (4% sucrose + 2% glucose) and in the placebo trial, an artificially sweetened beverage. In the CHO trial, RER was lower in boys than men and decreased in men but remained constant in boys. In both trials, average CHO_{total} oxidation was lower and Fat_{total} oxidation was higher in boys compared to men. CHO_{total} oxidation tending to be lower (P = 0.07) and Fat_{total} oxidation tending to be higher (P = 0.08) in the placebo compared to the CHO trial. In conclusion, this study showed that pre- and early pubertal boys demonstrate a greater reliance on CHO_{exo} for energy compared to men. In addition, boys oxidized ~70% more fat and ~ 23% less CHO during exogenous CHO feeding before and during exercise. The authors speculated that children may have a relatively higher endogenous fat oxidation due to higher intramuscular triglyceride availability compared with adults. Another hypothesis, although less likely, is that the higher fat oxidation is related to a default mechanism caused by underdeveloped glycogenolytic or glycolytic systems. It is also possible that the rate of intestinal absorption of ingested CHO_{exo} might be greater in boys, resulting in more glucose being delivered to the muscles.

Other studies comparing girls (Timmons et al. 2007a) and boys (Timmons et al. 2007b) during prolonged exercise determined the impact of maturational status on CHO_{exo} oxidation and endogenous substrate utilization. Timmons et al. (2007b) evaluated 12-year-old boys and nine 14-year-old boys cycling for 60 min at 70% VO_{2peak} on two different occasions: with (CHO trial) and without (placebo trial) ^{13}C-enriched CHO intake. There was a significant group effect for RER with higher values in the older group. RER was also higher in CHO trial compared to the placebo trial. Older boys had lower FAT_{endo} oxidation compared to younger boys. CHO_{total} tend to be higher in older boys compared to younger, and was higher in CHO trial compared to placebo trial. No statistical significance was found in the CHO_{exo} oxidation in younger compared to older boy. These data do not support the idea of under-developed glycolytic flux in children. Timmons et al. (2007a) also verified the contribution of CHO_{exo} on metabolism during exercise in twelve preadolescent and ten adolescent girls. Girls performed the same 2 x 30min cycling exercise described above at 70% of their power output with or without CHO intake. RER values were lower during the placebo than CHO trial in younger girls, but were similar between trials in older girls. Fat oxidation was higher in the placebo compared to CHO trial in younger girls, but remained similar in older girls. CHO_{total} was lower in the placebo in young but not in older girls. The ratio between CHO_{exo} and CHO_{endo} was similar between younger and older girls. These data confirm higher fat and lower CHO_{endo} oxidation rates during exercise in young children compared to adolescents. However, these findings contrast with previous studies regarding of age-related differences in CHO_{exo} oxidation in males (Timmons et al. 2003) and suggest that in females there are no differences in CHO_{exo} reliance between young and older girls (Timmons et al. 2007) . According to these results, the authors do not believe that children possess an underdeveloped glycolytic capacity, as was previously proposed by Eriksson (1972). In summary, the available literature suggests that children have greater fat oxidation compared to adults during exercise. Pre-pubertal have greater reliance of CHO_{exo}, and consequently reduced reliance on fat metabolism compared with latepubertal boys (Riddell et al. 2000; Riddell et al. 2001; Timmons et al. 2007a; Timmons et al. 2007b). It has been suggested that both children and adolescents have higher rates of CHO_{exo} oxidation compared to adults; however, the optimal carbohydrate supplementation to sustain endurance performance during childhood and adolescence is unknown. Additionally, there is a lack of studies comparing the effects of CHO_{exo} on substrate utilization during exercise in the heat between boys and men. Given that both environmental conditions and biological maturation seem to affect substrate metabolism, additional studies are needed to understand these responses.

1.5 GENERAL PURPOSE, SPECIFIC PURPOSE AND HYPOTHESIS

1.5.1 General Purpose

The purpose of this thesis was to compare the thermoregulatory responses, the effects of hypohydration on muscle performance, and the substrate utilization between boys and men exercising in the heat.

1.5.2 Specific Purpose

Chapter 2. To compare the thermoregulatory and perceptual responses of boys and men exercising in the heat at two different exercise intensities prescribed to elicit a fixed metabolic heat production ($\dot{H}_p$) per unit body mass and a fixed absolute $\dot{H}_p$.

Chapter 3. To compare the effect of hypohydration, euhydration with and without carbohydrate intake during exercise in the heat on subsequent strength performance of exercised and non-exercised muscles in boys and men. Specifically, we observed the effects of: (1) 2% hypohydration (HY), (2) euhydration with a carbohydrate solution (EU-CHO) and (3) euhydration with flavoured water (EU-W), on isometric and isokinetic muscle strength performance tests of knee and elbow extensors and flexors following exercise in the heat.

Chapter 4*.* To compare the effects of CHO_{exo} on energy yield between boys and men exercising in the heat at the same metabolic heat production per unit body mass. Specifically, we examined total CHO (CHO_{total}), exogenous CHO (CHO_{exo}), endogenous CHO (CHO_{endo}), and total fat (Fat_{total}) oxidation rates during exercise in the heat.

1.5.3 Hypothesis

Chapter 2. We hypothesized that the use of similar metabolic heat production ($\dot{H}_p$) per unit BM would lead to similar changes in rectal temperature (ΔT_{re}) and perceptual responses in boys and men. Secondarily, we hypothesized that exercise at a similar absolute $\dot{H}_p$ would induce a similar sweat volume in boys and men.

Chapter 3. We hypothesized that a reduction in muscle strength due to hypohydration would be greater in boys compared to men both in legs (exercised) and arms (non-exercised) muscles, and that CHO intake would have no effect on muscle strength in the euhydration trial in both boys and men.

Chapter 4*.* We hypothesized that providing exogenous carbohydrate (CHO_{exo}) during exercise in the heat would lead boys and men to oxidize less fat and more carbohydrate. We also hypothesized that boys would use relatively more CHO_{exo} as energy during exercise in the heat compared to men.

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CHAPTER 2: THERMOREGULATION IN BOYS AND MEN EXERCISING AT THE SAME HEAT PRODUCTION PER UNIT BODY MASS

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1. Child Health & Exercise Medicine Program, Department of Pediatrics, McMaster University, Hamilton, ON, Canada.
2. Federal University of Rio Grande do Sul, Porto Alegre, RS, Brazil.

Author contributions: B.W.T. and G.T.L conceptualized and designed the research project; G.T.L acquired the data with assistance from G.S.C, J.O. and B.W.; G.T.L conducted the statistical analysis; G.T.L interpreted results of experiments with assistance from F.M., G.S.C, B.W. and B.W.T; G.T.L wrote the final manuscript with manuscript revisions from all authors. All authors reviewed and agreed upon the final manuscript.

Running head: Thermoregulation in boys and men

ABBREVIATIONS

ANOVA	Analysis of variance
$\dot{H}p$	Metabolic heat production
$\dot{V}O_{2peak}$	Peak oxygen uptake
BM	Body mass
BSA	Body surface area
BSA_r	Effective radiating area of the body
C	Convective heat loss
C_{res}	Respiratory losses through convection
E_{req}	Required evaporative loss for heat balance
E_{res}	Respiratory losses through evaporation
h_c	Convective heat transfer coefficient
HR	Heart rate
M	Rate of metabolic energy expenditure
MEN_{ABS}	Men cycling at a fixed $\dot{H}p$
MEN_{REL}	Men cycling at a fixed $\dot{H}p$ per kg body mass
R	Radiative heat loss
RER	Respiratory exchange ratio
RPE	Rate of perceived exertion
USG	Urine specific gravity

2.1 ABSTRACT

Purpose: Child-adult thermoregulatory comparisons may be biased by differences in metabolic heat production ($\dot{H}_p$). We compared thermoregulatory responses of boys and men exercising at two intensities prescribed to elicit either a fixed $\dot{H}_p$ per unit body mass (BM) or a fixed absolute $\dot{H}_p$. ***Methods:*** Ten boys (10-12 yrs) and 10 men (19-25 yrs) performed 4×20-min cycling at a fixed $\dot{H}_p$ per BM ($W \cdot kg^{-1}$) at 35℃ and 35% relative humidity (MEN_{REL}). Men also cycled (MEN_{ABS}) at the same absolute $\dot{H}_p$ (in W) as the boys. ***Results:*** $\dot{H}_p$ was lower in boys compared with MEN_{REL}, but similar to MEN_{ABS} (mean ± SD, 233.6 ± 38.4, 396.5 ± 72.3, 233.6 ± 34.1 W, respectively, P<0.001). Conversely, $\dot{H}_p$ per unit BM was similar between boys and MEN_{REL}, and lower in MEN_{ABS} (5.7 ± 1.0, 5.6 ± 0.8 and 3.3 ± 0.3 $W \cdot kg^{-1}$, respectively; P<0.001). The change in rectal temperature was similar between boys and MEN_{REL} (0.6 ± 0.2 vs. 0.7 ± 0.2 ℃, P=0.92) but was lower in MEN_{ABS} (0.3 ± 0.2 ℃, P=0.004). Sweat volume was lower in boys compared to MEN_{ABS} (500 ± 173 vs. 710 ± 150 mL; P=0.041), despite the same evaporative heat balance requirement (E_{req}) (199.1 ± 34.2 vs. 201.0 ± 32.7 W, P=0.87). ***Conclusion:*** Boys and men demonstrated similar thermoregulatory responses to 80 min of exercise in the heat performed at a fixed $\dot{H}_p$ per unit BM. Sweat volume was lower in boys compared to men, despite similarities in absolute $\dot{H}_p$ and E_{req}.

Keywords: Temperature regulation · Dehydration · Sweating · Children · Exercise

2.2 INTRODUCTION

Responses to exercise in the heat are influenced by a number of factors, some of which have been used to explain child-adult differences in thermoregulation. Morphological and physiological characteristics of the pediatric population include lower sweat rate, higher body surface area (BSA) to body mass (BM) ratio, slower acclimatization to the heat, and reduced cardiac output at a given metabolic rate compared to adults (Bar-Or et al. 1980a; Bar-Or and Rowland 2004; Falk et al. 1992a; Inoue et al. 2004; Meyer et al. 1992). Such features may also be associated with differences in perceived exertion (Bar-Or and Inbar 1977) and lower tolerance of children exercising in the heat (Drinkwater et al. 1977a). These observations have led to the common belief that youth are at a greater risk for exercise-induced heat illness and exercise intolerance in hot environments. However, it must be noted that the outcomes of these early studies may have been biased by the use of exercise intensities that failed to elicit a fixed metabolic heat production ($\dot{H}_p$) (Cramer and Jay 2014).

To date, most comparisons of children and adults exercising in the heat have employed similar relative exercise intensities (% $\dot{V}O_{2peak}$) (Bar-Or et al. 1980a; Inbar et al. 2004), and have not considered the impact of differential $\dot{H}_p$ on thermoregulatory responses (Jay et al. 2011). In fact, exercise at the same absolute rate of $\dot{H}_p$, but different relative intensities (% $\dot{V}O_{2peak}$), in two groups of adults matched by BM and BSA led to similar changes in body core temperature and whole body sweat loss (Jay et al. 2011). Similar results were observed when comparing thermoregulatory responses during exercise at a fixed $\dot{H}_p$ per unit BM when groups of adults were not matched for BM and BSA (Cramer and Jay 2014).

A diminished sweat rate has been described as a distinguished thermoregulatory response between boys and men when they exercised at a fixed % $\dot{V}O_{2peak}$ in the heat (Falk et al. 1992a; Inbar et al. 2004; Meyer et al. 1992; Shibasaki et al. 1997). In these studies however, the required evaporation for heat balance (E_{req}) does not seem to be considered. In male adults (Gagnon et al. 2013) the E_{req} (in W) was the only variable which independently explained the variation in end-exercise evaporative loss. The % $\dot{V}O_{2peak}$ did not independently contribute to the variation in end-exercise evaporative heat loss, and only described a very small (<2%) amount of variability in whole-body sweat rate. An experimental design that elicited the same absolute E_{req} would serve to clarify differences in sweating responses despite age and maturational factors. Because evaporation of sweat is the primary avenue for heat loss during exercise in the heat and a fixed absolute $\dot{H}_p$ is the primary determinant of E_{req}, the use of exercise intensity protocols based on these variables enables unbiased comparison in sweat responses between boys and men.

To the best of our knowledge, no study has yet compared boys and men exercising at an intensity prescribed to elicit a fixed $\dot{H}_p$ per BM, and a fixed absolute $\dot{H}_p$. Therefore, the selection of an appropriate exercise intensity to compare thermoregulatory responses in children and adults is important to allow appropriate comparisons in thermoregulatory responses. The purpose of this study was to compare the thermoregulatory and perceptual responses of boys and men exercising in the heat at two different exercise intensities prescribed to elicit a fixed $\dot{H}_p$ per unit BM and a fixed absolute $\dot{H}_p$. It was hypothesized that the use of similar $\dot{H}_p$ per unit BM would lead to similar ΔT_{re} in boys and men. We also hypothesized that exercise at the same absolute $\dot{H}_p$ per unit BM would induce similar ΔT_{re} and and perceptual responses in boys and men. Secondarily, we hypothesized that exercise at a similar absolute $\dot{H}_p$ would induce a similar sweat volume in boys and men.

2.3 METHODS

Participants

Ten healthy, active boys (10- to 12-years-old) and ten healthy, active men (19- to 25-years-old) were recruited from the local community and the student community at McMaster University, respectively. Participants were matched for relative $\dot{V}O_{2peak}$ (within 5%) and % body fat (within 10%). Sample size was calculated with 95% statistical power and a 5% significance level based on boys' and men's ΔT_{re} during exercise in the heat, according to Armstrong and Maresh (1995). Participants' physical characteristics are presented in Table 1. Participants were not heat acclimatized as data collection took place during the winter and spring in Ontario, Canada; they reported no medical conditions and were not taking any medications at the time of participation. Boys and their guardians, and men, were informed of the experimental protocol and potential risks and provided written informed assent, where appropriate, and/or consent prior to participation in this study, which was approved by the Hamilton Integrated Research Ethics Board.

Preliminary session

Each participant attended a preliminary session where a physical activity questionnaire (Bar-Or and Rowland 2004) was used to confirm similar activity levels between boys and men. Biological maturation was determined using self-assessed Tanner staging (Matsudo and Matsudo 1994). Standing height, BM (BWB-800, Tanita Corporation, IL) were assessed in all participants; sitting height was also measured in boys to calculate age at peak height velocity (Mirwald et al. 2002). BSA was calculated from the measurements of body height and mass (Dubois and Dubois 1916). Body composition was measured via Dual-energy X-ray absorptiometry (Hologic QDR 4500A scanner, Hologic Inc., Waltham, MA).

To determine $\dot{V}O_{2peak}$, an incremental exercise test was performed in a thermoneutral room on a calibrated, electromagnetically-braked cycle ergometer (*Lode Corival*, Groningen, the Netherlands) using the the McMaster All-Out Progressive Continuous Cycling protocol, as previously described (Bar-Or and Rowland 2004). Measurements of expired O_2 and CO_2 were made continuously using a calibrated metabolic cart (Vmax29, SensorMedics, Mixing Chamber method, Yorba Linda, CA, U.S.A.). Peak oxygen consumption ($\dot{V}O_{2peak}$) was defined as the highest 30-s oxygen uptake value. The test was terminated when 2 of the 4 following criteria were achieved: 1) inability to maintain a cycling cadence above 60 rpm, despite strong verbal encouragement; 2) heart rate (HR) >195 beats·min^{-1}; 3) rate of perceived exertion (RPE) >19; and 4) respiratory exchange ratio (RER) >1.0.

Experimental Trial

Boys performed one experimental trial at a fixed $\dot{H}_p$ per unit BM. Men performed two experimental trials (separated by 4-10 days), in a counterbalanced fashion; trials were identical with the exception of cycling intensity, which consisted of cycling at either 1) a fixed $\dot{H}_p$ per unit BM (MEN_{REL}), or 2) the same absolute $\dot{H}_p$ as their fitness-matched boy (MEN_{ABS}). Prior to the experimental trials, participants were asked to maintain their eating habits, and to avoid any strenuous physical activity, caffeine and alcohol (adults). Participants were prescribed a 3-day hydration protocol before the experimental visit of 12 mL·kg^{-1} of water per day, in addition to their usual intake. On the morning of the experimental trials, participants ingested 6 mL·kg^{-1} of water. No fluids were provided during exercise in the heat. A standardized lunch was consumed 3-4 hours prior to the exercise.

All experimental trials were scheduled in the afternoon. Upon arrival to the laboratory, hydration status was verified from an initial urine sample analyzed for urine specific gravity (USG) (Atago refractometer, 2722-E04; Tokyo, Japan) and color (Armstrong et al. 1994). A USG cutoff of 1.025 was applied as the need for additional hydration prior to exercise. This was followed by a measurement of nude weight. Participants performed the exercise in athletic shorts and shoes only. Rectal temperature (T_{re}) was measured using a flexible thermometer (YSI 400 series thermistor, USA). Skin temperature (T_{sk}) was measured using skin thermistors (YSI 400 series temperature sensors, USA), placed on the chest, upper back, and thigh.

Participants received standardized instructions on how to answer the following four perceptual evaluations: RPE using Borg scale (Borg and Dahlstrom 1962); thermal sensation (9-point scale from “very cold” to “very hot”) and comfort (6-point scale from “very comfortable” to “very uncomfortable”) (Arens et al. 2006); and intensity of thirst (9-point scale from “not thirsty” to “very thirsty”) (Maresh et al. 2001).

Prior to exercise, participants rested in a seated position for 5 min in the climatic chamber, which was set to 35˚C and 35% relative humidity (for figure of experimental design see Appendix E). Room temperature was verified with a wet-bulb globe temperature (3M QUESTemp® QT34 Wet Bulb Globe Temperature Heat Stress Monitors, Illinois, USA) and air velocity using an anemometer. The exercise protocol consisted of 80 min of cycling divided into 4×20 min bouts, with a 10-min rest in between bouts, at a fixed $\dot{H}_p$ per BM ($W \cdot kg^{-1}$). Men performed an additional session at the same $\dot{H}_p$ (in W) as the boys. $\dot{V}O_2$ was measured in the last 5 min of each cycling bout. T_{re}, T_{sk}, HR and RPE were recorded every 5 min. Thermal sensation, thermal comfort, and intensity of thirst were recorded at the beginning, mid-, and in the last min of each cycling bout. Cycling was terminated if two of the following events occurred simultaneously: T_{re} > 39°C, HR ≥ 200 beats·min^{-1}, RPE >19, participant displayed symptoms of heat exhaustion (nausea, disorientation, headache, and dizziness), and/or an inability to maintain a cycling cadence of 60 to 80 rpm.

After each bout, participants were asked to void their bladder, and weight was measured with their shorts, shoes, and electrodes to determine cumulative body weight changes, which were corrected to the pre-exercise nude BM. Change in body weight was used as the indicator of dehydration. Upon completion of the exercise, participants were instructed to void their bladder and dry the sweat on their body, and nude body weight was measured to calculate sweat volume (ΔBM before and after cycling and corrected for urine loss and vapor losses from the respiratory tract).

Heat exchange parameters

The rate of metabolic energy expenditure (M; in W·m^{-2}) was estimated using the average of VO_2 (L·min^{-1}) and RER measured during the experimental trials, and calculated as (Nishi 1981):

$$M = \dot{V}O_2 \cdot \frac{\left[\left(\frac{RER - 0.7}{0.3}\right) \cdot e_c\right] + \left[\left(\frac{1.0 - RER}{0.3}\right) \cdot e_f\right]}{60 \cdot BSA} \cdot 1000$$

where e_c is the caloric equivalent per liter of oxygen for the oxidation of carbohydrates (21.13 kJ), and e_f is the oxidation of fat (19.62 kJ). $\dot{H}_p$ (in W·m^{-2}) was calculated as the difference between *M* and the external work rate (W).

$$\dot{H}_p = M - W$$

Sensible heat loss at the skin (C + R, in W·m^{-2}) was calculated as the combination of convective heat loss exchange from skin (C) and heat loss via radiation (R) (Kerslake 1972). Convective heat loss exchange from skin (C, in W·m^{-2}) was calculated as:

$$C = h_c \cdot (T_{sk} - T_a)$$

where h_c is the convective heat transfer coefficient (in W·m^{-2}·K^{-1}) (Mitchell 1974), determined by $8.3 \cdot v^{0.6}$, here v equals air velocity in m·s^{-1}. Heat loss via radiation (R, in W·m^{-2}) was calculated as:

$$R = h_r \cdot (T_{sk} - T_a)$$

where T_a is the ambient temperature (in ℃), and h_r is the radiant heat transfer coefficient ($W \cdot m^{-2} \cdot K^{-1}$) calculated as:

$$h_r = 4 \cdot \varepsilon \cdot \sigma \cdot (BSA_r / BSA) \cdot [(T_{sk} + T_r)/2 + 273.15]^3$$

where ε denotes the emissivity of the skin (0.95), σ is the Stefan-Boltzmann constant ($5.67 \cdot 10^{-8}$ $W \cdot m^{-2} \cdot K^{-4}$); BSA_r/BSA is the effective radiating area of the body (0.72 since participants were seated (ISO 7933/2004)), and T_r is mean radiant temperature in °C (assumed to be equal to T_a).

Respiratory losses through evaporation (E_{res}) and convection (C_{res}) (in $W \cdot m^{-2}$) were determined by:

$$E_{res} + C_{res} = 0.173 \cdot (\dot{H}_p) \cdot (5.87 - P_a) + 0.0014 \cdot (\dot{H}_p) \cdot (34 - T_a)$$

where P_a is the ambient vapor pressure (kP_a).

The required evaporative heat loss per unit area for heat balance (E_{req}, in W) was calculated as:

$$E_{req} = \dot{H}_p - (C + R + C_{res} + E_{res})$$

Statistical analyses

Paired student *t*-tests were used for the mean participant characteristics comparisons and matched pair comparisons (boys vs. men) (i.e. heat exchange parameters, 80-min ΔT_{re}, 80-min cumulative whole body sweat volume). Condition-specific 2-way ANOVAs were used to compare the groups over time (i.e T_{re}, T_{sk}, HR, RPE, thermal sensation, thermal comfort and intensity of thirsty, cumulative weight loss, USG and color every rest period). Bonferroni post-hoc analyses were used to examine significant interactions (group × time). One-way repeated-measures ANOVAs were performed to analyse cumulative weight loss, USG, and urine colour throughout each trial, with Bonferroni post hoc tests when necessary. No comparisons were performed across conditions in men (i.e., MEN_{REL} was not compared to MEN_{ABS}). All data are expressed as means and standard deviations (SD). Statistical significance was set at $\alpha \leq 0.05$, and all analyses were performed using GraphPad Prism (version. 4.0, GraphPad Software, La Jolla, CA).

2.4 RESULTS

In the present study, boys performed one experimental trial at a fixed $\dot{H}_p$ per unit BM. Men performed two experimental trials cycling at either 1) a fixed $\dot{H}_p$ per unit BM (MEN_{REL}), or the same absolute $\dot{H}_p$ as their fitness-matched boy (MEN_{ABS}). Mean environmental conditions were similar for all experimental trials for boys, MEN_{REL} and MEN_{ABS}. Exercise intensity as % VO_{2peak} was similar between boys and MEN_{REL}, but lower in MEN_{ABS} (47.9 ± 5.0, 48.7 ± 1.8 and 27.3 ± 5.0, respectively; P < 0.001). Conversely, average workload was lower in boys vs. MEN_{REL}, but similar to MEN_{ABS} (31.7 ± 9.4, 86.1 ± 22.9, and 35.3 ± 7.0 W, respectively; P <0.001). Mechanical efficiency, defined as the ability of an individual to transfer energy consumed by external work, was lower in boys vs. MEN_{REL}, but similar to MEN_{ABS} (13.0, 17.7, and 11.7 %, respectively, P < 0.001).

Heat Exchange Parameters

Table 2 presents means heat exchange values during the experimental trials. On average, $\dot{H}_p$ was 41% lower in boys compared to MEN_{REL}, and by design, similar in boys vs. MEN_{ABS}. Conversely, when normalized to BM, $\dot{H}_p$ was similar between boys and MEN_{REL}, and lower in MEN_{ABS}. (R+C) was similar in boys, MEN_{REL}, and MEN_{ABS}; however, (R+C) normalized by BSA was greater in boys compared to MEN_{ABS}. E_{req} was lower in boys and compared to MEN_{REL}, but was similar in boys and men MEN_{ABS}.

Rectal Temperature, Skin Temperature and Heart Rate

Figure 1 depicts T_{re}, T_{sk} and HR over the 4×20-min bouts of exercise. Initial T_{re} was similar among boys, MEN_{REL} and MEN_{ABS} (37.6 ± 0.2, 37.4 ± 0.2 and 37.4 ± 0.2 ˚C, respectively). The pattern of T_{re} was similar in boys and MEN_{REL} during exercise ($F(1,19) = 1.1$; $P = 0.32$). A group × time interaction was seen for boys vs. MEN_{ABS} ($F(1,19) = 7.1$; $P < 0.001$), whereby T_{re} was greater in boys vs. MEN_{ABS}

from the 35th minute to the end of exercise (P < 0.05). The pattern of average T_{sk} increased similarly between boys and MEN_{REL} throughout the session. Conversely, the magnitude of average T_{sk} change was higher in boys vs. MEN_{ABS}, as indicated by a group × time interaction (F(1,19) = 2.2; P = 0.003). The regional T_{sk} patterns for the back, chest, and thigh were similar in boys and MEN_{REL}, but were higher in boys vs. MEN_{ABS}, for T_{sk} of the chest and thigh (group × time interaction, F(1-19) = 2.05; P = 0.006 and F(1-19) = 2.43, respectively; P <0.001) but not T_{sk} for the back. HR was similar among boys, MEN_{REL} and MEN_{ABS} at the beginning of exercise (93 ± 12, 87 ± 13, and 89 ± 13 beats·min^{-1}, respectively), and increased to a similar degree during exercise in boys vs. MEN_{REL}, but was lower in MEN_{ABS} (group × time interaction F(1-19) = 6.76; P < 0.001).

Body Hydration and Sweating Responses

Table 3 shows body hydration status during the exercise trials. All participants arrived to the experimental trials with similar hydration levels according to mean ± SD values of USG and urine colour. USG and urine colour throughout the session were similar in boys and MEN_{REL} (F(1-4) = 0.18 and F(1-4) = 0.13, respectively, P >0.90), as well as in boys and MEN_{ABS} (F(1-4) = 0.02 and F(1-4) = 0.05, respectively, P >0.90). The cumulative weight loss was similar between boys and MEN_{REL} (F(1-3) = 2.61; P = 0.06), but it was greater in boys compared to MEN_{ABS} (F(1-3) = 4.09; P = 0.011).

On average, sweat volume was 55% lower in boys compared to MEN_{REL}, and 30% lower in boys compared to MEN_{ABS} (500 ± 173, 1122 ± 304 and 710 ± 150 mL, respectively; $P < 0.001$ and $P = 0.041$). When normalized to BSA, the sweat volumes were 38.6% lower in boys compared to MEN_{REL} (371 ± 116 vs. 604 ± 149 mL·m^2, $P = 0.002$), but were similar between boys and MEN_{ABS} (384 ± 78 mL·m^2, $P = 0.81$).

Perceived Responses

Figure 2 shows the results of RPE, thermal sensation, thermal comfort and thirst in boys and men exercising in the heat. Since exercise intensity was not identical for all groups, RPE was normalized to HR (RPE·HR^{-1}). No differences in RPE·HR^{-1} were observed between the boys and MEN_{REL}, but a group × time interaction was found between boys and MEN_{ABS} ($F(1\text{-}15) = 1.88$; $P = 0.025$), indicating a greater perceived effort in the boys. Boys perceived the room to be warmer and less comfortable than MEN_{ABS} ($F(1\text{-}11) = 2.01$; $P = 0.029$ and $F(1\text{-}11) = 2.51$; $P = 0.005$, respectively). The boys' perceived intensity of thirst was similar to that of MEN_{REL} ($F(1\text{-}11) = 1.96$; $P = 0.34$) but higher than MEN_{ABS} ($F(1\text{-}11) = 4.41$; $P < 0.001$) at the end of each exercise bout.

2.5 DISCUSSION

The main finding of this study is that thermoregulatory responses were similar between boys and men during 80 min of exercise in the heat performed at a fixed $\dot{H}_p$ per unit BM (i.e., boys vs. MEN_{REL}). Moreover, the increase in T_{re} was similar in boys and MEN_{REL} (0.6 vs. 0.7^o C, respectively). However, in the condition where men produced the same absolute $\dot{H}_p$ as the boys (MEN_{ABS}), $\dot{H}_p$ normalized by BM was lower in men, resulting in smaller T_{re} increase compared with boys (0.3 vs. 0.6 ˚C). Our results suggest that when $\dot{H}_p$ per unit BM is equal in boys and men, there is no thermoregulatory disadvantage for boys exercising in the heat, despite their marked lower BM, lean BM, height, BSA, age and biological maturation.

In the present study, ΔT_{re} was similar in boys and MEN_{REL} during exercise, but greater in boys compared to $MEN_{ABS.}$ Previous studies have also reported no differences in the rise of T_{re} between children and adults during exercise in the heat, but the exercise was prescribed as $\%VO_{2peak}$ (Inbar et al. 2004; Rivera-Brown et al. 2006; Rowland et al. 2008; Shibasaki et al. 1997), which may have elicited similar $\dot{H}_p$ per unit BM. Indeed, Armstrong and Maresh (1995) compiled data from studies comparing children and adults, and reported an average increase in T_{re} of 1.24 ± 0.4 ˚C and 1.21 ± 0.39 ˚C, respectively. Falk et al. (1992b) compared pre-, mid- and late- pubertal boys cycling at 50% VO_{2peak} during 3×20 min bouts in dry heat condition. ΔT_{re} was similar among groups, with no differences in $\dot{H}_p$ per unit BM, although lower body heat storage was reported in pre- compared to late-pubertal boys. Similarly, an earlier study by Inbar et al. (2004) also found no difference in ΔT_{re} between boys and men cycling in the heat at 50% of $\dot{V}O_{2peak}$; however, the boys demonstrated greater $\dot{H}_p$ per unit BM and lower heat storage compared to men. The use of exercise intensity based on $\dot{H}_p$ normalized to BM, as in the current study, has been suggested to ensure an unbiased comparison of thermoregulatory responses in heterogeneous groups (i.e. adults not matched by body composition (Cramer and Jay 2014) or aerobic fitness (Jay et al. 2011)). Importantly, exercise prescription based on heat exchange parameters with appropriate normalization may also avoid the aforementioned conflicting results reported in the pediatric literature. In the present study, it is unlikely that similarities in ΔT_{re} are due to similarities in % $\dot{V}O_{2peak}$, per se. $\dot{H}_p$ normalized to BM was the same therefore from a biophysical perspective it resulted in similarities in change in T_{re}. To confirm this, future studies should examine thermoregulatory responses in boys and men of different fitness levels exercising at the same $\dot{H}_p$ per unit BM, and in turn, different relative intensities.

Several factors may affect the body's ability to regulate body temperature, including BSA, BSA per BM, total number of heat activated sweat glands, sweat droplet size and their distribution, and the sweating sensitivity (Armstrong and Maresh 1995). Boys in the current study presented with 18% higher BSA normalized to BM compared with men, which may have conferred a thermoregulatory advantage by allowing for more dry heat dissipation from the skin to the environment when T_{sk} exceeded T_a. In fact, dry heat loss in the present study was greater in boys compared to MEN_{ABS} (22.6 vs. 16.5 $W.m^2$). However, this difference likely had negligible effects on our findings as the gradient of T_{sk} and T_a was close to zero.

Sweating is the primary method of heat dissipation during exercise in the heat, and importantly, it is the only effective method when T_a exceeds T_{sk}. Differences in sweating between boys and men may be explained by biophysical factors (i.e. E_{req}). Indeed, compared to MEN_{REL}, absolute sweat volume was 55% lower in boys and E_{req} was 44% lower in boys. Inbar and colleagues (2004) also showed a lower sweat volume in boys compared to men (479 vs. 1172 mL, respectively) and a lower E_{req} (235.1 vs. 488.6 W) during exercise prescribed as % $\dot{V}O_{2peak}$ in a hot and dry condition, with water available to drink *ad libitum*. In adults, young men with vastly different BM and BSA demonstrated similar whole body sweat loss when exercising at a similar absolute E_{req} in a physiologically compensable environment (Jay et al. 2011). These findings suggest that E_{req} may be more influenced by differences in exercise intensity rather than differences in body size.

The use of protocols based on a fixed % $\dot{V}O_{2peak}$ may lead to a different rate of $\dot{H}_p$ between independent groups which are not matched for absolute $\dot{V}O_{2peak}$ (in $L \cdot min^{-1}$), resulting in a greater sweat production to offset the greater $\dot{H}_p$ elicited by the protocol. Actually, Gagnon et al. (2013) showed that % $\dot{V}O_{2peak}$ has a minimal contribution to whole body sweat rate and does not independently contribute to the variation of end-of-exercise sweat production in adult males. On the other hand, E_{req} explained ~ 90% of the variance in whole-body sweat rate. Interestingly, in the present study despite similarities in absolute E_{req} (199.1 vs. 201.0 W) boys showed lower sweat volume compared to MEN_{ABS} (500 vs. 710 mL). The observed lower sweating rate in our boys corroborates previous findings highlighting the effect of biological maturation on sweating rate in males (Falk et al. 1992a; Inbar et al. 2004; Meyer et al. 1992; Rowland et al. 2008). Size of sweat drops (drop area) increases with greater pubertal stage, as well as the sweat rate (Falk et al. 1992a; Meyer et al. 1992). Studies suggested that these maturation-related effects reflect hormonal differences as well as underdeveloped peripheral sweating mechanisms in children rather than any impairment of central-driven sudomotor function (Rowland 2008; Sato et al. 1987). Even though differences in sweating rate were found between boys and men in the present study it seems that the greater volume was not an advantage for the men considering the level of hypohydration and experimental conditions. Future studies should compare children and adults exercising at intensities based on the E_{req} in different environmental conditions (i.e. physiologically compensable environment) so as to clarify the effects of growth, maturation, exercise intensity and environment on sweating responses.

Previous studies have compared perceptual responses between children and adults during exercise in the heat (Drinkwater et al. 1977a; Meyer et al. 1994; Passe et al. 2007), but to our knowledge, no studies have compared child-adult perceptual responses to exercise in the heat at the same relative and absolute intensities. In our study, $RPE \cdot HR^{-1}$ was similar in all groups. The perceived thermal sensation and thermal discomfort were higher for the boys compared to MEN_{ABS}. Drinkwater et al. (1977a) observed a higher occurrence of heat-acclimatized pre-menarcheal girls than young women stopping exercise in the heat because they "felt it was too hot" (five vs. one); thermal perception was also higher at the point of fatigue in girls compared to women. The authors suggested that this might serve as a protective mechanism preventing girls from experiencing exertion-related heat illness. In the present study, thirst intensity was greater in boys compared to men at both exercise intensities even though hypohydration levels were similar in all groups. It is interesting to note that in adults, thirst alone is insufficient at completely preventing significant dehydration during prolonged exercise in the heat (Passe et al. 2007), while in children, thirst can trigger sufficient voluntary fluid consumption to replace sweat losses (Meyer et al. 1994).

Regardless of age, body size and biological maturation, in the current study boys and men demonstrated similar thermoregulatory and perceptual responses to 80 min of exercise in the heat performed at a fixed metabolic heat production per unit BM. Sweat volume was lower in boys compared to MEN_{ABS} despite the same absolute E_{req}.

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Conflict of interest

None of the authors had a conflict of interest regarding any aspect of this research.

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Table 2.1 Physical and physiological characteristics of boys and men.

Characteristics	Boys (n = 10)	Men (n = 10)
Age (years)	11.5 ± 1.3	22.9 ± 2.3*
Body mass (kg)	42.3 ± 9.7	71.1 ± 9.7*
Body height (cm)	152.1 ± 1.2	174.5 ± 5.9*
Body fat (%)	17.4 ± 4.8	16.7 ± 4.6
Total muscle mass (kg)	31.6 ± 5.8	58.5 ± 8.9*
Body surface area (BSA) (m^2)	1.34 ± 0.20	1.85 ± 0.14*
BSA per BM ($m^2 \cdot kg^{-1}$)	0.0322 ± 0.0024	0.0263 ± 0.0015*
VO_{2peak} ($mL \cdot min^{-1}$)	1641.2 ± 352.5	2875.7 ± 640.4*
VO_{2peak} ($mL \cdot kg^{-1} \cdot min^{-1}$)	39.6 ± 7.1	40.4 ± 6.5
Heart $Rate_{max}$ ($beats \cdot min^{-1}$)	187 ± 12	185 ± 9
$Workload_{max}$ (W)	112.8 ± 19.6	240.0 ± 47.4*
Biological maturation		
Tanner stage 1/2/3, n	4/2/4	–
Years from peak high velocity	-2.7 ± 0.9	–

Tanner stage 1 = prepubertal; tanner stages 2 and 3 = early pubertal. Values are expressed in mean ± SD. *$P < 0.05$.

Table 2.2 Average heat exchange values during exercise in the heat.

	Boys	MEN_{REL}	MEN_{ABS}
$\dot{H}_p$ (W)	233.6 ± 38.4	396.5 ± 72.3*	233.6 ± 34.1
$\dot{H}_p$ /mass (W·kg^{-1})	5.7 ± 1.0	5.6 ± 0.8	3.3 ± 0.3#
$\dot{H}_p$ / BSA (W·m^{-2})	175.2 ± 24.3	213.3 ± 30.0*	125.5 ± 14.2#
(C + R) (W)	29.9 ± 4.7	36.1 ± 11.7	30.5 ± 8.5
(C + R) / BSA (W·m^2)	22.6 ± 3.7	19.3 ± 5.4	16.5 ± 4.4#
E_{req} (W)	199.1 ± 34.2	352.7 ± 63.9*	201.0 ± 32.7
E_{req} / BSA (W·m^2)	149.3 ± 21.0	189.8 ± 27.1*	108.9 ± 15.4#

Values are expressed as mean ± SD. * $P < 0.05$ boys vs. MEN_{REL}; # $P < 0.05$ boys vs. MEN_{ABS}. $\dot{H}_p$: metabolic heat production; BSA: body surface area; C: convective heat loss; R: radiative heat loss; E_{req}:. required evaporative loss for heat balance.

Table 2.3 Hydration status during exercise in the heat in boys and men.

Groups	Prior to exercise	Bout 1	Bout 2	Bout 3	Bout 4	*P*
Cumulative body weight loss (%)						
Boys	-	0.3 ± 0.2	0.6 ± 0.3[b]	0.9 ± 0.4[b,c]	1.6 ± 0.4[b,c,d]	<0.0001
MEN_{REL}	-	0.5 ± 0.1	1.0 ± 0.2[b]	1.4 ± 0.4[b,c, *]	1.9 ± 0.4[b,c,d]	<0.0001
MEN_{ABS}	-	0.4 ± 0.2	0.7 ± 0.2[b]	1.0 ± 0.2[b,c]	1.3 ± 0.3[b,c,d]	<0.0001
Urine Specific Gravity						
Boys	1.018±0.008	1.020±0.011	1.027±0.005[a]	1.028±0.007[a,b]	1.029± 0.004[a,b]	<0.0001
MEN_{REL}	1.015±0.006	1.018±0.007	1.022±0.005[a,b]	1.024±0.004[a,b]	1.027±0.004[a,b,c]	<0.0001
MEN_{ABS}	1.012±0.005	1.015±0.006	1.019±0.006[a,b]	1.021±0.005[a,b]	1.024±0.004[a,b]	<0.0001
Urine colour						
Boys	2.1 ± 0.9	2.2 ± 1.0[a]	2.9 ± 1.0	3.3 ± 0.7[a,b]	3.5 ± 1.0[a,b,c]	<0.0001
MEN_{REL}	2.1 ± 0.9	2.4 ± 1.1	3.0 ± 1.1	3.3 ± 0.9	3.6 ± 0.9[a,b]	<0.0001
MEN_{ABS}	1.8 ± 0.9	1.8 ± 1.1	2.3 ± 1.0	2.6 ± 0.7[a,b]	3.1 ± 0.8[a,b,d]	<0.0001

Values are expressed as mean ± SD. * $P < 0.05$ boys *vs.* MEN_{REL}. a ≠ from prior to exercise, b ≠ from bout 1, c ≠ from bout 2, d ≠ from bout 3.

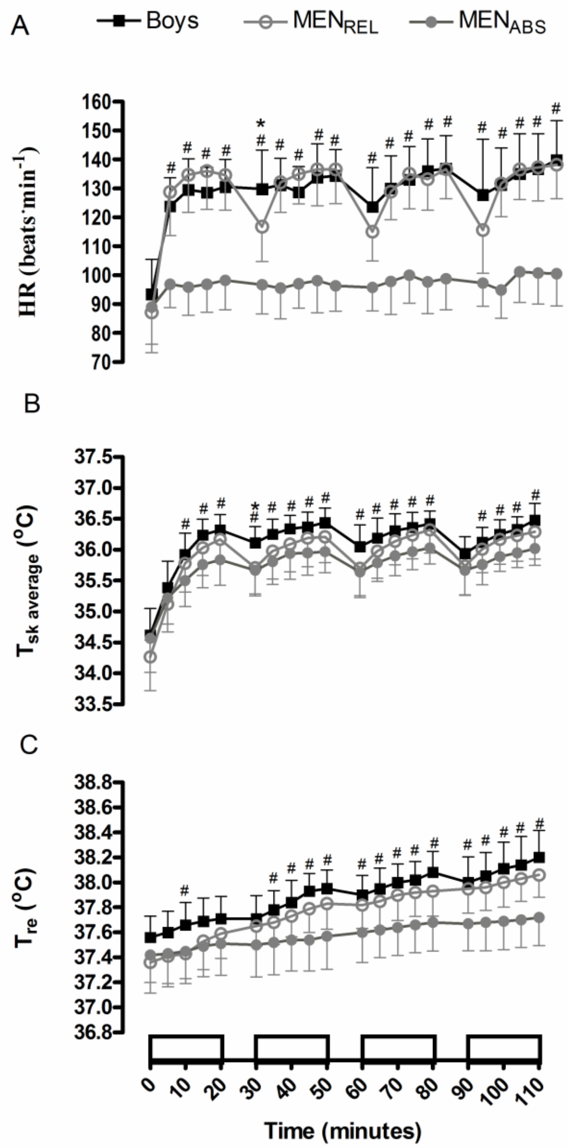
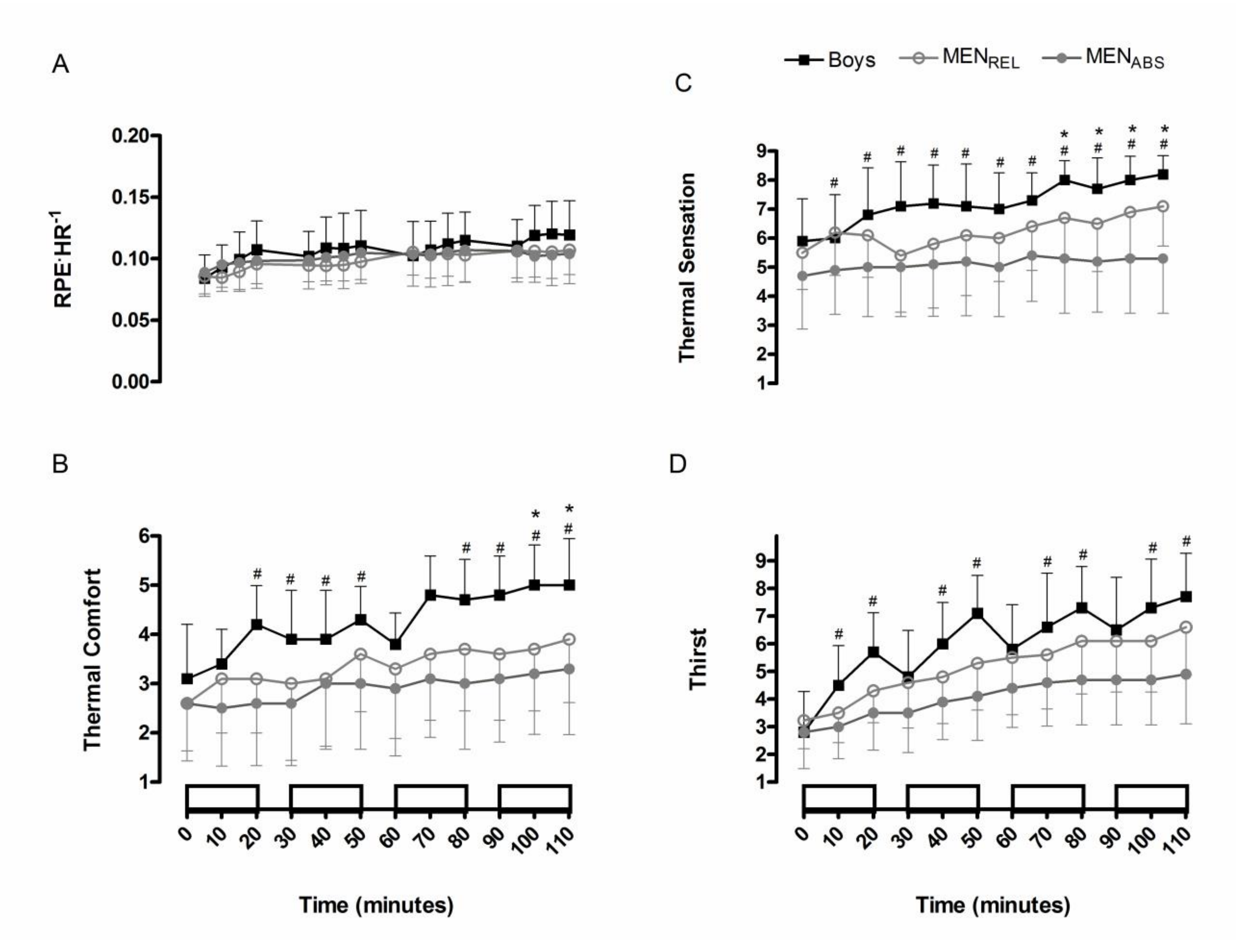


Figure 2.1 A) Heart rate (HR), B) average skin temperature (T_{sk}), C) rectal temperature (T_{re}), D) in boys (■), MEN_{REL} (○) and MEN_{ABS} (●) exercising in the heat. Values are expressed as mean ± SD. # *P* < 0.05 boys vs. MEN_{ABS}.

Figure 2.2 A) Rating of perceived exertion normalized to heart rate (RPE·HR^{-1}), B)



thermal comfort, C) thermal sensation, and D) thirst in boys (■), MEN_{REL} (○) and MEN_{ABS} (●) exercising in the heat. Values are expressed as mean ± SD. * $P < 0.05$ boys vs. MEN_{REL}; # $P < 0.05$ boys vs. MEN_{ABS}.

CHAPTER 3: STRENGTH PERFORMANCE IN BOYS AND MEN: EFFECT OF HYPOHYDRATION AND CARBOHYDRATE INTAKE

Submitted for publication as:

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3. Child Health & Exercise Medicine Program, Department of Pediatrics, McMaster University, Hamilton, ON, Canada.
4. Federal University of Rio Grande do Sul, Porto Alegre, RS, Brazil.

Author contributions: B.W.T. and G.T.L conceptualized and designed the research project; G.T.L acquired the data with assistance from G.S.C, J.O and B.W; G.T.L conducted the statistical analysis; G.T.L interpreted results of experiments with assistance from G.S.C, J.O., F.M., B.W. and B.W.T; G.T.L wrote the final manuscript with manuscript revisions from all authors. All authors reviewed and agreed upon the final manuscript.

Running head: Hypohydration on Muscle Strength

Keywords: Muscle strength. Hydration. Exercise. Heat. Children.

3.1 ABSTRACT

Purpose: To compare the effect of hypohydration (HY), euhydration with (EU-CHO) and without (EU-W) carbohydrate intake on subsequent strength performance of exercised and non-exercised muscles in boys and men. **Methods:** In a randomized counterbalanced order, seven boys (11.2±0.5 yr) and nine men (24.0±3.3 yr) attended three identical sessions, except for experimental beverages. Participants performed 4×20-min bouts of cycling at a fixed metabolic heat production (6 $W \cdot kg^{-1}$) at 38ºC and 50%RH. Experimental beverages were provided in a double-blinded manner. Following one hour of recovery (25ºC and 35%RH), participants performed isometric and isokinetic ($60^{o} \cdot s^{-1}$, $120^{o} \cdot s^{-1}$ and $240^{o} \cdot s^{-1}$) maximal voluntary contractions (MVCs) of knee and elbow extensors and flexors. Participants performed 30 MVCs at $240^{o} \cdot s^{-1}$ and the fatigue index was calculated as the rate of drop from the average of the first 5 MVCs and the 5 last MVCs. **Results:** No differences in strength performance were found among HY, EU-CHO and EU-W in boys or men. Knee peak torque in boys, compared to men, was lower by: 19.0% for isometric extensors in EU-CHO and 17.2 to 21.4% for isometric flexors (depending on trial); 12.1% for extensors in EU-W and 16.8 to 23.9% for flexors at $60^{o} \cdot s^{-1}$; 15.6 to 16.9% for extensors and 20.3 to 23.8% for flexors at $120^{o} \cdot s^{-1}$; 16.0% for extensor in HY and 11.6% in EU-CHO at $240^{o} \cdot s^{-1}$. Elbow peak torque in boys, compared to men, was lower by: 20.6% for flexors at $60^{o} \cdot s^{-1}$; 24.4 to 25.6% for flexors at $120^{o} \cdot s^{-1}$; 16.5% for extensors in HY and 20.9%

for flexors in EU-W. The drop in strength (fatigue index) during 30 repetitions at 240$^{o}\cdot s^{-1}$ in boys, compared to men, was greater by: 27.0 to 47.1% for knee flexors and 21.5 to 29% for elbow flexors. **Conclusion:** Hydration state with or without CHO supplement did not influence strength performance in boys and men. Boys showed lower peak torques and greater fatigability compared to men.

3.2 INTRODUCTION

Muscle strength is a key parameter for successful performance across a variety of sports and fitness programs (1, 18). During sport practices and competitions, players become progressively dehydrated when they fail to properly replenish sweat loss (2-3, 10, 17). Many studies have examined the effects of hypohydration on endurance performance in children (27, 40) and adults (12-13, 35-36) and reported consistent detriments in outcomes such as heart rate, body core temperature and effort and thirst perceptions (36). As a result, supplementary fluid intake is used to avoid the adverse effects of hypohydration and early fatigue during prolonged exercise (10). Conversely, fewer studies have assessed the impact of hypohydration on strength performance and the results in adults are a mix of either detrimental (21, 23) or no effect (8, 34). Such inconsistent results may be due to the exercise/heat protocol used to induce hypohydration, and the associated confounding variables that may affect strength (9, 26) such as increased core temperature, muscle fatigue, baseline hydration levels, and caloric restriction. The presence of carbohydrate (CHO) in the beverage ingested in the euhydration but not the hypohydration trial may also be an important consideration, given that both fluid and CHO equally improve cycling performance, and demonstrate additive effects in adults (6). Isolating the effects of hypohydration would allow for a clearer understanding of its consequences on muscle strength both in children and adults.

Currently, the effect of hydration status on neuromuscular function, and consequently muscle strength performance, is unclear. The most widely accepted theory is that hypohydration could affect some neuromuscular component that impairs the ability of the central nervous system to stimulate the musculature or muscle membrane excitability (26). Maximal muscle strength is generally lower in children compared to adults, even after normalization for body mass (15) or muscle cross-sectional area (20). This difference could be related to neuromotor maturation. It is therefore possible that children's strength performance is impaired by hypohydration to a greater extent than adults

Drinks containing CHO in the form of spo-rts drinks are used to offset the deleterious effects of hypohydration before and during prolonged exercise (25, 29). The effects of CHO consumption on strength performance are controversial, with evidence to suggest that CHO supplementation might increase (38) or have no effect (19) on strength in adults. Primary mechanisms related to maintenance of high rate of CHO oxidation and increased activation of pleasure and reward centers of the brain (24, 25). The ergogenic effects of CHO (6-8%) may be attributable to the maintenance of a higher CHO oxidation rate during exercise (25). Children may have a greater reliance on CHO ingested during prolonged exercise compared to adults (37); however, exercise in the heat seems to influence CHO oxidation (24), which might in turn affect the ergogenic role of CHO in children and adults.

Exercise in the heat and hypohydration as a result of habitual hypohydration or failure to fully rehydrate from previous exercise may promote fatigue in the exercised muscles, thereby compounding impaired strength performance (21, 23). This may be a concern as many active children and young players are involved in sports characterized by intermittent bouts of exercise that demand strength performance over a long period of time. Moreover, repeated exercise bouts, such as training sessions or same-day rounds of tournament competition, are a common practice in organized youth sports (7, 33). A limited number of studies suggest that hypohydration can impair (39) and CHO can enhance (17) high intensity endurance performance in children. However, no study verified the impact hypohydration and CHO intake before and during exercise on subsequent strength performance. Understanding the effects of hypohydration on strength performance in children and adults will allow for more specific recommendations for the appropriate hydration needs of these groups and help guarantee their optimal physical performance.

The purpose of the present study was to compare the effect of hypohydration (HY), euhydration with (EU-CHO) and without (EU-W) carbohydrate intake during exercise in the heat on subsequent strength performance of exercised and non-exercised muscles in boys and men. Specifically, we observed the effects of: (1) 2% hypohydration (HY), (2) euhydration with a carbohydrate solution (EU- CHO) and (3) euhydration with flavoured water (EU-W), on isometric and isokinetic muscle strength performance tests of knee and elbow extensors and flexors following exercise in the heat. We hypothesized that a reduction in muscle strength due to hypohydration would be greater in boys compared to men both in legs (exercised) and arms (non-exercised) muscles, and that CHO intake would have no effect on muscle strength in the euhydration trial in both boys and men.

3.3 METHODS

Participants. Seven healthy active boys (10 to 12 yr) and nine healthy active men (20 to 30 yr) participated in this study. Sample size was calculated with 90% statistical power and a 5% significance level based on the expected impact of hypohydration on performance in boys according to Wilk et al (2014) and on expected differences between boys and men with respect to the average knee extension isokinetic torque, according to De Ste Croix et al (2009). Participants were not heat acclimatized as data collection took place during the winter months in Ontario. Participants were excluded if they reported any medical conditions, medication use at the time of participation, history of surgery or musculoskeletal injury in lower and upper limbs, and cardiovascular, respiratory or metabolic disease that could affect thermoregulatory responses or strength performance. Boys were recruited from the local community and men were recruited from the student community of McMaster University. Boys and their guardians, and men, were informed of the experimental protocol and potential risks and provided written informed assent, where appropriate, and/or consent prior to participation in this study, which was approved by the Hamilton Integrated Research Ethics Board. Participants involved in this study are the same as in study #3 of this thesis.

Preliminary session. Each participant attended a preliminary session where a questionnaire (5) was used to confirm similar physical activity levels between boys and men. Participants were also asked about their health status. Biological maturation was determined using self-assessed Tanner staging. Standing height and body mass (BM) (BWB-800, Tanita Corporation, IL) were assessed in all participants; sitting height was measured in boys to calculate age at peak height velocity. Body surface area (BSA) was calculated from the measurements of body height and mass. Body composition was measured using Dual-energy X-ray absorptiometry (DXA) (Hologic QDR 4500A scanner, Hologic Inc., Waltham, MA). Lower limb muscle mass (LLMM) and upper limb muscle mass (ULMM) were used to correct maximal peak torque. Additionally, lean body mass (LBM) was used to calculate the total beverage volume consumed by each participant.

Aerobic fitness. To determine peak oxygen uptake ($\dot{V}O_{2peak}$), incremental exercise testing was performed using the McMaster All-Out Progressive Continuous Cycling Test (5). All participants were verbally encouraged to give their best performance. An electronically-braked cycle ergometer (*Lode Excalibur*, Groningen, the Netherlands) was used for all tests. Measurements of O_2 and CO_2 were made continuously using a calibrated metabolic cart (Vmax29, SensorMedics, Mixing Chamber method, Yorba Linda, CA, U.S.A). Peak was considered the highest 30-s oxygen uptake ($\dot{V}O_{2peak}$ value). To gauge the participant's perception of their effort, we asked them to rate their perceived exertion using the Borg 6-20 scale. The test ended when 2 of the 4 following criteria were reached: 1) inability to maintain a cycling cadence above 60 rpm in spite of strong verbal encouragement; 2) heart rate (HR) >195 beats$\cdot$min^{-1}; 3) rate of perceptive exertion (RPE) > 19; and 4) respiratory exchange rate > 1.0.

Familiarization with muscle strength performance tests. The complete test protocol was explained and practiced by the participants to familiarize them with the assessments to be performed in the subsequent experimental trials. To determine muscle strength performance (peak torque), maximal voluntary contractions (MVCs) of knee and elbow extensors and flexors were performed on an isokinetic dynamometer (Biodex System 4 Pro, Biodex Medical Systems, Shirley, NY, USA). All tests were conducted with the dominant limb, determined by the leg used to kick a ball with precision and writing hand. Participants were seated in the straight-backed chair of the dynamometer, and fitted with two straps secured diagonally across the chest in 'X', one strap across the hips, and another strap across the dominant leg so as to limit extraneous movement. The rotation axis of the dynamometer was aligned with the apparent axis of the lateral femoral condyle (knee) or through the center of the trochlea and the capitulum, bisecting the longitudinal axis of the shaft of the humerus (elbow). After establishing individual range of movement, the gravitational correction for limb weight was performed (15).

The protocol consisted of isometric MVCs of the knee extensors and flexors at 60º and, of the elbow extensors and flexors at 90º, each of which were performed 3 times and maintained for five seconds with 30-second rest periods. After two-min rest, three concentric MVCs of knee and elbow extensors and flexors were performed at $60^{\circ}\ s^{-1}$ and $120^{\circ}{\cdot}s^{-1}$. Next, participants rested for 2 minutes then performed 30 MVCs at $240^{\circ}{\cdot}s^{-1}$. The fatigue index was calculated as the rate of drop from the average of the first 5 MVCs and the 5 last MVCs performed on the 30 MVCs at $240^{\circ}{\cdot}s^{-1}$.

Participants were given explicit standardized instructions to contract from a relaxed state. Also, instructions were given to reach maximum strength in all tests, and verbal stimuli were provided by the same investigator at each test to encourage participants to achieve their best performance (i.e., "keep the torque output on the display as high as possible" for isometric contractions, and "pull back and move forward as hard and as fast as possible", for isokinetic contractions).

Visual feedback of the dynamometer force signal was provided on the computer screen. The highest torque value of the MVCs was considered for analysis performed on the software of the isokinetic dynamometer. The test–retest reliability coefficient (ICC) in children for knee flexors and extensors isokinetic concentric peak torque ranges from 0.49 to 0.81

Handgrip strength was also measured using a handheld dynamometer adjusted to the size of the participant's hand. The test was performed with the dominant limb, in a standing position with the shoulder of tested arm adducted, the elbow extended, and the forearm and wrist set in neutral position. The testing protocol consisted of three MVCs for five seconds with a rest period of 30 seconds between each trial, and the highest value was considered the maximal grip strength. Participants were instructed to squeeze the dynamometer as hard and as fast and as possible, and were verbally encouraged to achieve their best performance. The ICC for the dominant handgrip strength test for children ranges from 0.994 to 0.997.

To appropriately compare muscle strength between boys and men, values of knee extensors and flexors torque were corrected for the respective LLMM, while elbow extensors and flexors, and handgrip strength were normalized by dominant ULMM obtained by DXA.

Experimental trials. The three experimental trials were performed in a counterbalanced fashion using the Latin Square method to set the order.

Beverages for the euhydration session were provided in a double-blind manner, and experimental sessions were performed as close as possible to the same time each day. Trials were identical with the only difference being body hydration level or the type of beverage consumed: 1) 2% of hypohydration (HY, no fluids provided) 2) euhydration with a carbohydrate solution (EU-CHO) and 3) euhydration with flavoured water (EU-W) (for figure of experimental design see Appendix G). Prior to the experimental trials, participants were provided with verbal and written instructions to: refrain from practicing any strenuous physical activity 24 hours, abstain from caffeine and alcohol (adults), and avoid changing their eating habits over the course of the entire protocol. To guarantee participants arrived euhydrated, they were instructed to ingest an individually calculated amount of water corresponding to 12 $mL{\cdot}kg^{-1}$ per day, on top of their usual intake on the day prior to each experimental trial. This extra volume was divided between morning and night. On the morning of the experimental trials, participants ingested an additional 6 $mL{\cdot}kg^{-1}$ of water. A standardized meal containing 40% of the individual daily energy requirement was consumed at least 3 hours prior arriving to the experimental trial.

Trials consisted of cycling in the heat, followed by a recovery period in a thermoneutral environment, and finally, muscle strength performance tests in a thermoneutral room following the procedures described in the familiarization visit. Additionally, handgrip was measured prior to and after cycling, as well as at the end of the recovery period.

Experimental beverages and drinking protocol. The CHO beverage contained 80 g of glucose per 1,000 ml of water (8% solution), whereas the flavoured water was artificially sweetened with 3.2 g of Splenda per 1,000 ml of water. The beverages were coded by one of the investigators who was not involved in data collection or analysis. The total beverage volume consumed by each participant was calculated based on lean body mass (LBM) (1.3g CHO per $kg{\cdot}LBM^{-1}$). Participants were given their first drink (0.65g CHO per $Kg{\cdot}LBM^{-1}$) 30 min before the start of the exercise (time = -30), and consumed additional drinks (0.16 g CHO per $Kg{\cdot}LBM^{-1}$ each) at four subsequent time points (0, 25, 55 and 85 min). Additional bottled water was provided to maintain euhydration levels during EU-CHO and EU-W at the same time points. The volume of fluid (in L) for boys and men was, respectively: 0.8 ± 0.3 and 1.2 ± 0.3 in EU-CHO (P = 0.01), and 0.8 ± 0.2 and 1.4 ± 0.3 in EU-W (P = 0.01).

Pre-climate chamber protocol. Upon arrival to the laboratory on the day of each experimental trial, participants ate a standardized meal that consisted of two portions of white bread (40 g) and one portion of strawberry jam – sugar free (15 g) and, they drank either the CHO beverage (EU-CHO) or water (EU-W and HY).

After the first beverage was consumed participants waited thirty minutes in a thermoneutral room before entering the climate chamber. During this time, hydration status was verified from an initial urine sample analyzed for urine specific gravity (USG) (Atago refractometer, 2722-E04; Tokyo, Japan) and color. A USG cutoff of 1.025 was applied as the need for additional hydration prior to exercise. This was followed by a measurement of nude weight. Participants exercised wearing only shorts and athletic shoes. Rectal temperature (T_{re}) was measured using a flexible thermometer (YSI 400 series thermistor, USA). Skin temperature (T_{sk}) was measured using skin thermistors (YSI 400 series temperature sensors, USA), placed on the upper back, arm and thigh. Participants received standardized instructions on how to answer the following four perceptual evaluations: RPE using Borg scale; thermal sensation (9-point scale from “very cold” to “very hot”) and comfort (6-point scale from “very comfortable” to “very uncomfortable”); and intensity of thirst (9-point scale from “not thirsty” to “very thirsty”). Finally, handgrip was measured using the same dynamometer and protocol as the preliminary visit.

Climate chamber protocol. Upon entering the climate chamber, which was set at 38˚C and 50% relative humidity, participants rested in a seated position for 5 min. The chamber temperature was verified continuously with a wet-bulb globe temperature monitor (3M QUESTemp® QT34 Wet Bulb Globe Temperature Heat Stress Monitors, Illinois, USA). The exercise protocol consisted of 80 min of cycling divided into 4 × 20-min bouts, with a 10-min rest between bouts. Cycling was performed at a fixed metabolic heat production ($\dot{H}_p$) per unit BM (6.0 $W \cdot kg^{-1}$). $\dot{V}O_2$ was measured for 3 min in the middle (7-10 min) and at the end (17-20 min) of each bout. T_{re}, T_{sk}, HR and RPE were recorded every 5 min. Thermal sensation, thermal comfort, and thirst perception were recorded at the beginning, mid-, and in the last min of each bout.

After each bout, participants were asked to void their bladder and BM was measured with their shorts, shoes, and electrodes. Change in BM was used as the indicator of body water balance. Upon completion of the entire exercise protocol and recovery period, participants were instructed to void their bladder and dry their skin with a towel before a nude BM was obtained.

Calculations. The rate of metabolic energy expenditure (M; in $W{\cdot}m^{-2}$) was estimated using the average VO_2 ($L{\cdot}min^{-1}$) and RER measured during the experimental trials, and calculated as (30):

$$M = \dot{V}O_2 \cdot \frac{\left[\left(\frac{RER - 0.7}{0.3}\right) \cdot e_c\right] + \left[\left(\frac{1.0 - RER}{0.3}\right) \cdot e_f\right]}{60 \cdot BSA} \cdot 1000$$

where e_c is the caloric equivalent per liter of oxygen for the oxidation of carbohydrates (21.13 kJ), and e_f is that of fat (19.62 kJ). $\dot{H}_p$ (in $W{\cdot}m^{-2}$) was calculated as the difference between *M* and the external work rate (W):

$$\dot{H}_p = M - W$$

Post-chamber protocol. Participants rested in a seated position for one hour in a thermoneutral room (25 ^oC and 35% relative humidity) to ensure T_{re} was similar between trials at the time muscle strength performance tests were performed. At the end of the recovery period, urine was collected and a nude BM was measured.

Statistical analysis. An unpaired Student *t*-test was used for the between group comparisons (e.g. participant characteristics, hydration levels, and peak torque between boys and men within a trial). Two-way ANOVAs were used for parametric findings to compare the groups over time within each trial (e.g T_{re}, T_{skin}, HR, perceived responses). Post-hoc analyses were performed where appropriate using a Tukey's test. One-way, repeated measures ANOVAs were performed to analyze trials (e.g. peak torque among HY, EU-CHO and EU-W) for boys and men separately. All data are expressed as mean and standard deviation (SD), and the analysis was conducted using a statistical software package (STATISTICA for Windows 7.0, StarSoft). The significance level adopted was 5%.

3.4 RESULTS

Participant's physical characteristics are presented in Table 1. Boys and men arrived to the experimental trials with similar hydration levels according to their respective USG and urine colour: 1.021 ± 0.006 and 2.1 ± 0.8, and 1.019 ± 0.007 and 2.3 ± 1.2 (HY); 1.019 ± 0.008 and 2.2 ± 1.1, and 1.016 ± 0.006 and 2.0 ± 0.9 (EU-CHO); and 1.015 ± 0.008 and 1.7 ± 0.7, and 1.012 ± 0.010 and 2.1 ± 1.1 (EU-W).

Mean environmental conditions were similar during all experimental trials for boys and men during exercise (38.0 ± 0.1 ˚C and 50.1 ± 3.2 % relative humidity) and recovery (26.3 ± 2.7 ˚C and 32.0 ± 3.8 % relative humidity). By design, exercise intensity according to $\dot{H}_p$ (in W·kg^{-1}) was similar between boys and men, respectively: 5.7 ± 0.6 and 6.3 ± 0.6 (HY); 5.7 ± 0.6 and 6.0 ± 0.6 (EU-CHO); and 6.0 ± 0.9 and 6.5 ± 0.6 (EU-W). The percent of maximal workload was similar between boys and men in all experimental trials (28.9 ± 4.2 and 33.5 ± 5.2 %, P = 0.09; boys and men respectively).

The calculated levels of hypohydration at the end of exercise in the heat in boys and men, respectively, were: - 2.1 ± 0.2 and - 2.4 ± 0.4 % (HY); - 0.2 ± 0.2 and - 0.2 ± 0.2 % (EU-CHO); and - 0.2 ± 0.3 and - 0.2 ± 0.1 % (EU-W). Differences in the levels of hypohydration between HY vs. EU-CHO and HY vs. EU-W were highly significant ($P < 0.001$).

Physiological Responses. Figure 1 depicts T_{re}, T_{sk} and HR over the 4 × 20-min bouts of exercise. Initial T_{re} was similar among trials for boys and men. A group × time interaction was observed for boys vs. men in in HY trial ($P = 0.003$), but not in EU-CHO ($P = 0.32$) or EU-W ($P = 0.68$). More specifically, the increase in T_{re} was similar between boys and men in EU-CHO (0.8 ± 0.2 and 0.9 ± 0.2 ºC, respectively) and EU-W (0.8 ± 0.3 and 0.9 ± 0.1 ºC), but it was lower in boys compared to men in HY (0.8 ± 0.2 vs. 1.1 ± 0.1 ºC, $P = 0.012$). By the end of recovery, ΔT_{re} relative to baseline was similar between boys and men in all experimental trials: 0.1 ± 0.2 and 0.2 ± 0.3 (HY), 0.0 ± 0.2 and 0.1 ± 0.2 (EU-CHO), 0.1 ± 0.2 and 0.1 ± 0.2 (EU-W). A group × time interaction was found for weighted

T_{sk} for boys vs. men during HY (P < 0.0001). During recovery, men's weighted T_{sk} was greater in HY compared to EU-CHO from minutes 110 to 120, and it was also greater in HY compared to EU-W from minutes 110 to 140. By the end of recovery, weighted T_{sk} were similar between trials. A group × time interaction was found for HR for boys vs. men in all experimental trials (*P* < 0.001).

Perceived responses. Figure 2 shows the results of RPE, thermal sensation, thermal comfort and thirst perception during exercise and recovery A group × time interaction was observed for RPE in boys vs. men in both the HY (*P* < 0.001) and EU-W (*P* = 0.002) trials, but not EU-CHO (*P* = 0.053). RPE was greater overtime in boys compared to men. No group × time interaction was observed for thermal sensation in HY (*P* = 0.82), EU-CHO (*P* = 0.21) and EU-W (*P* = 0.82). A group × time interaction in boys vs. men was observed for thermal comfort in HY (*P* < 0.001), as well as for thirst perception in HY (*P* = 0.015), EU-CHO (*P* < 0.001) and EU-W (*P* = 0.037) trials. Boys showed a greater thermal discomfort overtime compared to men. Boys perceived higher levels of thirst in HY compared to EU-CHO and EU-W (P < 0.001) from the end of exercise to the end of recovery; and men perceived higher levels of thirst in HY compared to EU-CHO and EU-W from the middle of exercise to the end of recovery.

Muscle Strength. Figure 3 presents the results of isometric and isokinetic peak torque for knee extensors and flexors. No differences were observed between trials in boys or in men for peak torque of knee extensors (*P* = 0.70 and *P* = 0.89, boys and men, respectively) and flexors for isometric MVCs (*P* = 0.08 and *P* = 0.67) and isokinetic knee MVCs at 60 $^{o}\cdot s^{-1}$ (Extensors: *P* = 0.32; *P* = 0.93. Flexors: *P* = 0.07 and *P* = 0.38), at 120 $^{o}\cdot s^{-1}$ (Extensors: *P* = 0.76 and *P* = 0.65. Flexors: *P* = 0.65 and *P* = 0.57), and at 240 $^{o}\cdot s^{-1}$ (Extensors: *P* = 0.50, and *P* = 0.97. Flexors: *P* = 0.79 and *P* = 0.89) (see Appendix I).

On average, isometric knee extensors peak torque ($Nm\cdot LLMM^{-1}$) was 19.0 % lower in boys compared with men during EU-CHO (*P* = 0.012), but similar for HY (*P* = 0.13*)* and EU-W (*P* = 0.14*)*. Isometric knee flexors peak torque ($Nm\cdot LLMM^{-1}$) was lower in boys compared to men by 17.2% in HY (*P* = 0.018), 19.8 % in EU-CHO (*P* = 0.044) and 21.4 % in EU-W (*P* = 0.004*).* Isokinetic knee extensors peak torque ($Nm\cdot LLMM^{-1}$) at 60 $^{o}\cdot s^{-1}$ was similar in boys and men for HY (*P* = 0.06) and EU-CHO (*P* = 0.60), but 12.1% lower in boys during EU-W (*P* = 0.022). Conversely, isokinetic knee flexors peak torque ($Nm\cdot LLMM^{-1}$) at 60 $^{o}\cdot s^{-1}$ were 16.8 % lower in boys compared with men for HY (*P* = 0.021), 23.9 % lower in EU-CHO (*P* = 0.005), and 26.4 % lower in EU-W (*P* = 0.001). Isokinetic knee extensors and flexors peak torque ($Nm\cdot LLMM^{-1}$) at 120 $^{o}\cdot s^{-1}$ were lower in boys compared to men in all experimental trials (Extensors: 16.8 %, *P* = 0.02; 15.6 %, *P* = 0.014; and 15.2 %, *P* = 0.001. Flexors 20.3 %, *P* = 0.01; 20.4 %, *P* = 0.02; and 23.8 %, *P* = 0.01, in HY, EU-CHO and EU-W, respectively). Isokinetic knee extensors peak torque ($Nm\cdot LLMM^{-1}$) at 240 $^{o}\cdot s^{-1}$ was 11.6 % lower in boys compared to men in HY (*P* = 0.029) and was 16.0 % lower in EU-CHO (*P* = 0.013), but not in EU-W (*P* = 0.07).

However, isokinetic knee flexors peak torque (Nm·LLMM^{-1}) at 240 $^{o}\cdot s^{-1}$ were similar in HY (P = 0.103), EU-CHO (P = 0.13) and EU-W (P = 0.076) in boys compared with men.

Figure 4 shows the results for isometric and isokinetic peak torque for elbow extensors and flexors in boys and men during HY, EU-CHO, and EU-W. No differences among trials were observed in boys and in men for peak torque for isometric MVCs at 90^{o} for elbow extensors (P = 0.09; and P = 0.78, for boys and men respectively) and flexors (P = 0.30 and P = 0.44), as well as for isokinetic MVCs at 60$^{o}\cdot s^{-1}$ for both extensors (P = 0.65 and P = 0.10) and flexors (P = 0.46 and P = 0.31). Similarly, no differences were observed 120$^{o}\cdot s^{-1}$ for elbow extensors (P = 0.11 and P = 0.12) and flexors (P = 0.9 and P = 0.58). Finally, no differences were seen between boys and men at 240$^{o}\cdot s^{-1}$ for elbow extensors (P = 0.21; and P = 0.40) and flexors (P = 0.90 and P = 0.32).

Isometric elbow extensors peak torque (Nm·ULMM^{-1}) was similar between boys and men in all HY (P = 0.4), EU-CHO (P = 0.13), and EU-W (P = 0.5). Isometric flexors peak torque tended to be lower (16.7 %) in boys compared to men in EU-CHO (P = 0.06). Isokinetic elbow extensors peak torque (Nm·ULMM^{-1}) at 60 $^{o}\cdot s^{-1}$ was similar between boys and men for all trials. Conversely, isokinetic elbow flexors peak torque (Nm·ULMM^{-1}) at 60 $^{o}\cdot s^{-1}$ was 20.6% lower in boys compared to men in EU-CHO (P = 0.017). Isokinetic elbow extensors peak torque (Nm·ULMM^{-1}) at 120 $^{o}\cdot s^{-1}$ was 19.1 % greater in boys compared with men in HY only (P = 0.03), while flexors peak torque was lower in boys than in men in all trials

(HY: 24.6 %, P = 0.006; EU-CHO: 24.4 %, P = 0.038; and EU-W: 25.6 %, P = 0.004). Isokinetic elbow extensors peak torque ($Nm \cdot ULMM^{-1}$) at 240 $^{o} \cdot s^{-1}$ was 16.5 % greater in boys compared to men in EU-CHO (P = 0.05), but not in EU-W or HY. Isokinetic elbow flexors peak torque ($Nm \cdot ULMM^{-1}$) were 20.9 % lower at 240 $^{o} \cdot s^{-1}$ in boys compared to men in EU-W only (P = 0.05).

Figure 5 shows the fatigue index (% change) measured from the 30 MVCs at 240$^{o} \cdot s^{-1}$ for the knee and elbow extensors and flexors for boys and men in HY, EU-CHO, and EU-W. No differences were found among trials for both boys and men. However, compared to men the fatigue index (% drop) for knee extensors were 47.1%, 27.0% and 47.0% greater in boys during HY, EU-CHO and EU-W. Additionally, compared to men the fatigue index were 21.5 %, 21.7 % and 29.6 % greater in boys compared to men for knee flexors during HY, EU-CHO and EU-W. Finally, compared to men, the fatigue index in boys was 25.1% for knee extensor 21.6% in HY.

Handgrip strength ($Kg \cdot ULMM^{-1}$) was similar for HY, EU-CHO, and EU-W prior to exercise (12.3 ± 0.5 and 11.2 ± 0.2 $Kg \cdot ULMM^{-1}$, pooled average for trials, boys and men, respectively), after exercise in the heat (12.1 ± 0.2 and 10.8 ± 0.1 $Kg \cdot ULMM^{-1}$) and after recovery (12.3 ± 0.7 and 11.0 ± 0.4 $Kg \cdot ULMM^{-1}$). Compared with men, boys demonstrated greater handgrip strength after recovery in the EU-CHO trial (13.1 ± 1.4 vs. 11.3 ± 1.3 $Kg \cdot ULMM^{-1}$) (see Appendix I).

3.5 DISCUSSION

To our knowledge, this is the first study to compare the effects of hypohydration, euhydration with CHO, and euhydration with water during cycling in the heat on subsequent muscle strength performance of exercised (knee) and non-exercised (elbow and handgrip) muscles between boys and men. The main finding of this study is that muscle strength performance was not influenced by 2% hypohydration (HY) and euhydration (EU-CHO and EU-W) in either exercised or non-exercised muscles both in boys and in men. Moreover, boys showed overall lower peak torque (normalized by LLMM or ULMM) for both knee and elbow flexors and extensors compared with men following exercise in the heat. The overall drop in strength (fatigue index) in boys was greater for knee and elbow flexors in all trials, and for knee extensor in HY and elbow extensors in HY and EU-W in boys compared to men.

Hypohydration and muscle strength performance. Hypohydration up to 2-3% of body mass is commonly observed in children and adolescents during sport practice and competitions (2, 11, 17). The lack of detrimental effects of 2% hypohydration compared to euhydration on muscle strength performance in boys and men following an 80-min exercise challenge in the heat and 60 min of recovery is similar to some studies (8, 34) in adults but not others (21, 23). Several confounding factors may influence the apparent effects of hypohydration on force generation (26), including the method employed to induce hypohydration (i.e., active heat exposure, passive heat, fluid restriction, diuretics, combined techniques), the outcome measured (i.e., isometric vs isokinetic muscle strength), and other factors including fatigue, body core temperature, baseline hydration status, and caloric restriction (9, 26). However, when these factors were controlled, hypohydration resulted in strength performance reductions of 2% in adults (26). In the present study, to isolate the effects of hypohydration from body temperature and fatigue, participants rested for one hour in a thermoneutral room after exercising in the heat. Using this approach, no differences between boys and men or between trials were observed for T_{re} and T_{sk} prior to the strength performance evaluations.

Moderate hypohydration and euhydration had similar effects on post-exercise muscle strength performance in boys and men. In contrast, previous studies in children have reported impairments in high intensity endurance and power performance due to hypohydration (17, 39). Wilk *et al.* (39), for example, showed that the mean time to exhaustion on a high intensity endurance performance test was 26.8% (5.4 min) earlier at 2% hypohydration compared to euhydration (7.4 min). A possible explanation for this difference is that the MVC in the present study was maintained for seconds (i.e., 5 seconds), while the high intensity endurance tests used in earlier studies were of a longer duration (i.e., > 5 min) (39), resulting in a greater reliance on more oxidative metabolism. Additionally, the boys in Wilk et al. (39) began the high intensity endurance performance test with higher HR (~14 beats$\cdot$min^{-1}) and T_{re} (~ 0.3 ^{o}C) in the hypohydration trials which also may explain differences. Hyperthermia can impair strength production in humans, so it is important to isolate the effects of body temperature from those related to hypohydration. Hyperthermia-induced central fatigue may also reduce motor drive to skeletal muscles (31). Additionally, the combined effects of hyperthermia and exercise-induced dehydration lead to reduced plasma volume, a reduction in cardiac output, and limited perfusion of the exercising muscle (31). To verify the impact of increased body temperature superimposed onto dehydration on muscle strength, a grip strength performance test was completed. Our findings revealed grip strength was not different before and after exercise in the heat, and after one hour of recovery in thermoneutral conditions in boys and in men in all trials.

Exogenous carbohydrate and muscle strength performance. Both fluid and CHO ingestion equally improved cycling performance and their effects were additive in adults (6). The effects of CHO consumption on strength performance are controversial, with evidence to suggest that CHO supplementation might increase (38) or have no effect (19) on MVCs in adults. In the present study, an 8 % CHO-solution before and during exercise had similar effects on muscle strength performance in boys and men when compared to water. This is likely explained by a reduction in the fatigue induced by exercise in the heat during the one-hour recovery in a thermoneutral room, thereby isolating the effects of hypohydration on strength. A study (22) showed that adolescent boys who lost ~1% BM while consuming a CHO-electrolyte beverage *ad libitum* ran for a longer period of time and performed more sprints compared with those who dehydrated by 2% with fluid restriction. Another study (17) reported that compared to euhydration with water, suicide sprint (high-intensity exercise) time was higher in euhydration with a 6% CHO-electrolyte solution in adolescent boys, while maximal vertical jump (lower limb muscle power) was similar between trials. Taken together, these results suggest that a CHO solution during exercise seems to enhance aerobic performance in children, but its effects on muscle power and post-exercise muscle strength are minimal.

Age-related differences in muscle strength. Maximal muscle strength is lower in children compared to adults, even after normalization to BM (15) or muscle cross-sectional area (20). Our novel findings revealed that on average, boys demonstrated lower knee and elbow flexors and extensors peak torque compared to men following a prolonged exercise in the heat. The overall drop in strength (fatigue index) in boys was 27 to 41 % greater for knee flexors, 21 to 29% greater in elbow flexors and 25% greater for elbow extensors compared to men following exercise in the heat. These findings are contrary to previous studies, where boys showed lower fatigue compared to men (20,23). Differences in strength production between children and adults may be attributable to neuro-motor maturation, neuromuscular activation, and biomechanical factors (4, 16, 20). A possible explanation for the differences found is that in past studies (20,23), to examine central and peripheral factors of fatigability between children and adults a series of muscle performance tests, usually isometric performance tests, were done. In the present study, before undergo to the strength performance protocol with isometric and isokinetic contractions, participants cycled during 80 minutes of exercise in the heat, which may affects not only peripheral, but also central factors of fatigue. However, additional work is necessary to comprehensively understand these differences in children and adults muscle strength following exercise in the heat.

Perceptual responses. Perceived factors may contribute to early fatigue during exercise in the heat. A greater increase in RPE was observed during HY compared to EU-CHO and EU-W, suggesting that hypohydration affects RPE. Similarly, in HY both boys and men described higher thirst perception compared to EU-CHO and EU-W. A greater intensity of thirst was reported in children during exercise in the heat at lower (28) and moderate (39) levels of hypohydration compared with euhydration. Authors suggested that greater intensity of thirst might affect motivation during exercise, and consequently aerobic performance (39). However, in the present study perceived exertion and intensity of thirst did not affect muscle strength performance among experimental sessions in boys and men.

Limitations. This study was conducted in a double-blinded manner for EU-CHO and EU-W; however, it was impossible to blind for HY since no beverages were provided during exercise. Additionally, we only included a sample of male participants; future studies should also investigate female and male participants since biological maturation may have different effects on muscle strength response to hypohydration and CHO intake.

Conclusion. There is limited information about the role of hypohydration and CHO intake on strength performance. Although some evidence supports the idea that hypohydration may impair strength, while CHO improves strength, in the present study we found that 2% hypohydration and 8%-CHO solution had similar effects on subsequent isometric and isokinetic strength performance of exercised and non-exercised muscles compared with flavoured water. Boys showed lower peak torque in various knee and elbow extensor and flexor tests compared to men. Fatigue index was greater in boys compared to men in elbow and knee flexors in all trials, and for knee extensors in HY and EU-W trials. A practical application of this study is that moderate hypohydration or CHO intake before and during exercise are not expected to impair or immediately improve performance of boys and men.

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Conflict of Interest

None of the authors had a conflict of interest regarding any aspect of this research.

3.7 REFERENCES

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Table 3.1 Physical and physiological characteristics of boys and men.

Characteristics	Boys (n = 7)	Men (n = 9)
Age (years)	11.2 ± 0.5	24.0 ± 3.3*
Body mass (kg)	38.7 ± 6.0	72.9 ± 6.0*
Body height (cm)	148 ± 7.1	174 ± 5.3*
Body fat (%)	19.1 ± 4.0	15.1 ± 4.4
Total muscle mass (kg)	29.9 ± 4.1	60.3 ± 4.8*
Upper limb muscle mass (kg)	1.6 ± 0.2	3.9 ± 0.4*
Lower limb muscle mass (kg)	5.2 ± 0.9	10.2 ± 1.0*
Body surface area (m^2)	1.26 ± 0.12	1.85 ± 0.08*
Aerobic fitness		
$\dot{V}O_{2peak}$ (mL·min^{-1})	1671 ± 260	3185 ± 806*
$\dot{V}O_{2peak}$ (mL·Kg^{-1}·min^{-1})	44.7 ± 4.3	44.9 ± 10.8
HR_{max} (bpm)	189 ± 4	182 ± 9
$Workload_{max}$ (w)	137 ± 20	306 ± 75*
Biological maturation		
Tanner stage	1 (n=3) / 2 (n= 4)	–
Peak height velocity (years)	-2.1 ± 0.6	–

Tanner stage 1 = pre-pubertal; tanner stage 2 = early-pubertal. Values are mean ± SD. *P < 0.05. Dominant lower and upper limb muscle mass.

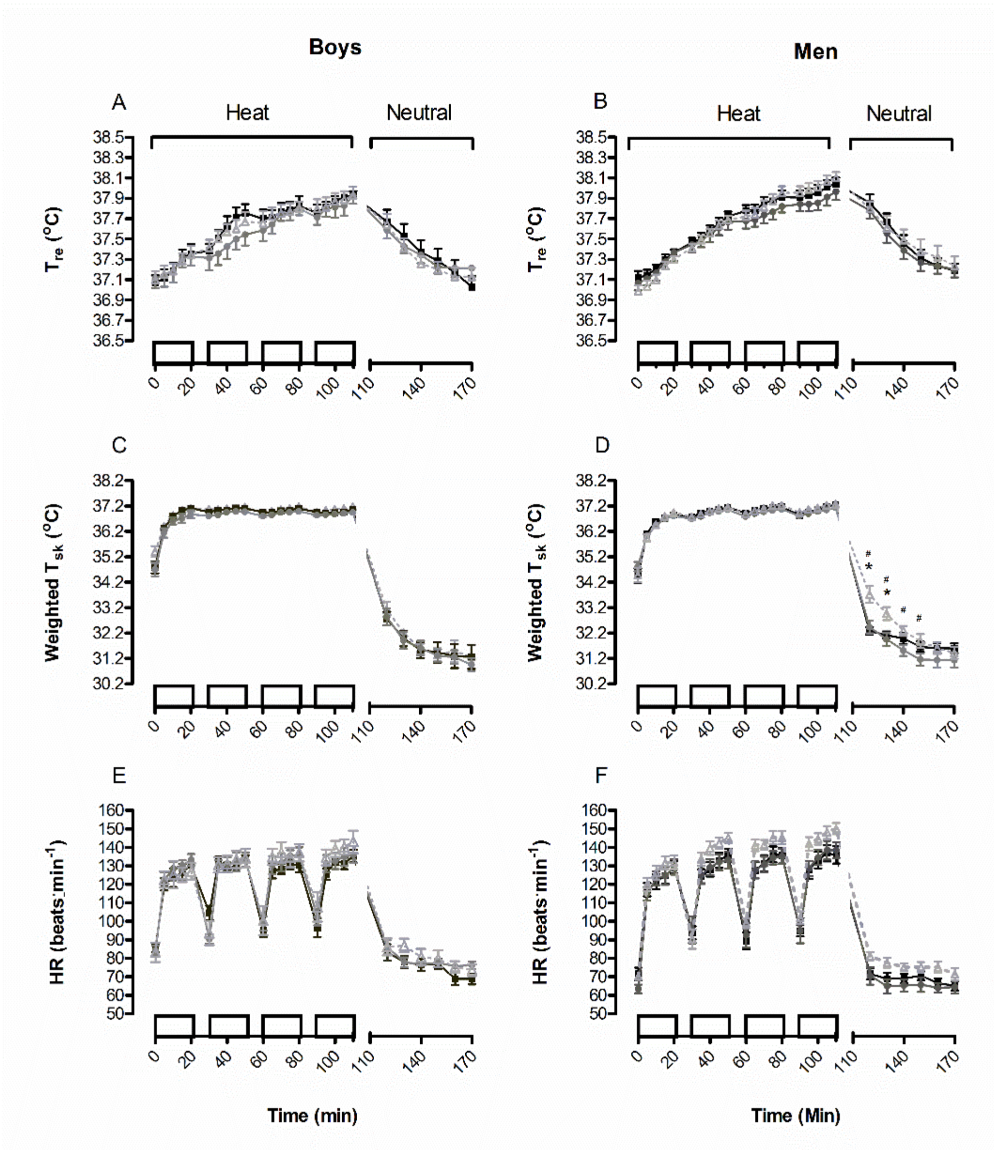

Figure 3.1 Rectal temperature (T_{re}) (A and B), weighted skin temperature (T_{sk}) (C and D) and heart rate (HR) (E and F) in boys and men during exercise in the heat (Heat) and recovery in a thermoneutral room (Neutral) during HY (∆), EU-CHO (■) and EU-W (●). Values are means ± SD. Bars along the x-axis represent exercise bouts. *Significant difference between HY and EU-W, P < 0.05. #Significant difference between HY and EU-W, P < 0.05.

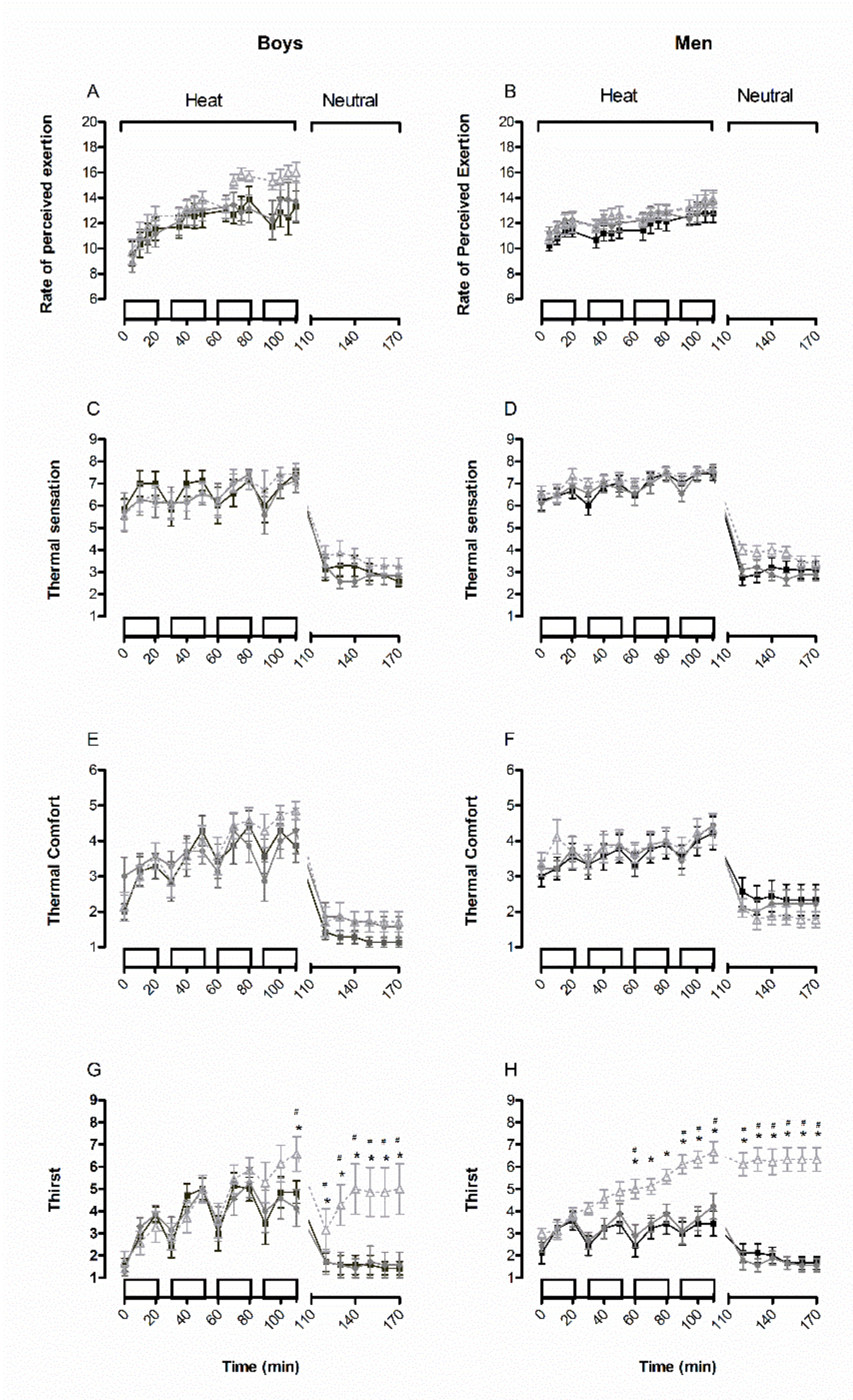
Boys
Men
A
B
C
D
E
F
G
H
Heat
Neutral
Rate of perceived exertion
Rate of Perceived Exertion
Thermal sensation
Thermal Comfort
Thirst
Time (min)

Figure 3.2 Ratings of perceived exertion (A and B), thermal sensation (C and D), thermal comfort (E and F) and intensity of thirst (G and H) in boys and men during exercise in the heat (Heat) and recovery in a thermoneutral room (Neutral) during HY (∆), EU-CHO (■) and EU-W (●). Values are means ± SD. Bars along the x-axis represent exercise bouts. *Significant difference between HY and EU-CHO. $P < 0.05$. #Significant difference between EU-CHO and EU-W, $P < 0.05$.

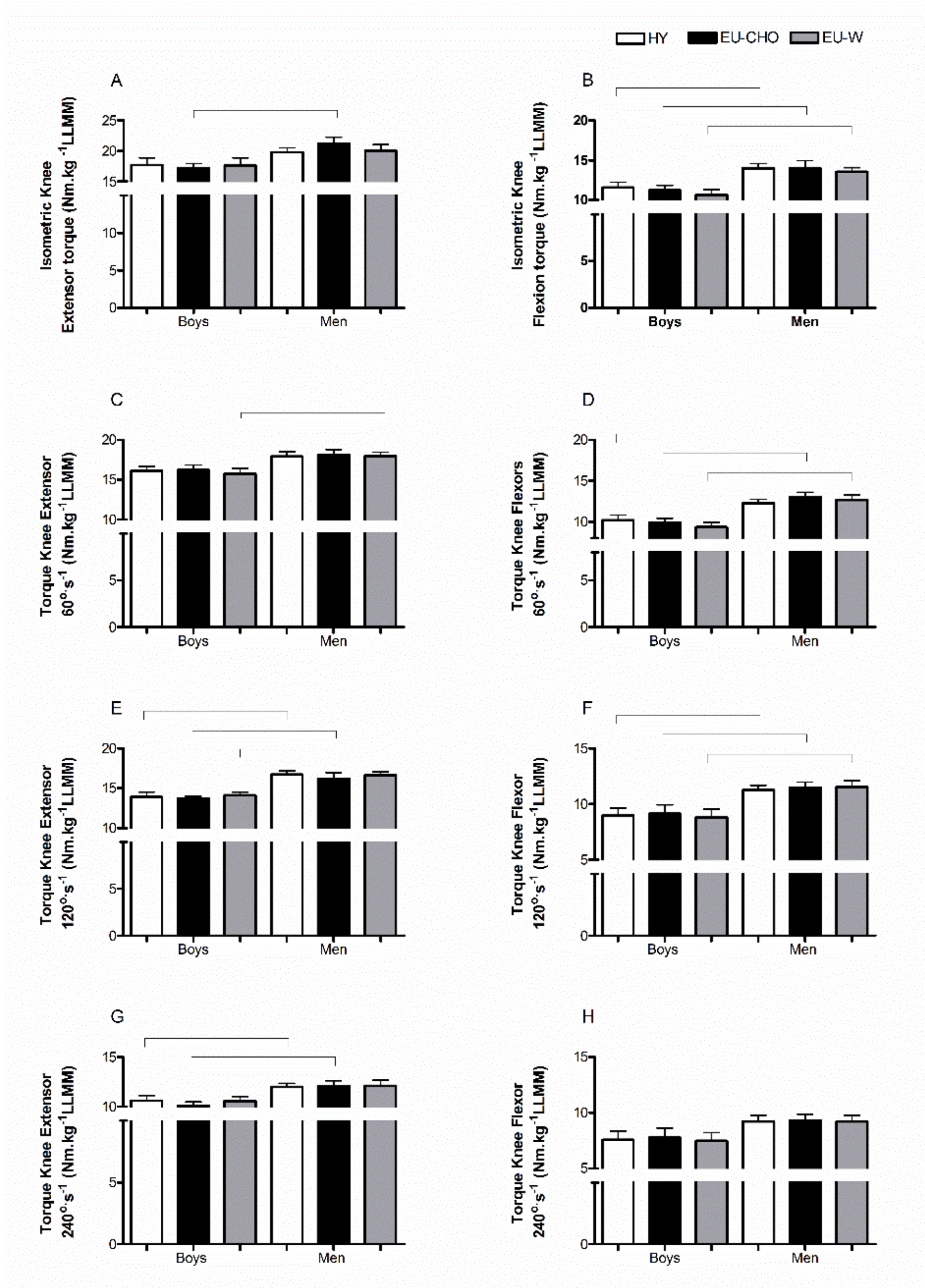

Figure 3.3 Isometric and isokinetic muscle strength performance of knee flexors and extensors in boys and men during HY (white), EU-CHO (black) and EU-W trials (grey).

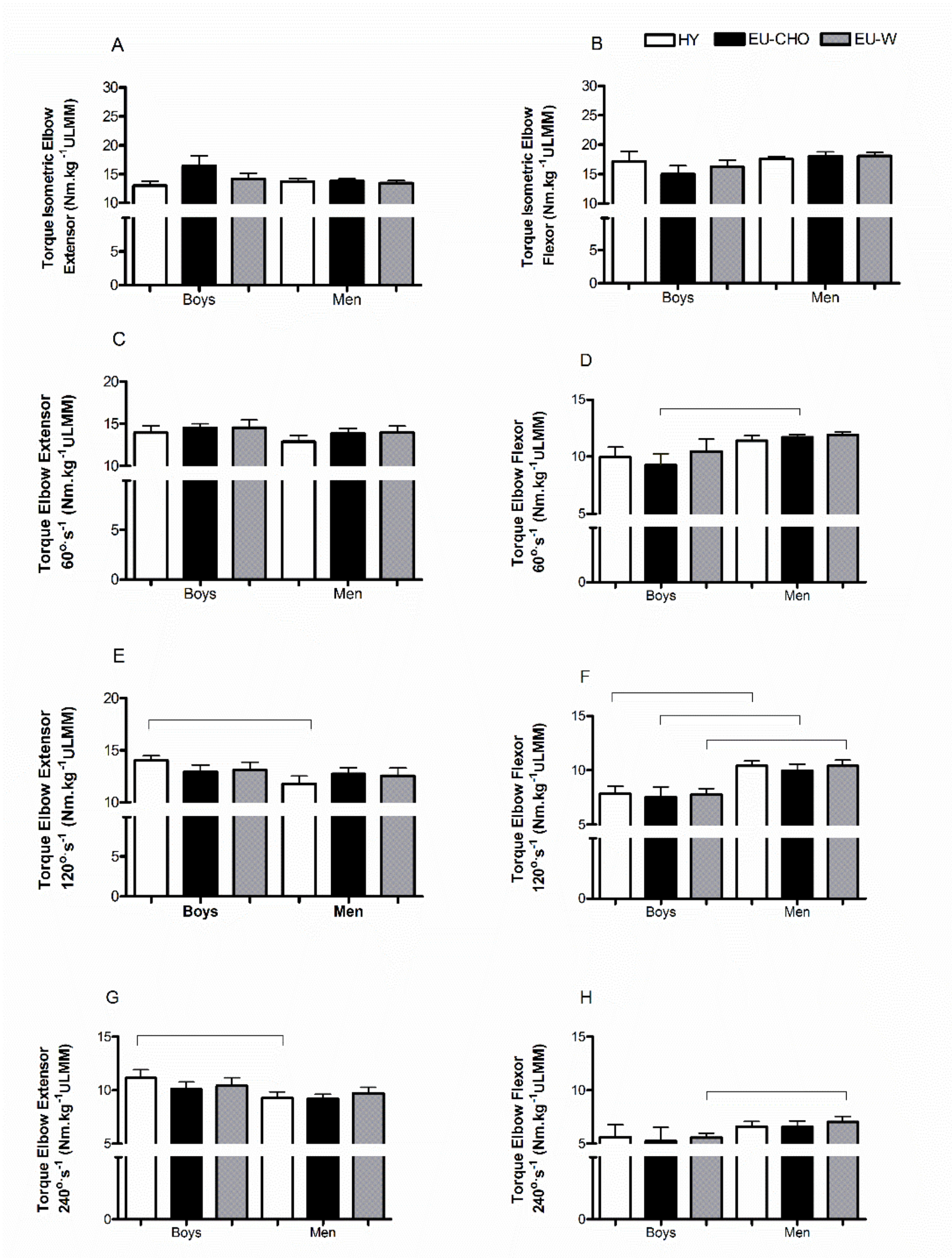

Figure 3.4 Isometric and isokinetic muscle strength performance of elbow flexors and extensors in boys and men during HY (white), EU-CHO (black) and EU-W trials (grey).

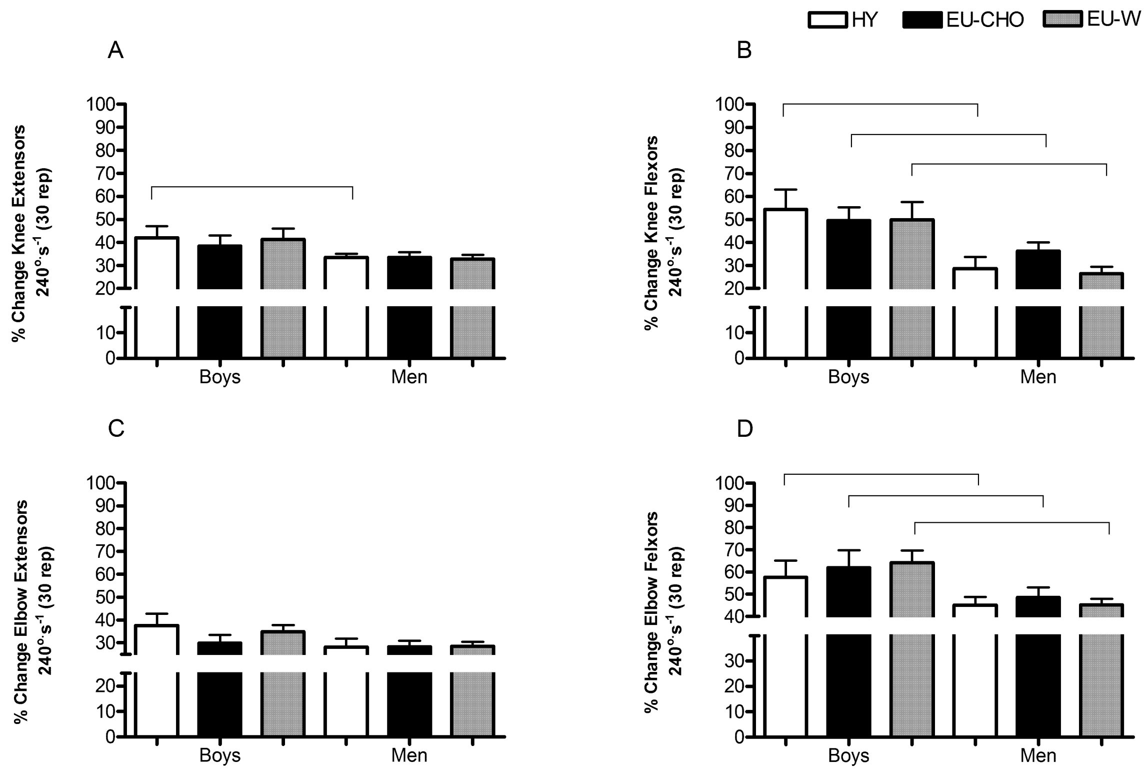

Figure 3.5 Fatigue index (% change/drop) measured from the 30 MVCs at 240º·s⁻¹ for the knee and elbow extensors and flexors in boys and men during HY (white), EU-CHO (black) and EU-W trials (grey).

CHAPTER 4: ENERGY SUBSTRATE UTILIZATION WITH AND WITHOUT EXOGENOUS CARBOHYDRATE INTAKE IN BOYS AND MEN EXERCISING IN THE HEAT

Submitted for publication as:

Gabriela T. Leites[1], Giovani S. Cunha[1,2], Lisa Chu[1], Flavia Meyer[2] and Brian W. Timmons[1]. Energy substrate utilization with and without exogenous carbohydrate intake in boys and men exercising in the heat. Journal Applied Physiology

5. Child Health & Exercise Medicine Program, Department of Pediatrics, McMaster University, Hamilton, ON, Canada.
6. Federal University of Rio Grande do Sul, Porto Alegre, RS, Brazil.

Author contributions: B.W.T. and G.T.L conceptualized and designed the research project; G.T.L acquired the data with assistance from G.S.C, L.C; G.T.L conducted the statistical analysis; G.T.L interpreted results of experiments with assistance from G.S.C, L.C., F.M. and B.W.T; G.T.L wrote the final manuscript with manuscript revisions from all authors. All authors reviewed and agreed upon the final manuscript.

Running tittle: Substrate utilization exercising in the heat

Keywords: children, substrate utilization, exercise, heat, hydration, stable isotope.

4.1 ABSTRACT

Little is known about energy yield during exercise in the heat in boys compared to men. To investigate substrate utilization with and without exogenous carbohydrate (CHO_{exo}) intake, seven boys (11.2 ± 0.5 yr) and nine men (24.0 ± 3.3 yr) cycled (4×20 min bouts) at a fixed metabolic heat production ($\dot{H}_p$) per unit body mass (6 $W \cdot kg^{-1}$) in a climate chamber (38° C and 50% relative humidity), on two occasions. Participants consumed a ^{13}C-enriched 8% CHO beverage (CARB) or placebo beverage (CONT) in a double-blinded, counter balanced manner. Substrate utilization was calculated for the last 60 min of exercise. CHO_{exo} oxidation rate (2.0 ± 0.3 vs. 2.5 ± 0.2 $mg \cdot kgFFM^{-1} \cdot min^{-1}$, $P = 0.02$) and CHO_{exo} oxidative efficiency (12.8 ± 0.6 vs. 16.0 ± 0.9 %, $P = 0.01$) were lower in boys compared to men exercising in the heat. Total carbohydrate (CHO_{total}), endogenous CHO (CHO_{endo}) and total fat (FAT_{total}) remained stable in boys and men ($P > 0.05$) during CARB, whereas CHO_{total} oxidation rate decreased ($P < 0.001$) and FAT_{total} oxidation rate increased over time similarly in boys and men during CONT ($P < 0.001$). The relative contribution of CHO_{exo} to total energy yield increased over time in both groups ($P < 0.001$). Although CHO_{exo} oxidation rate and CHO_{exo} oxidative efficiency were lower in boys compared to men, endogenous substrate metabolism and the relative contribution of fuels to total energy yield were not different between groups during exercise in the heat. CHO_{exo} intake spared endogenous substrate utilization in both groups.

NEW AND NOTEWORTHY

This study provides the new finding that during exercise in the heat boys experience a lower exogenous carbohydrate oxidation rate and oxidative efficiency compared to men. In contrast, boys and men were not different regarding endogenous substrate metabolism and the relative contribution of substrate for the total energy during exercise. These age-related differences observed in the heat are noteworthy because they expand our knowledge of exercise metabolism in children.

4.2 INTRODUCTION

A number of studies have describe marked differences in energy metabolism during exercise between children and adults (11, 17, 29, 30). Understanding child-adult differences in energy metabolism might be particularly important for children mainly because puberty is associated with a conservation of endogenous substrate, most likely due to growth-related energy requirements (30). Evidence suggests that CHO supplementation before or during exercise can shift the relative amount of substrate utilization (10, 30). More information on energy metabolism during exercise in pediatric population would be beneficial as children may have a greater reliance on CHO_{exo} compared to adults (30). Environmental conditions have been described as an important factor that alter energy metabolism during exercise (13). Heat stress impairs the oxidation rate of CHO_{exo}

in adults (14); however, there is no information about CHO_{exo} oxidation in children compared to adults under heat stress.

In adults, prolonged exercise combined with heat stress can decrease CHO_{exo} oxidation and increase reliance on endogenous metabolism (14). Possible mechanisms could explain the heat effect in lowering CHO_{exo} oxidation during exercise: 1) reduction of intestinal blood flow and CHO_{exo} absorption due to increased shift of blood flow to the skin for evaporative cooling 2) reduced gastric emptying, and 3) reduced glucose transport into the muscle. Heat stress impaired CHO_{exo} oxidation during exercise in trained male cyclists (14), although there may have been an influence of hypohydration levels during exercise in the heat. Dumke, et al. (10) showed similar CHO_{total} oxidation after consuming a placebo and a 6% glucose beverage in males during exercise in the heat. However, fat oxidation was 25% lower in the CARB trial compared to CONT, suggesting that CHO_{exo} availability can alter substrate utilization during exercise in the heat.

From a pediatric point-of-view, the potential for reduced CHO_{exo} metabolism due to heat stress may require special considerations, given the existing evidence that supports a greater reliance on CHO_{exo} during the exercise in youth compared with adults in thermoneutral conditions (25, 26, 29). However, few studies have considered maturity status as a factor influencing the rate of CHO_{exo} during exercise. Pre- and early-pubertal boys demonstrated a greater reliance on CHO_{exo} for energy compared to men, measured by ^{13}C isotope technique, during 60 minutes of cycling in thermoneutral conditions at 70% of $\dot{V}O_{2peak}$ (30). It was suggested that a greater reliance on CHO_{exo} in boys may be related to pubertal status and important for preserving endogenous fuels (30). In addition, boys utilized ~ 70% more fat and 23% less CHO_{total} compared to men during exercise without CHO_{exo} (30). While the majority of studies have focused on substrate utilization in thermoneutral environments (28, 29), exercise in the heat may impair metabolism (6, 13, 14). An experimental design that directly compares the effects of CHO_{exo} on energy metabolism is lacking, but would be important to clarify possible age- or maturity-related differences. Therefore, more information is required to identify the effects of CHO intake before and during exercise in the heat for children, in light of its potential to alter metabolism.

The effect of CHO intake on endogenous substrate utilization has not yet been evaluated during exercise in the heat in the pediatric population. Another consideration when comparing children and adults exercising in the heat is to select an experimental design that guarantees similar thermoregulatory responses to avoid bias on child-adult comparisons. To date, most comparisons of children and adults exercising in the heat have employed similar relative exercise intensities (% $\dot{V}O_{2peak}$) (2, 12), and have not considered the impact of differential metabolic heat production. A recent study (16) suggested that exercise prescribed to elicit a fixed metabolic heat production per unit body mass guaranteed similar thermoregulatory responses between boys and men exercising in the heat.

The purpose of this study was to compare the effects of CHO intake on energy metabolism between boys and men during exercise in the heat at the same metabolic heat production per unit body mass. Specifically, we examined total CHO (CHO_{total}), CHO_{exo}, endogenous CHO (CHO_{endo}), and total fat (Fat_{total}) oxidation rates during exercise in the heat. We hypothesized that providing CHO_{exo} during exercise in the heat would lead boys and men to oxidize less fat and more CHO_{exo}. We also hypothesized that boys would use relatively more CHO_{exo} as energy during exercise in the heat compared to men.

4.3 Methods

Participants**.** Seven healthy active boys (10- to 12-years-old) and nine healthy active men (20- to 30-years-old) participated in this study. Sample size was calculated with 95% statistical power and a 5% significance level based on CHO_{exo} oxidation rate in the heat, according to Jentjens et al. (13). Participants' physical and fitness characteristics are summarized in Table 1. Participants were not heat acclimatized as data collection took place during the winter months in Ontario, Canada. No medical conditions were reported and no medications were taken at the time of participation. Boys and their guardian and men were informed of the experimental protocol and potential risks and provided written informed assent, where appropriate, and/or consent to participate. The study was approved by the Hamilton Integrated Research Ethics Board.

Experimental design and procedures**.** A repeated-measures, crossover, counterbalanced, double-blinded, placebo-controlled study design was used. Participants were required to attend three visits, one preliminary and two experimental trials (see appendix G).

Preliminary trial. At the preliminary session, a physical activity questionnaire (3) was used to confirm similar activity levels between boys and men. Participants were also asked about their health status, including any previous diagnosis of chronic disease and use of medication. Biological maturation was determined using self-assessed Tanner staging (18). Standing height and body mass (BWB-800, Tanita Corporation, IL) were assessed in all participants; sitting height was also measured in boys to calculate estimated age of peak height velocity (19). Body surface area (BSA) was calculated from the measurements of body height and mass (9). Body composition was measured via Dual-energy X-ray absorptiometry (DXA) (Hologic QDR 4500A scanner, Hologic Inc., Waltham, MA).

To determine $\dot{V}O_{2peak}$, an incremental exercise test in a thermoneutral room was performed, using the McMaster All-Out Progressive Continuous Cycling Test (3). The test began at 25 W and had 25-50 W increments every 2 min, according to participant's height, while maintaining a cadence between 60 and 80 rpm. All participants were verbally encouraged to give their best performance. A calibrated electronically-braked cycle ergometer (*Lode Excalibur*, Groningen, the Netherlands) was used for all testing. Measurements of expired O_2 and CO_2 were made continuously using a calibrated metabolic cart (Vmax29, SensorMedics, Mixing Chamber method, Yorba Linda, CA, U.S.A.). $\dot{V}O_{2peak}$ was considered the highest 30-s oxygen uptake value. To gauge the participant's perception of their effort, Borg's 6-20 categorical scale was used. The test ended when 2 of the 4 following criteria were reached: 1) inability to maintain a cycling cadence above 60 rpm in spite of strong verbal encouragement; 2) HR (Polar S610; Polar Electro Oy, Kempele, Finland) >195 beats$\cdot$min^{-1}; 3) rating of perceived exertion (RPE) > 19; and (4) respiratory exchange rate > 1.1.

Hydration, diet and activity before experimental trials. Prior to the experimental trials, participants were asked to abstain from caffeine and alcohol (adults), to refrain from strenuous physical activity for 24 hours and avoid corn or corn-based products (27) to reduce background enrichment of expired CO_2 from naturally derived ^{13}C for 48 hours. On the day prior to the experimental trials, a hydration protocol was prescribed, whereby participants were instructed to consume 12 mL$\cdot$kg^{-1} of bottled water, in addition to their usual intake. This extra volume was divided between morning and night. On the morning of the experimental trials, participants ingested 6 mL$\cdot$kg^{-1} of water. The same standardized meal was consumed prior to each experimental visit, consisting of 40% of total daily energy calculated individually, and contained 50% of energy from carbohydrate, 30% from protein, and 20% from fat. Participants arrived at the laboratory at least 3 h following their standardized meal.

Experimental trials. On two separate occasions, participants cycled for 80 min divided into 4 × 20 min bouts with 10-min rest between bouts at a fixed metabolic heat production per unit body mass (~ 6 W·kg^{-1}). To avoid any influence of circadian variance, trials were completed as close as possible to the same time of day for each participant. The experimental trials were identical except for the beverage before and during the exercise in the heat. Participants were given either a ^{13}C-enriched 8% CHO beverage (CARB) or a placebo beverage (CONT). The CHO solution was artificially enriched with uniformly labeled ^{13}C-glucose to an estimated isotopic composition of +100.0 change per 1000 difference versus the $^{13}C/^{12}C$ ratio from the international standard ^{13}C Pee Dee Belemnitella-1 (+100.0% [δ-^{13}C] PDB^{-1}). The placebo was artificially-sweetened with 3.2 g of Splenda per 1,000 mL of fluid and, therefore, was identical in flavour to the CHO beverage. The beverages were coded appropriately by one of the investigators who was not involved in data collection or analysis. The total volume of beverage consumed by each participant was calibrated to his fat-free mass (FFM; 1.3 g CHO per kg·FFM^{-1}). Participants were given their first drink (0.65g CHO per kg·FFM^{-1}) 30 min before the start of the exercise (time = -30 min) and consumed additional drinks (0.16 g CHO per kg·FFM^{-1} each) at four subsequent time points (0, 25, 55 and 85 min). Additional water was provided to maintain euhydration levels during CARB and CONT trials at the same time points.

Experimental protocol. Prior to the consumption of the experimental beverage, each participant provided a resting, baseline breath sample for background enrichment of ^{13}C in expired CO_2. CHO (total, endogenous and exogenous) and fat oxidation rates were calculated from 3-min gas collection periods (SensorMedics Vmax29, Yorba Linda, CA). A 60-mL syringe was used to draw a sample of the expired gas directly from the tube connecting the participant's mouthpiece to the metabolic cart and later analyzed for the ratio of $^{13}C/^{12}C$. Participants ate a standardized meal that consisted of two portions of white bread (40 g) and one portion of strawberry jam - no sugar added (15 g) and drank the initial bolus of the experimental beverage (carbohydrate or placebo).

After the first beverage was consumed, participants waited 30 min in a thermoneutral room before entering the climate chamber. Within this time, the following procedures were performed: Participants provided a urine sample, which was immediately analyzed for urine specific gravity (USG) (Atago refractometer, 2722-E04; at Q17 a resolution of 1.000 to 1.050 density, Tokyo, Japan) and color using an 8-point scale that ranges from very pale yellow (number 1) to brownish green (number 8) to ensure similar initial hydration status (1). A USG cutoff value of 1.020 was applied (15), considering higher values in need of additional hydration prior to exercise. After emptying their bladder, nude body mass was recorded. A HR monitor (Polar Electro Oy, S610, Finland) was used, and T_{re} was measured using a flexible thermometer (YSI 400 series thermistor, USA) inserted 10 cm for boys and 12 cm for men beyond the anal sphincter. Skin temperature (T_{sk}) was measured using skin thermistors (YSI 400 series temperature sensors, USA), placed on the upper back, arm and calf. Participants with shorts, shoes and electrodes were weighed.

Participants then went into the climate chamber and mounted a cycle ergometer. Prior to exercise, participants rested for 5 min in the climate chamber, which was set to 38 ˚C and 50% relative humidity (verified by a 3M QUESTemp® QT34 Wet Bulb Globe Temperature Heat Stress Monitors, Illinois, USA) and another 3-min resting expired gas sample was collected. Participants then received an aliquot of the experimental beverage. Additional breath samples were collected for 3-min periods during the middle (7-10 min) and at the end (17-20 min) of each cycling bout to verify metabolic heat production and calculate substrate utilization. Breath samples were collected from both CARB and CONT trials to determine ^{13}C-enrichment.

T_{re}, T_{sk} and HR were recorded every 5 min. Participants were weighed with shorts, shoes and electrodes after emptying their bladder at the beginning and end of each trial to determine body weight changes (post-exercise - pre-exercise body mass).

Calculations. The rate of metabolic energy expenditure (M; in $W \cdot m^{-2}$) was estimated using the average of VO_2 ($L \cdot min^{-1}$) and RER measured during the experimental trials, and calculated as (21) :

$$M = \dot{V}O_2 \cdot \frac{\left[\left(\frac{RER - 0.7}{0.3}\right) \cdot e_c\right] + \left[\left(\frac{1.0 - RER}{0.3}\right) \cdot e_f\right]}{60 \cdot \mathrm{BSA}} \cdot 1000$$

where e_c is the caloric equivalent per liter of oxygen for the oxidation of carbohydrates (21.13 kJ), and e_f is the oxidation of fat (19.62 kJ). Metabolic heat production ($\dot{H}_p$; in $W \cdot m^{-2}$) was calculated as the difference between *M* and the external work rate (W).

$$\dot{H}_p = M - W$$

CHO_{total} and Fat_{total} oxidation rates were calculated according to the following equations (23)

$$\mathrm{CHO_{total}}(\mathrm{g \cdot min^{-1}}) = 4.59 \cdot \dot{V}\mathrm{CO_2}(\mathrm{L \cdot min^{-1}}) - 3.23 \cdot \dot{V}\mathrm{O_2}(\mathrm{L \cdot min^{-1}})$$

$$\mathrm{Fat_{total}}(\mathrm{g \cdot min^{-1}}) = -1.70 \cdot \dot{V}\mathrm{CO_2}(\mathrm{L \cdot min^{-1}}) + 1.69 \cdot \dot{V}\mathrm{O_2}(\mathrm{L \cdot min^{-1}})$$

The energy provided from CHO_{total} and fat oxidation was calculated from their energy potentials (3.87 and 9.75 $kcal \cdot g^{-1}$, respectively). Breath samples stored in Exetainer tubes (Labco Exetainer, Lampeter, Ceredigion, UK) were subsequently analyzed for $^{13}C/^{12}C$ ratio in expired CO_2 by isotope ratio mass spectrometry (IDMicro Breath Version 2.0, Compact Science Systems, Staffordshire, UK). CHO_{exo} oxidation was calculated using the equation modified from Mosora et al. (20):

$$CHO_{exo}(g \cdot min^{-1}) = \dot{V}CO_2\left[\left(R_{exp} - R_{ref}\right)\right]/(R_{exo} - R_{ref})\,(l/\kappa)$$

Where $\dot{V}CO_2$ ($L \cdot min^{-1}$) is in STPD, R_{exp} is the isotopic composition of expired CO_2 during CARB, R_{ref} is the isotopic composition of expired CO_2 during CONT at the corresponding time point, R_{exo} is the isotopic composition of the CHO_{exo}, and k (0.7426 $L \cdot g^{-1}$) is the volume of CO_2 produced by the complete oxidation of 1g of glucose. CHO_{endo} oxidation during CARB was calculated by subtracting CHO_{exo} from CHO_{total}. Because of the presence of a large bicarbonate pool in the body and because of the delay in measuring $^{13}CO_2$ production by the tissues at the mouth (22), computations of CHO_{exo} oxidation were made for the last 60 min of exercise. The oxidative efficiency of CHO_{exo} (%) was calculated as the area under the curve (AUC) of CHO_{exo} during the entire 80-min of exercise in the heat divided by the amount of glucose ingested (in g) and multiplied by 100.

Statistical Analysis

All data are expressed as mean ± SE. The significance level adopted was 5%, and the analysis was conducted using a statistical software package (STATISTICA for Windows 7.0, StarSoft). Independent *t*-tests were used for the inter-group comparisons (i.e. differences in physical and fitness characteristics, exercise intensity, urinary parameters, hypohydration levels). T_{re}, T_{sk}, HR, RER, substrate oxidation responses of boys and men over time were determined by using a group x trial x time mixed factorial ANOVAs (2 x 2 x 3 analysis). Where appropriate, a Tukey's post hoc test was used to determine significance among means. Partial eta-squared (η^2) was calculated as a measure of effect size for ANOVAs, where values of 0.01, 0.06 and above 0.15 were considered as small, medium and large, respectively (7). Cohen's d (*d)* was calculated as a measure of effect size for pairwise comparisons. Values of 0.2, 0.5, and above 0.8 were considered as small, medium and large, respectively (7).

4.4 Results

Hydration levels. All participants arrived with similar hydration levels for the experimental trials according to mean ± SE values of USG and urine colour, boys and men, respectively: 1.019 ± 0.003 and 2.2 ± 0.4, and 1.016 ± 0.002 and 2.0 ± 0.3 (CARB); 1.015 ± 0.003 and 1.7 ± 0.3, and 1.012 ± 0.002 and 2.1 ± 0.4 (CONT). The recorded body fluid balance at the end of exercise in the heat in boys and men respectively, were: - 0.2 ± 0.2 and 0.2 ± 0.2 % in CARB, and - 0.2 ± 0.3 and 0.2 ± 0.1 % in CONT. The volume of fluid (in L) required to maintain hydration during exercise for boys and men were, respectively: 0.8 ± 0.3 and 1.2 ± 0.3 L in CARB (P = 0.01), and 0.8 ± 0.2 and 1.4 ± 0.3 L in CONT (P = 0.01).

$\dot{H}_p$, T_{re}, T_{sk} and HR. Exercise intensity according to metabolic heat production per unit body mass was similar between boys and men, respectively: 5.7 ± 0.3 and 6.0 ± 0.2 $W \cdot kg^{-1}$ in CARB, and 6.0 ± 0.4 and 6.5 ± 0.2 $W \cdot kg^{-1}$ in CONT. Figure 1 depicts T_{re}, T_{sk} and HR over the 4 × 20-min bouts of exercise in the heat. No differences were found in resting T_{re} between boys and men in CARB (Figure 1A). T_{re}

increased similarly during exercise in the heat in boys and men in CARB and CONT (P = 0.48, η^2 = 0.07), resulting in a similar ΔT_{re} between boys and men in CARB (0.8 ± 0.2 and 0.9 ± 0.2 ^oC, respectively) and CONT (0.8 ± 0.3 and 0.9 ± 0.1 ^oC). T_{sk} increased in the first 15 min of exercise in the heat, and then reached a steady-state value of ~37 ^oC, with no differences between CARB and CONT (P = 0.70, η^2 = 0.05) (Figure 1B). A significant group × time interaction was found in HR ($P < 0.001$, η^2 = 0.17) (Figure 1C), but no group x trial interaction was found (P = 0.63 η^2 = 0.02). Average HR was similar between boys and men during exercise in the heat in CARB and CONT.

RER. The average resting RER was not different between boys and men in CARB (0.86 ± 0.01 and 0.89 ± 0.02, respectively) and CONT trials (0.84 ± 0.01 and 0.89 ± 0.02, respectively). The average RER during exercise was similar between boys (Figure 2A) and men (Figure 2B). In both groups, RER was greater in CARB compared to CONT over time (P = 0.011, η^2 = 0.83).

Breath enrichment. The isotopic composition of expired CO_2 during exercise in the heat in CARB and CONT are shown in Figure 3. No differences were observed at rest between groups and trials (pooled average was - 19.7 ‰ [δ-^{13}C] PDB^{-1}). During CARB, a marked increase in breath enrichment of $^{13}CO_2$ was observed over time in both boys and men, indicating a strong measurement signal compared to at rest (difference of 9.9 ‰ [δ-^{13}C]PDB^{-1}), with no differences between groups

(P = 0.10). During CONT, the ratio of $^{13}C/^{12}C$ remained constant over time in both boys and men (P = 0.08), with a pooled average of -19.7 ± 0.07 and -20.9 ± 0.31‰ [δ-^{13}C] PDB^{-1}, respectively.

Substrate utilization. CHO_{exo}, CHO_{total}, CHO_{end}, and FAT_{total} oxidation rates were calculated over the last 60 min of exercise and presented in Table 2. To control for differences in body size, values are expressed relative to FFM ($mg \cdot FFM^{-1} \cdot min^{-1}$).

CHO_{exo} oxidation rates were lower in boys compared to men (group effect: P = 0.015, d **=** 0.9). Both boys and men increased the oxidation rates of CHO_{exo} over time, with no differences between groups (group x time effect: P = 0.73, η^2 = 0.022). Oxidation efficiency of CHO_{exo} was lower in boys compared to men (12.8 ± 0.6 vs. 16.0 ± 0.9 %, respectively; P = 0.01, d **=** 1.5). CHO_{total} and CHO_{end} oxidation rates were not different between boys and men (group effect: P = 0.10 and P = 0.13, respectively), even when trial was considered (group x trial: P = 0.80, η^2 = 0.05; and P = 0.66, η^2 = 0.014, respectively). CHO_{total} oxidation rates were stable in boys and decreased in men over time (group x time: P = 0.041, η^2 = 0.20). During CARB trial, CHO_{total} and CHO_{endo} did not change over time in both groups. However, during CONT trial CHO_{total} decreased over time (time x trial effect: P < 0.001; η^2 = 0.54). Fat_{total} oxidation rates were not different between boys and men (group effect: P = 0.41). Fat_{total} oxidation rates were greater in CARB compared to CONT (trial effect: P = 0.004). FAT_{total} oxidation during CARB remained constant over time, but during CONT oxidation increased over time (trial x time: $P < 0.001$, $\eta^2 = 0.36$) similarly in both groups (group x trial: $P = 0.80$, $\eta^2 = 0.04$).

Energy yield. To account for inter-group differences in energy yield, the percent of total energy provided from CHO_{exo}, CHO_{total}, CHO_{endo}, and Fat_{total} was calculated for boys and men as shown in Figure 3.

The percent of energy contribution of CHO_{exo} was similar between boys and men (group effect: $P = 0.076$, d **=** 0.7). The relative contribution of CHO_{exo} oxidation increased over time in boys from 4.8 to 7.7 %, and in men from 6.0 to 8.9 %, showing no differences in the increment (group x time effect: $P = 0.85$, $\eta^2 = 0.011$). The percent of energy contribution of CHO_{endo} for energy yield was not different between boys and men (group effect: $P = 0.23$). The relative contribution of CHO_{endo} remained stable in CARB trial, but decreased over time in both groups in CONT trial (group x time: P = 0.073, $\eta^2 = 0.3$, group x trial: $P = 0.81$, $\eta^2 = 0.04$; and trial x time: $P < 0.0001$, $\eta^2 = 0.69$). The percent energy contributions of CHO_{total} and Fat_{total} to total yield were not different between boys and men (group effect: $P = 0.15$). The percent contribution of CHO_{total} for energy yield was lower in CONT trial (66.8 ± 3.0 % in boys and 72.2 ± 3.7 % in men, Figure 3A and B) compared to CARB (72.2 ± 0.66 % in boys vs. 81.1 ± 1.06 % in men, Figure 3C and D) (trial effect: $P = 0.008$). During CONT, the percentage of total energy provided from CHO_{total} decreased over time from 71.6 to 61.2 % in boys, and from 78.5 to 65.8% in men. However, it remained stable in CARB trial (76.2 ± 0.7% in boys, and 81.1

± 1.0 % in men) (group x time effect: $P = 0.054$, $\eta^2 = 0.19$; and trial x time effect: $P < 0.0001$, $\eta^2 = 0.69$). Consequently, Fat_{total} contribution increased over time in CONT trial, but remain stable over time in CARB trial.

4.5 Discussion

This study demonstrated that CHO_{exo} oxidation rate and CHO_{exo} oxidation efficiency were lower in boys compared to men during exercise in the heat, although the percent contribution of CHO_{exo} to total energy yield was similar. Further, no differences were observed in CHO_{total}, CHO_{endo} and Fat_{total} utilization between boys and men. CHO_{total}, CHO_{endo} and Fat_{total} oxidation did not change over time during CARB trial; while CHO_{total} decreased and Fat_{total} increased over time similarly in boys and men during CONT trial. Finally, compared to CONT trial, a higher percent contribution of CHO_{total} and lower percent contribution of Fat_{total} to total energy yield were reported in the CARB trial.

Our findings with respect to age-related differences in CHO_{exo} oxidation rate and efficiency, and relative contribution of CHO_{exo} to total yield support some but not all previously reported studies (28, 29). Unlike our results, Timmons, et al. (30) showed a greater reliance on CHO_{exo} in boys compared to men (10.7 *vs.* 8.4 $mg \cdot kg^{-1} \cdot min^{-1}$) by the end of 60 min of cycling at 70% VO_{2peak} with an ^{13}C-enriched CHO-solution (4% sucrose + 2% glucose) in a thermoneutral environment. The contribution of CHO_{exo} to total energy yield increased over time in both groups, but the average contribution was higher in boys. Other studies also verified the impact of biological maturational on CHO_{exo} oxidation and endogenous substrate with the same protocol and showed younger boys have a greater reliance on CHO_{exo} compared to older boys (29), but not girls (28). Although it is difficult to directly compare our results to those of the aforementioned studies, plausible explanations for our differences include the amount and type of CHO_{exo} provided, exercise intensity and duration, as well as environmental conditions. More specifically, the amount of CHO_{exo} provided by Timmons, et al. (30) was normalized by kg of body weight (6 aliquots x 4 mL/kg body weight), which may been advantageous for children compared to adults. In contrast, we provided CHO_{exo} normalized by fat free mass (1.3 g CHO $kg.FFM^{-1}$) since this is related to the active muscle mass during exercise and allows for a more realistic comparison between heterogeneous groups. The extent to which the rate of CHO ingestion influences metabolic responses during exercise in children has not been systematically investigated. In adults (31), the highest rates of CHO_{exo} oxidation were attained when CHO was ingested at rates of 1.0 $g{\cdot}min^{-1}$ (moderate dose). However, CHO_{exo} oxidation did not further increase when larger amounts of CHO (1.5 $g{\cdot}min^{-1}$, high dose) were ingested during exercise. The source of CHOs might also relate to the discrepant findings since the combination of CHOs (fructose + glucose) in the experimental beverage offered in early studies (25, 26, 29, 30) may have contributed to a higher relative contribution of CHO_{exo} oxidation to total energy yield in boys (15-30%) and in men (15-20%) compared to boys and men in the present study (~8.3%). However, when glucose alone was provided during prolonged exercise in the heat, the reported relative contribution of CHO_{exo} to total energy yield (~10%) for adults (14) was similar to our study.

In adults, heat stress has been suggested as an important variable that affects energy substrate utilization during exercise resulting in a greater reliance on CHO metabolism, however with lower CHO_{exo} oxidation in hot environments compared to cool environments (14). No previous study has examined the effect of CHO intake on endogenous substrate utilization during exercise in the heat in children. The possible mechanism for lower CHO_{exo} oxidation during exercise in hot compared to cool environments might be related to an increased blood flow to the skin for evaporative cooling, resulting in a reduction of flow to other organs and limiting intestinal absorption of CHO_{exo}, which may be influenced by differences in T_{re}. For this reason, we chose a low-to-moderate exercise intensity to allow both groups to complete the prolonged exercise and to minimize potential effects of higher-intensity exercise on gastric emptying or intestinal fluid absorption. The same metabolic heat production, rather than % VO_{2peak} or maximal power output, was used to match exercise intensity to induce the same thermoregulatory responses. The use of exercise intensity based on metabolic heat production normalized to body mass can provide an unbiased comparison of thermoregulatory responses (i.e. ΔT_{re}) in heterogeneous groups, such as children and adults (16).

While most studies examining CHO_{exo} oxidation during exercise in adults have been performed in cool or thermoneutral environments, a few studies have evaluated CHO_{exo} oxidation and substrate utilization during exercise in the heat.

Heat stress, compared to a cool environment, reduced CHO_{exo} oxidation rate (0.76 vs. 0.84 $g \cdot min^{-1}$) and increased CHO_{total} oxidation rate (3.18 vs. 2.85 $g \cdot min^{-1}$) in trained male cyclists pedaling for 90 minutes at 55% VO_{2peak} (13). Unlike our study, an important limitation of this work is that hypohydration levels and final T_{re} were greater during exercise in the heat, which could have reduced muscle blood flow and in turn limited CHO_{exo} oxidation. Dumke, et al. (10) showed that absolute CHO_{total} oxidation ($g \cdot min^{-1}$) was similar between a placebo and a 6% CHO beverage in males during exercise in the heat, but absolute fat oxidation was 25% lower in the experimental trial compared to placebo. Our study corroborates these findings by showing an increase in fat oxidation in CONT trial and maintenance of fat during CARB trial in boys and men. Altogether, these studies suggest that CHO_{exo} availability during exercise in the heat can alter metabolic response and possibly increase reliance on fat stores when CHO_{exo} availability is low.

In thermoneutral conditions, investigations have shown that children use proportionally more Fat_{total} and less CHO_{total} compared with adults exercising in the same relative intensity ($\%\dot{V}O_{2peak}$)[1]. This suggests that children may have a higher endogenous fat oxidation due to a higher intramuscular triglyceride availability compared with adults (4). A study showed (31) that CHO_{total} oxidation was lower and Fat_{total} oxidation was higher during exercise in boys compared to men at 70% $\dot{V}O_{2peak}$. In contrast, our findings showed that Fat_{tota} and CHO_{total} oxidation rates were similar between boys and men during exercise at a lower intensity (~ 6 W.kg-

[1]). In our study, both boys and men demonstrated a similar decrease in CHO_{total} oxidation rate and an increase in Fat_{total} oxidation rate over time in the CONT trial. This was confirmed by changes in RER values, which decreased in both boys and men over time in CONT trial. During prolonged exercise, an increased reliance on fat stores is expected with glycogen depletion and/or when CHO_{exo} availability decreases and fatty acids are released from triacylglycerol stores in adipose tissue and muscle (24).

As expected, CHO_{exo} availability shifted substrate utilization from Fat_{total} to a greater reliance on CHO_{total} during exercise in both boys and men. In the present study, CHO_{total} and Fat_{total} were relatively constant overtime in CARB trial in boys and men, compared to the CONT trial. Compared to the CONT trial the oxidation rate of CHO_{total} by the end of exercise was 18% greater in boys and 12% greater in men in the CARB trial; and oxidation rate of FAT_{total} was 66% lower in boys and 104% lower in men by the end of exercise. Based on our results, CHO intake seems to spare endogenous substrate in both boys and men during exercise in the heat, but future studies are needed to elucidate the location of this spared fuel (i.e. intra or extra muscular stores).

It is generally accepted that carbohydrate supplementation during exercise can postpone fatigue and improve exercise capacity and performance during prolonged exercise in both children (8) and adults (5, 6). The present findings may be applicable for active boys and men exercising in the heat and may vary for other populations (i.e female, sedentary individuals and chronic conditions) and environmental conditions (i.e thermoneutral and cool). There is a lack of studies comparing the effects of CHO intake on substrate utilization during exercise in the heat between boys and men. Future studies should investigate the impact of different amounts of CHO, different time points of CHO ingestion, different combinations of CHO (i.e glucose + fructose), and compare the oxidation rate in other environmental conditions (i.e., thermoneutral and cool). Future studies should also verify the impact of different exercise intensities on CHO_{exo} oxidation between children and adults.

In summary, boys had a lower CHO_{exo} oxidation rate and CHO_{exo} oxidative efficiency compared to men. However, boys and men did not show differences in endogenous substrate metabolism or the relative contribution of substrate to total energy yield over the last 60-min of exercise at the same metabolic heat production in a hot environment. Recommendations for CHO intake among active youth should consider that the ingestion of a CHO beverage during exercise in the heat may be beneficial to spare endogenous substrate in children and adults; and it may translate into delayed fatigue onset and enhanced aerobic performance, as it results in energy and fluid availability.

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Conflict of interest

None of the authors had a conflict of interest regarding any aspect of this research.

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Table 4.1 Physical and fitness characteristics of boys and men.

	Boys (n = 7)	Men (n = 9)
Age (years)	11.2 ± 0.2	24.0 ± 1.1*
Body mass (kg)	38.7 ± 2.2	72.9 ± 1.9*
Body Height (cm)	148.3 ± 2.8	174.4 ± 1.8*
Body fat (%)	19.1 ± 1.5	15.1 ± 1.5*
$\dot{V}O_{2peak}$ (mL·kg^{-1}·min^{-1})	44.7 ± 1.7	44.9 ± 3.6
HR_{max} (beats·min^{-1})	189 ± 2	182 ± 3
Tanner stage 1/2, n	3/4	–
Years from peak high velocity	-2.1 ± 0.2	–

Values are expressed in mean ± SE. *P < 0.05. $\dot{V}O_{2peak}$: peak oxygen output; HR_{max}: maximal heart rate. Tanner stage 1 = pre-pubertal; tanner stage 2 = early-pubertal.

Table 4.2. Substrate utilization during exercise in the heat in CARB and CONT for boys and men.

	CHO_{total}		CHO_{exo}		CHO_{endo}		Fat_{total}	
	CARB	CONT	CARB	CONT	CARB	CONT	CARB	CONT
Boys								
50 min	23.6 ± 1.5	24.0 ± 2.5	1.5 ± 0.1	–	22.0 ± 1.4	24.0 ± 2.5	3.1 ± 0.3	3.6 ± 0.5
80 min	24.2 ± 1.5	22.7 ± 2.1	2.0 ± 0.1	–	22.2 ± 1.5	22.7 ± 2.1	2.9 ± 0.3	4.2 ± 0.3
110 min	25.0 ± 1.8	20.4 ± 2.2	2.5 ± 0.1	–	22.5 ± 1.8	20.4 ± 2.2	3.0 ± 0.5	5.0 ± 0.4
Men								
50 min	28.0 ± 1.0	28.0 ± 1.1	2.0 ± 0.1	–	25.9 ± 1.0	28.0 ± 1.1	2.3 ± 0.3	3.1 ± 0.6
80 min	26.0 ± 1.0	25.5 ± 1.3	2.5 ± 0.1	–	23.5 ± 0.8	25.5 ± 1.3	2.6 ± 0.3	4.0 ± 0.6
110 min	26.6 ± 1.2	23.4 ± 1.6	2.8 ± 0.2	–	23.7 ± 1.1	23.4 ± 1.6	2.4 ± 0.4	4.9 ± 0.7

Values are presented in means ± SE given in $mg \cdot kgFFM^{-1} \cdot min^{-1}$. CHO_{total}, total carbohydrate oxidation; CHO_{exo}, exogenous carbohydrate oxidation; CHO_{endo}, endogenous carbohydrate oxidation; Fat_{total}, total fat oxidation. Statistical analysis is presented in the Results section.

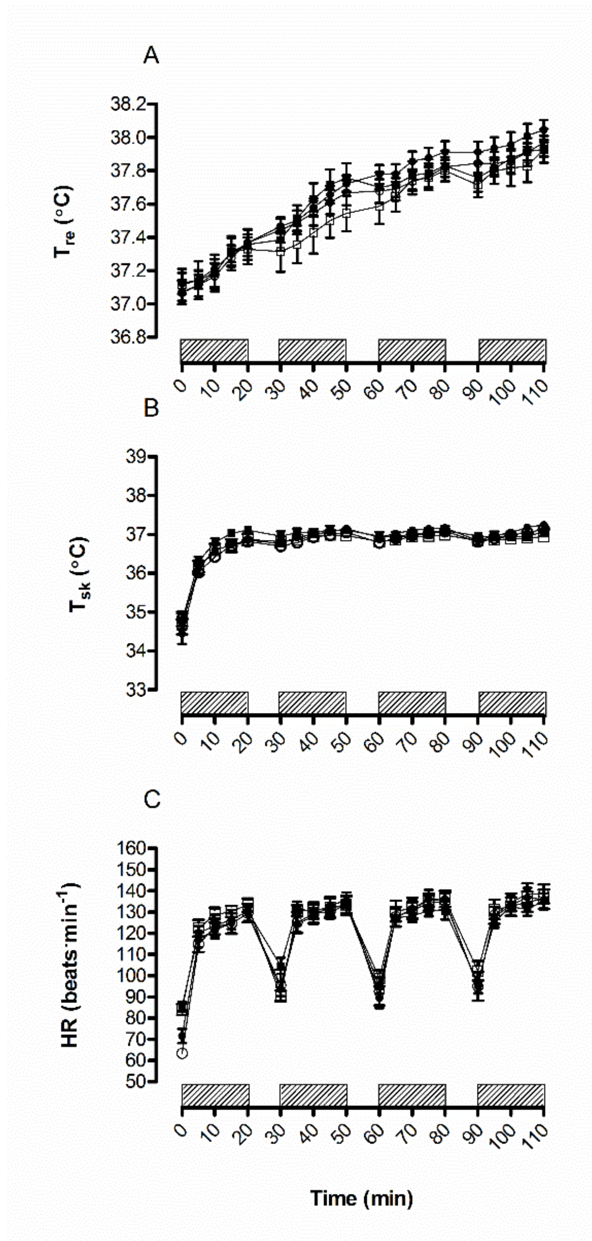

Figure 4.1 Rectal temperature (T_{re}) (A), weighted skin temperature (T_{sk}) (B) and heart rate (HR) (C) in CARB trial (solid symbols) and CONT trial (open symbols) for boys (■ and □) and for men (● and ○). Values are means ± SE. Hatched bars

represent exercise in the heat. No significant differences were found between groups for T_{re}, T_{sk} or HR (P > 0.05).

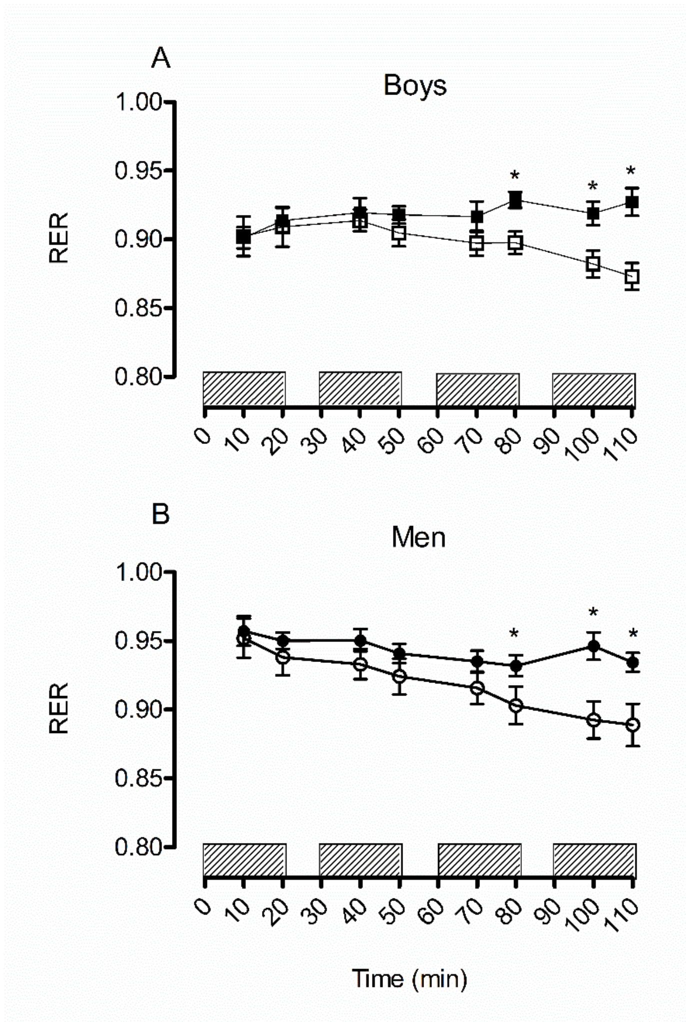

Figure 4.2. Respiratory exchange ratio (RER) during exercise in CARB trial (solid symbols) and CONT trial (open symbols) for boys (A; ■ and □) and for men (B; ● and ○). * Significant different between CARB and CONT in boys and in men (*P* < 0.05). Hatched bars represent exercise in the heat.

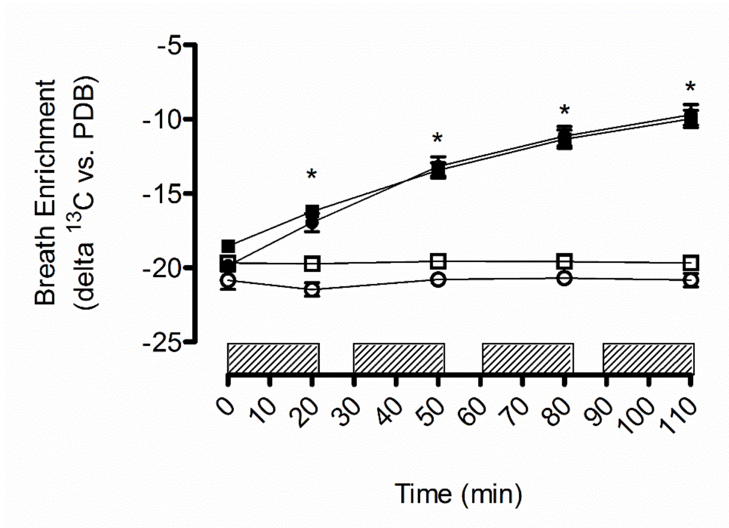

Figure 4.3. Breath enrichment of ^{13}C in expired air [δ% Pee Dee Bellemnitella (PDB)] at rest and during exercise in the heat with (CARB, solid symbols) and without (CONT, open symbols) CHO ingestion for boys (■ and □) and for men (● and ○). Values are expressed in mean ± SE. Hatched bar represent exercise. * Significantly different between CARB and CONT in boys and men (*P* < 0.05).

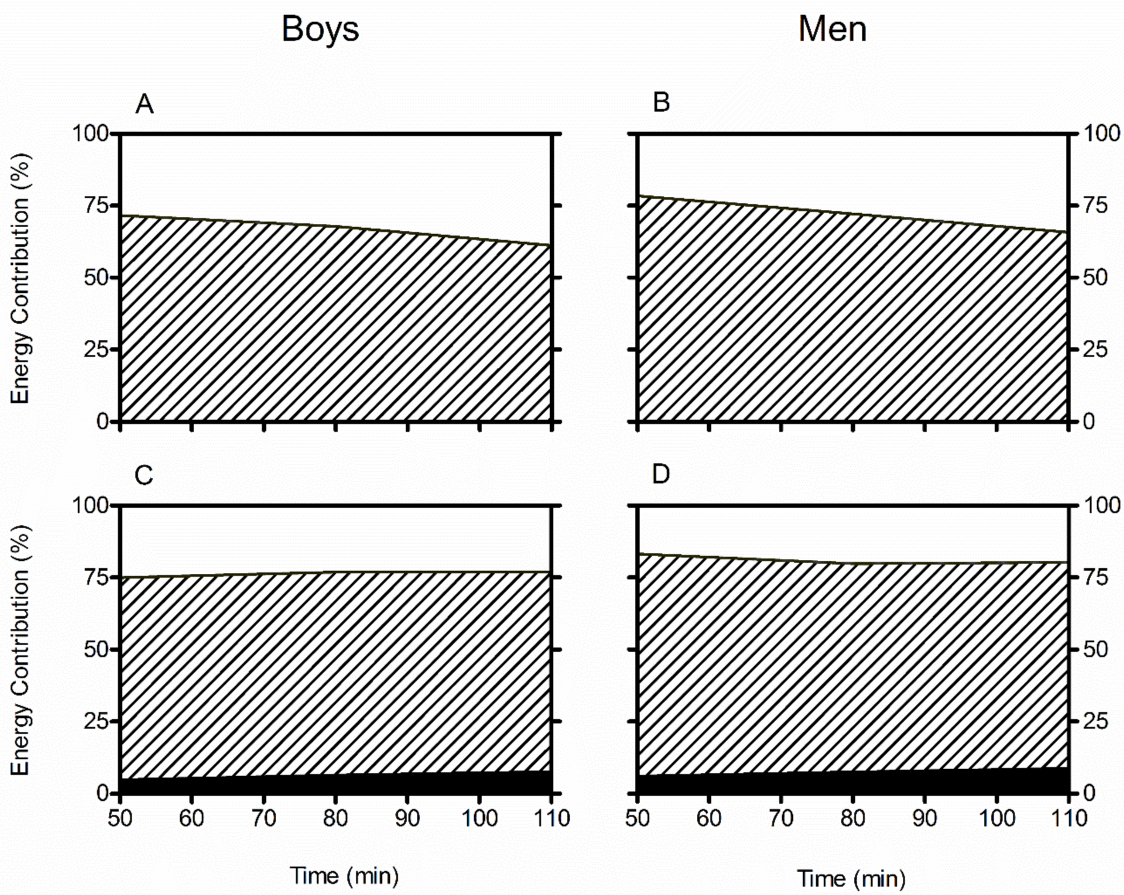

Figure 4.4. Percent of energy contribution from substrate during exercise in CONT trials (A and B) and CARB trial (C and D). The white portions represent total fat (Fat_{total}). The hatched portions represent endogenous carbohydrate (CHO_{endo}). The black portions represent exogenous carbohydrate (CHO_{exo}). CHO_{exo} percent contribution to total energy expenditure was similar between boy and men (P = 0.076). Additional statistical results are presented in the Results section.

CHAPTER 5. SUMMARY OF MAIN FINDINGS AND GENERAL DISCUSSION

In order to investigate the physiological and metabolic responses during exercise in the heat and whether maturation is an important variable, we conducted three independent but related studies comparing the responses between boys and men. The first investigation we assessed the differences in thermoregulatory responses to exercising in the heat at two different exercise intensities prescribed to elicit a fixed metabolic heat production per unit body mass and a fixed absolute metabolic heat production **(Chapter 2)**. Second, the effects of hypohydration and euhydration with and without carbohydrate on muscle strength performance in boys and men were compared. More specifically, isometric and isokinetic muscle strength performance of knee and elbow extensors and flexors were assessed following exercise session in the heat in three conditions: a) 2% of hypohydration, b) euhydration with a carbohydrate solution and c) euhydration with flavoured water (**Chapter 3**). Lastly, the effects of exogenous carbohydrate on energy metabolism were compared between boys and men who exercised in the heat at the same metabolic heat production per unit body mass and ingested the same amount of exogenous carbohydrate per kg fat free mass (**Chapter 4**).

5.1 Highlights of results

The main finding of the present thesis is that age, body size, and biological maturation differences between boys and men did not influence thermoregulatory and perceptual responses to 80 min of exercise in the heat performed at a fixed metabolic heat production per unit body mass.

Lower sweat volume was observed in boys compared to men at a fixed absolute heat production mass, but it seems that the greater sweat volume was not a thermoregulatory advantage for the adults (i.e ΔT_{re}) considering the levels of hypohydration and experimental conditions (**Chapter 2**).

Additionally, age and maturation did not impact post-exercise muscle strength performance in the euhydration with water trial (EU-W) compared to the hypohydration (HY) and euhydration with carbohydrate (EU-CHO). Boys showed lower peak torque (normalized to limb muscle mass) in various knee and elbow extensor and flexor tests compared to men. These differences in strength production between children and adults may be attributable to neuro-motor maturation, neuromuscular activation (e.g. maturation voluntary activation of agonist muscles, coactivation of antagonist muscles and muscle coordination), and biomechanical factors (lever arm and joint moment) (Dotan et al. 2012, Falk et al 2009). Fatigue index was also greater in boys compared to men for elbow and knee flexors in all trials, and for knee extensors in hypohydration and euhydration with water trials (**Chapter 3**). Additional work is necessary to comprehensively understand these differences in children and adults muscle strength following exercise in the heat.

The key identifiable difference related to age was a lower rate of exogenous carbohydrate oxidation, as well as lower exogenous carbohydrate oxidative efficiency in boys compared to men during exercise in the heat. This occurred in spite of no observed differences between boys and men with regard to endogenous substrate metabolism, nor with respect to the relative substrate contribution to total energy during 80-min of exercise at the same metabolic heat production (**Chapter 4**).

The following discussion will address the main results and novel findings in this thesis, their relevance to previous research, as well as the practical applications of our findings, and future research directions.

5.2 Effects of exercise intensity and environmental conditions on thermoregulatory responses of boys and men

Environmental heat stress and physical exercise interact synergistically to increase strain on physiological systems (Sawka et al. 2011). There is a common belief that youth are at a greater risk for exercise-induced heat illness and exercise intolerance in hot environments. Indeed, according to the American Academy of Pediatrics, when children exercise in the heat, special considerations, preparation, adaptations, and monitoring are essential (Bergeron 2011). Research focused on heat stress is critical for improving heat safety for children and adolescents engaged in youth sports and other physical activities. Therefore, the overarching goal of **Chapters 2, 3 and 4** of the present thesis was to understand the effects of heat stress during exercise on physiological and metabolic responses between boys and men's

Exercise intensity

During exercise, the metabolic heat production seems independent of the environmental condition, rather, it is proportional to individual metabolic rate (Gonzalez et al. 1978). This concept, first presented by Nielsen (1938), is especially important when examining physiological and metabolic responses to the heat in heterogeneous groups, such as children and adults (Leites et al. 2016). A major proportion of metabolic heat is generated by active muscle, and approximately 75-80% of chemical energy used for muscle contractions is converted into heat (Bar-Or and Rowland 2004). Unlike heat production, the magnitude of heat loss occurs as a function of both environmental conditions and exercise intensity (Parsons 2014). **Chapter 2** of this thesis focused specifically on understanding the impact of two distinct exercise intensities that allowed for comparison of both metabolic heat production (i.e H_p per unit body mass) and evaporative heat loss (i.e absolute E_{req}) in boys and men exercising in the heat. The intensity that elicited similar metabolic heat production per unit body mass in boys and men provided an appropriate comparison of thermoregulatory responses during exercise. More specifically, this exercise intensity (5.7 ± 1.0 and 5.6 ± 0.8 $W \cdot kg^{-1}$) resulted in similar increases in rectal temperature between boys and men (0.6 ± 0.2 vs. 0.7 ± 0.2 ℃). This finding was important and served as justification for the greater exercise intensity prescribed in **Chapters 3 and 4**.

For **Chapters 3 and 4**, we decided to prescribe the exercise intensity based on metabolic heat production per unit body mass so as to avoid any differences in body core temperature that might influence our main outcomes (i.e. strength performance and substrate utilization). To date, no studies have compared these outcomes in children and adults with metabolic heat production as the basis for exercise intensity prescription. This novel exercise prescription offers an advantage by controlling body core temperature, thereby avoiding bias in the physiological and metabolic responses between groups.

Another interesting finding in **Chapter 2** was related to exercise prescription based on a fixed absolute metabolic heat production. Since the groups were matched by aerobic fitness (VO_{2peak} in $mL \cdot kg^{-1} \cdot min^{-1}$), this exercise intensity elicited the same absolute required evaporation for heat balance. Theoretically, an exercise intensity designed to elicit the same absolute required evaporation for heat balance would allow for a fair comparison of sweating responses between groups. For example, Gagnon et al. demonstrated that the required evaporation for heat balance was the only variable that independently explained the variation in end-exercise evaporative loss in male adults (Gagnon et al. 2013). The % $\dot{V}O_{2peak}$ did not independently contribute to the variation in end-exercise evaporative heat loss, and only accounted for a very small (<2%) proportion of the variability in whole-body sweating rate. In **Chapter 2,** the sweat volume was lower in boys compared to men exercising at a fixed absolute metabolic heat production (500 ± 173 vs. 710 ± 150 mL; P = 0.041), despite the same evaporative heat balance requirement (E_{req}) (199.1 ± 34.2 vs. 201.0 ± 32.7 W, P = 0.87). This novel finding and requires further studies. An experimental trial that elicits the same absolute required evaporation for heat balance in a physiologically compensable environment would serve to clarify differences in sweating responses, specifically as they relate to age and maturational factors.

Environmental conditions

Climate can play a major role in health, subject comfort, and performance during exercise. Ambient temperature is just one component of heat stress, which also includes relative humidity, air velocity, and radiant heat (Bar-Or and Rowland 2004). The environmental condition set for **Chapter 2** was 35 ℃ and 35 % relative humidity, and air velocity averaged to 1 $m \cdot s^{-1}$. This environmental condition is a usual during summer time in many places across the world and it is related to an increased risk for heat illness. Skin temperature of boys and men (~ 36 ℃) was greater than environmental temperature. This scenario could be advantageous for children with respect to dry heat loss because of their greater surface area per unit body mass compared to adults. Indeed, the small difference (< 1 ℃) between the environmental and skin temperatures resulted statistically higher dry heat loss for boys compared to men ($P < 0.05$); however, it is important to note that from a practical perspective, this statistical significance did not likely yield any meaningful advantage for boys.

Sweat evaporation is the most effective mechanism of heat loss during the exercise and it is the only way humans dissipate heat when skin temperature is greater than environmental temperature (Arens and Zhang 2006). Based on results from **Chapter 2**, temperature and relative humidity were increased (38 ℃ and 50 % relative humidity) in **Chapter 3 and 4** so as to induce greater heat-stress, and in turn, higher rectal temperature, sweating volume, and level of hypohydration. Participants and experimental conditions in the Chapters 3 and 4 are the same. The environment conditions set for **Chapter 3 and 4** are an interesting experimental design because very few studies have imposed such environmental conditions during exercise in the children.

5.3 Effects of hypohydration during exercise in the heat in boys and men

During sport practices and competitions, athletes become progressively dehydrated when sweat loss exceeds fluid intake (Casa et al. 2000, Dougherty et al. 2006, Aragon-Vargas et al. 2013, Arnaoutis et al. 2014). Many studies have examined the effects of hypohydration on endurance performance in children (Wilk et al. 2010, Kavouras et al. 2012) and adults (Cheuvront et al. 2003, Sawka et al. 2007, Cheuvront et al. 2010, Shirreffs and Sawka 2011, Bardis et al. 2013, Davis et al. 2015), with detriments consistently observed in outcomes such as heart rate, body core temperature, effort, and thirst perceptions (Shirreffs and Sawka 2011). However, the effects of hypohydration on strength performance are controversial.

The average levels of hypohydration achieved in **Chapter 2** were 1.6% for boys, and 1.3% (fixed absolute metabolic heat production) and 1.9% (fixed metabolic heat production per unit body mass) for men. In **Chapter 3 to** induced higher levels of hypohydration (2.1% for boys and 2.4% for men) compared to

Chapter 2, higher ambient temperature (35 ºC *vs* 38 ºC) and relative humidity (35% *vs* 55%) were set. It has been suggested that children and adolescents may not ingest enough fluid to compensate their losses during (Bar-Or et al. 1980, Wilk and Bar-Or 1996, Rivera-Brown et al. 1999) or after (Bergeron 2009) prolonged periods of exercise in the heat, even when fluid is available. This may be a concern as repeated exercise bouts, such as training sessions or same-day rounds of tournament competition, are a common practice in organized youth sports (Bergeron et al. 2009, Rowland 2011). Hypohydration at the beginning of an exercise bout, resulting from failure to fully rehydrate from previous exercise, may compound the risk of heat injury or impair performance (Rowland 2011, Wilk et al. 2013).

Chapter 3 was the first study to compare the effects of hypohydration, euhydration with carbohydrate and euhydration with water during exercise in the heat and the subsequent muscle strength performance of exercised (knee) and nonexercised (elbow) muscles between boys and men. The main finding of this study was that the effects of 2% hypohydration and euhydration on subsequent muscle strength performance were similar to the trial with water for both the exercised and nonexercised muscles in boys and men. In adults, some studies demonstrated detrimental effects of hypohydration on muscle strength (Ftaiti et al. 2001, Hayes and Morse 2010), while others did not (Saltin 1964, Bigard et al. 2001). However, when controlling for confounding factors (i.e. method employed to induce hypohydration, fatigue, body core temperature, hydration status in baseline, caloric restriction), hypohydration resulted in reductions of only 2% in strength performance in adults (Judelson et al. 2007, Bowtell et al. 2013). In **Chapter 3**, to isolate the effects of hypohydration from those of body temperature and fatigue, participants spent one hour sitting in a thermoneutral room following exercise in the heat. This was verified by similar rectal and skin temperatures prior to cycling in the heat and prior to strength evaluations in boys and men across all experimental trials.

Studies showed that maximal muscle strength is lower in children compared to adults, even when normalized for body mass (De Ste Croix et al. 2003) or muscle cross-sectional area (Falk et al. 2009). However, no studies have compared strength in children and adults following exercise in the heat. It is therefore possible that children's strength performance is impaired by hypohydration to a greater extent than adults when they exercise at shorter and more intense efforts. In **Chapter 3**, boys showed lower knee and elbow flexors and extensors normalized per lower or upper limb muscle mass compared to men. The overall drop in strength in boys was greater compared to men. As previous studies suggested, this difference could be related to neuromotor maturation (De Ste Croix et al. 2003). Lastly, hypohydration may also influence the exogenous carbohydrate absorption and oxidation. For this reason, in **Chapter 4,** euhydration levels were maintained by providing water during exercise. However, future studies should verify the impact of hypohydration on exogenous carbohydrate and endogenous substrate oxidation in boys and men exercising in the heat.

5.4 Effects of exogenous carbohydrate during exercise in the heat in boys and men

Drinks containing carbohydrate in the form of sports drinks are often used to offset the deleterious effects of hypohydration, especially during prolonged (Jeukendrup 2004) or intermittent high-intensity exercises (Nicholas et al. 1995). Moreover, compared to water, there is some evidence that rehydration with sports drinks can improve high-intensity and power athletic performance in children (Menzel et al. 1999, Dougherty et al. 2006) and adults (Davis et al. 1997). One possible explanation is that the presence of carbohydrate (6-8% carbohydrate solution) may maintain a higher carbohydrate oxidation rate (Jeukendrup 2004). In **Chapter 3** of this thesis, we evaluated the impact of exogenous carbohydrate on strength performance in boys and men following exercise in the heat. Ours results suggest that ingesting an 8%-carbohydrate beverage during exercise in the heat and one hour of recovery in a thermoneutral condition had similar effects on strength performance in boys and men as the water ingestion or 2% hypohydration conditions. It is possible that the exercise in the heat may have also affected carbohydrate absorption and oxidation. Therefore, **Chapter 4** aimed to understand the effects of hot conditions on energy metabolism in boys and men.

Prolonged exercise, combined with heat stress, can affect metabolism by decreasing exogenous carbohydrate oxidation and increasing the reliance on endogenous metabolism. The possible mechanisms of lower exogenous carbohydrate oxidation during exercise in the heat compared to cool conditions might be related to: (1) increased blood flow to the skin for evaporative cooling, resulting in a reduction of flow to other organs and limiting intestinal absorption of exogenous carbohydrate; (2) reduced gastric emptying; and (3) reduced glucose transport into the muscle (Jentjens et al. 2002). **Chapter 4** investigated the effects of exogenous carbohydrate on endogenous metabolism during exercise in the heat in boys and men. An early study in thermoneutral conditions (Timmons et al. 2003) reported that exogenous carbohydrate oxidation was greater in boys compared to men. Conversely, in the present thesis, boys had a lower exogenous carbohydrate oxidation rates and exogenous carbohydrate oxidative efficiency compared to men. Groups were not different with respect to endogenous substrate metabolism and the relative contribution of fuels to total energy yields during 80-min of exercise at the same metabolic heat production. These findings suggest exogenous carbohydrate spared endogenous substrate utilization in both boys and men exercising in the heat. To clarify the controversies regarding substrate utilization in boys and men, future studies should compare the effect of environmental conditions (e.g., cold vs. thermoneutral) on substrate utilization in boys and men.

5.5 Novelty of findings

The key novel findings and the contributions of this thesis to advancing the state of knowledge in this field are summarized in Table 5.1.

Table 5.1 Summary of novel findings.

Chapter	Novel Findings	Contribution to knowledge
2	Fixed metabolic heat production per unit body mass allowed appropriate comparisons of thermoregulatory responses in children and adults, resulting in similar rectal and skin temperatures and perceptual responses between boys and men	First study to compare boys and men exercising at an intensity prescribed to elicit a fixed metabolic heat production per unit body mass and a fixed absolute heat production
	Despite similarities in absolute required evaporative loss for heat balance, boys showed lower sweat volume compared to MEN_{ABS}. The greater volume was not an advantage for adults	First study to compare boys and men exercising at a fixed intensity to elicit the same required evaporative loss for heat balance E_{req}
3	2% hypohydration and euhydration with or without carbohydrate had similar effects on subsequent muscle strength performance of exercised and non-exercised muscles in both boys and men	First study to compare the effects of hypohydration, euhydration with CHO and euhydration with water during cycling in the heat and the subsequent muscle strength performance of exercised (knee) and nonexercised (elbow and handgrip) muscles between boys and men
	Boys have lower strength performance than men. Strength performance and fatigue index were lower in boys compared to men after exercise in the heat	First study to compare strength performance between boys and men after a bout of intermittent exercise in the heat
4	Boys have lower exogenous carbohydrate oxidation rate and exogenous carbohydrate oxidative efficiency compared to men during exercise in the heat The ingestion of CHO_{exo} beverage during exercise in the heat is beneficial to spare endogenous substrate in children and adults	First study to show boys have a lower exogenous carbohydrate oxidation rate and exogenous carbohydrate oxidative efficiency compared to men during exercise in the heat. However, boys and men were not different regarding endogenous substrate metabolism and the relative contribution of substrate for the total energy during exercise

5.6 Practical applications

Information about hydration and exercise in the heat is of interest to players, athletes, coaches, parents, and sport scientists for many years. Children usually rely on their parents and coaches to provide advice and access to beverages during exercise, and to give them information about safety conditions during exercise, particularly in hot environments. Based on a common belief that exercise in the heat is a greater safety hazard for children compared to adults, a number of safeguards and limitations have been placed on exercise and sports practice in the heat, especially in the children. Indeed, children may avoid or be prohibited from playing in warm environmental conditions due to limited knowledge about exercise in the heat and the consequential hydration needs.

Boys and men had similar thermoregulatory and perceptual responses to 80 min of exercise in the heat performed at a fixed metabolic heat production per unit body mass (**Chapter 2**). When comparing children and adult thermoregulatory responses, future studies should apply a fixed metabolic heat production per unit body mass, resulting in similar increase in rectal temperature. Further, our findings suggested that thermoregulatory responses of children and adults exercising in the heat are similar with mild-to-moderate levels of hypohydration.

Mild-to-moderate hypohydration and 8%-carbohydrate solution showed no effect on the subsequent muscle strength performance compared to flavoured water in boys and men following exercise in the heat. Although boys have lower strength performance compared to men, which may be related to a number of factors including neuromotor maturation, no disadvantages were observed in boys regarding the impact of hypohydration and exogenous carbohydrate compared to water. Hydration is important to maintain and improve aerobic performance, but 2% hypohydration did not have any apparent detrimental effects on strength performance compared when compared with the euhydration trials (8% carbohydrate beverage or water). Therefore, moderate hypohydration or glucose intake before and during exercise is not expected to impair or immediately improve strength performance in boys or men.

Boys showed a lower exogenous carbohydrate oxidation rate and endogenous carbohydrate oxidative efficiency compared to men in **Chapter 4**. However, boys and men did not show differences in endogenous substrate metabolism or the relative contribution of substrate to total energy yield during 80-min of exercise at the same metabolic heat production. Recommendation of exogenous carbohydrate for young players should consider the lower oxidation rate during exercise in the heat compared to adults. The ingestion of a carbohydrate beverage during exercise in the heat is beneficial to spare endogenous substrate utilization in children and adults, and may translate into later fatigue onset and enhanced aerobic performance by increasing energy and fluid availability.

It is important that both players and individuals involved in advising players, such as parents and coaches, receive accurate information and guidance about the impact of exercise in the heat during practices and competitions. To accomplish this knowledge translation and exchange, pamphlets and websites could be created to provide easy-to-read, evidence-based resources for sporting organizations, coaches, players, and their parents. This would allow broad access and practical use of the knowledge acquired in this thesis.

5.7 Limitations and Future directions

The findings in this thesis begin to address some of the gaps in our understanding of physiological and metabolic responses to exercise in the heat in the children. Nevertheless, our results must be interpreted in light of some limitations, and further highlight a number of areas that require additional investigation, including:

Chapter 2:

1) By design, boys and men were matched by aerobic fitness and percent body fat. While this is an experimental strength, it may also limit the extrapolation of our findings to the general population of boys and men.

2) The increase in rectal temperature was similar in boys and men at a fixed metabolic heat production per unit body mass during exercise. Previous studies have also reported no differences in the rise of rectal temperature between children and adults during exercise in the heat, but the exercise was prescribed as $\%VO_{2peak}$ (Shibasaki et al. 1997, Inbar et al. 2004, Rivera-Brown et al. 2006,

Rowland et al. 2008), which may have elicited similar metabolic heat production per unit body mass. However, it is unlikely that the similar increases in rectal temperature are due to similarities in % $\dot{V}O_{2peak}$, per se. To confirm this, future studies should examine thermoregulatory responses in boys and men of different fitness levels exercising at the same metabolic heat production per unit body mass, and in turn, different relative intensities.

3) Boys experienced a lower sweating rate compared to men, despite the fact that the exercise intensity was elicited similar required evaporative loss for heat balance. Differences in sweating rate were found between boys and men, and it seems that the greater volume in adults was not an advantage given the similar levels of hypohydration. Future studies should compare children and adults exercising at intensities based on the required evaporative loss for heat balance in different environmental conditions (i.e. physiologically compensable environment) to clarify the effects of growth, maturation, exercise intensity and environment on sweating responses.

Chapter 3:

1) This study was conducted in a double-blinded manner for euhydration with and without carbohydrate trials; however, it was impossible to blind for the hypohydration trial based on the lack of beverages.

2) Future studies are needed to better understand children's strength performance, speed, and power after prolonged exercise in the heat, including more sport-specific task performance (i.e. kicking or throwing).

3) It is important to verify the effect of hypohydration and carbohydrate intake on peak torque development so as to better understand potential differences in both peak torque as well as the velocity of force production in boys and men.

4) More studies are needed to investigate the effect of perceptual responses throughout exercise in the heat in children, mainly as they relate to motivation and performance.

Chapter 4:

1) There is a lack of research comparing the effects of exogenous carbohydrate on substrate utilization during exercise in the heat between boys and men. A limitation in this study was the lack of a thermoneutral condition using the same exercise protocol and intensity, in order to differentiate the effects of biological maturation and environmental conditions. Future studies, in order to isolate the effects of the heat should examine different environmental conditions, including thermoneutral and cold environments, with similar experimental conditions.

2) Additional research is needed to examine the impact of volume, timing, concentration, and type (e.g. glucose + fructose) of exogenous carbohydrate on oxidation rates in a host of environmental conditions (i.e thermoneutral and cool).

This information is important to identify optimal carbohydrate intake before and during exercise in the heat for children to enhance performance, however also prevent excessive CHO consumption that may contribute to health complications over time.

5.8 Conclusions

The studies in this thesis examined the impact of hot environments during exercise on thermoregulatory responses, hypohydration, strength performance, and substrate utilization in boys and men. In summary, children do not present thermoregulatory or metabolic disadvantages compared to adults during exercise at moderate intensity. These findings together have practical implications for boys and men who exercise and play sports in the heat. Specifically, moderate exercise in the heat is equally safe for boys and men. The effects of hypohydration and a carbohydrate drink on muscle strength performance following exercise in the heat are minimal, compared to water. Finally, an important practical application is that carbohydrate intake spared endogenous fuels during exercise in the heat in both groups

5.9 References

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APPENDICES

APPENDIX A

Table 1.1 Impact of dehydration on power and high intensity endurance in children and muscle strength performance in adults.

Study	*Characteristics:* *Sample size* *Age* *Sex*	*Dehydration protocol* *Exercise/Rest* *Time* *Intensity* *Climatic conditions* *Recovery Period*	*Dehydration (%)*	*Δ Body Temperature (º C)*	*Main results*
Children					
Wilk et al. (2013)	N = 9 10- to 12 yr Male	Cycling 6 x 10 min 40-45% VO_{2max} 35ºC and 50-55% RH Seated 45 min neutral condition	A) 0 % B) - 1 % C) - 2 %	A) +0.1 B) +0.2 C) +0.2	**Time to exhaustion (min)** A) 7.35 ± 1.35 B) ↓ 15.6% (6.20 ± 1.26) C) ↓ 26.8% (5.38 ± 1.07) **Mechanical Work (kJ)** A) 49.3 ± 9.8 B) ↓ 15.5 % (42.3 ± 9.0) C) ↓ 23.3 % (35.5 ± 6.8)
Dougherty et al. (2006)	N = 15 12- to 15 yr Male	Treadmill and cycle ergometer 2h of 15-min exercise 50 % VO_{2max} 35ºC and 20% RH	A) 0% flavoured-water B) 0% CHO-electrolyte - 2%	A) +0.1 B) +0.02	**Suicide Sprints (s)** 37 ± 4 A) ↓ 5.4 % (35 ± 4) B) ↑ 7.5 % (40 ± 5) **Maximal vertical jump (cm)** 68 ± 15

		Rested 75 min neutral condition + Basketball protocol 4 x 12-min quarters and a 10-min halftime rest			A) ↓ 2.9% (66 ± 13) B) ↓ 2.9% (66 ± 13)
Carvalho et al. (2011)	N = 12 14- to 15 yr Male	3 separate 90-min basketball training sessions Followed by 30-min of basketball drills	A) - 2.4 % no fluid B) - 1.08 % *ad libitum* water C)- 0.65 % *ad libitum* 8% CHO-electrolyte beverage	N/A	**Suicide sprints (s)** A) 31.1 ± 0.89 B) ↓ 0.64% (30.9 ± 1.22) C) ↓ 0.64% (30.9 ± 1.39)
Adults					
Rodrigues et al. (2014)	N = 10 18- to 30 yr Male	Cycling Range of 80 – 120 min Constant load of 100 W 36–37º C and 45% RH Rest 30 min neutral condition	A) 0 B) - 2%	T_{re} < 38 ºC	**Knee extension isometric strength (Nm)** A) ↓ 3.4 % B) ↓ 16 % **Elbow flexion isometric strength (Nm)** A) ↓ 0.5 % B) ↓ 6.7 %
Hayes and Morse (2010)	N = 12 21.2 ±1.0 yr Male	Jogging 5 x 20-min 80% age predicted HR 48.5°C and 50% RH	A) 0 B) - 1 ± 0.5 C) - 1.9 ± 0.7 D) - 2.6 ± 0.8 E) - 3.3 ± 0.9 F) - 3.9 ± 1.0	Tympanic temperature < 39.5 °C	**Knee extension isometric strength ($N{\cdot}m^{-1}$)** A) 292.4 ± 53.7 B) ↓1.5 % C) ↓ 7.9 % D) ↓ 8.4 % E) ↓ 6.9 % F) ↓ 9.7 % **Knee isokinetic strength at 30º s^{-1} ($N{\cdot}m^{-1}$)** A) 241.3 ± 47.2 B) ↓ 3.4 % C) ↓ 2.5 % D) ↓ 8.2 % E) ↓ 12.8 % F) ↓ 10.8 %

					Knee isokinetic strength at 120º s^{-1} ($N{\cdot}m^{-1}$) A) 181.4 ± 49.4 B) = 0 % C) = 0 % D) ↓ 1.8 E) ↓ 2.2 F) ↓ 1.9
Judelson et al. (2007b)	N = 7 23 ± 4 yr Resistance-trained males	Day prior: abstained from the intake of fluids or fluid-rich foods Motor-driven treadmill $1.5\ m.s^{-1}$ and 3% grade inclination, 40-50% RH following morning: 10–12 h after rehydration	A) 0 B) - 2.5 C) - 5	A) 36.56 B) 36.84 C) 36.98	**Vertical jump height (cm)** A) 61.1 ± 9.4 B) ↑2.5 % C) ↑ 2.8 % **Jump Squat Power (W)** A) 5908 ± 660 B) ↑0.6 % C) ↑ 0.9 % **Isometric Squat Force (N)** A) 2829 ± 622 B) ↓ 3.7 % C) ↓ 2.4 % **Central activation ratio** A) 95.6 ± 4.9 B) ↓ 1.7 % C) ↓ 3.2 %
Vallier et al. (2005)	N = 8 male cyclist or triathletes 30.8 ± 7.3 yr	Cycling 180 min at 60%VO_{2max} 20-21ºC and 50% RH No rest (between bouts)	A) 2.2% (fluids) B) 4.1% (no fluids)	A) +0.5 B) +0.6	**Fluids vs. no fluids** ↓ 16 ± 10% maximal force isometric knee extensor muscles (N)
Gutierrez et al. (2003)	N = 12 six male athletes 21.6 ± 1.8 yr and, six female aged 24.5 ± 3.7 yr	Sauna-induced dehydration 3 x 20 min, 5-min rest, 70º C Cool shower after last sauna session	A) –1.8% male B) –1.3 % female	N/A	**Male** A) ↓ 4.0% in handgrip strength A) ↓ 4.7% squat jump height BMB A) ↓ 3.8% countermovement jump height **Female** B) ↓ 3.7 % in handgrip strength B) ↓ 1.2 % squat jump height BMB B) ↓ 2.2 % countermovement jump height

Evetovich et al. (2002)	N= 10 six male 22.5 ± 1.6 yr	Control or 48 hrs water restriction (no more than 500ml/day)	A) –0.3% B) –2.9%	N/A	**Control before and after** A) ↑ 1.7% isometric forearm flexion strength A) ↓ 4% isokinetic forearm flexion strength **Water restriction before and after** B) ↓ 5.6% isometric forearm flexion strength B) ↑ 2.2% isokinetic forearm flexion strength **Control (A) vs. Water restriction after (B)** ↓ 3.4% isometric forearm flexion strength ↓ 2.1% isokinetic forearm flexion strength
Kraemer et al. (2001)	N= 12 19.33 ± 1.16 yr wrestler male	1 week caloric restriction, thermal, and dehydration-induced weight loss (day before also exercised, i.e., cycle exercise about 20–30 min)	~ –6% (range to 4.63 from 6.75)	N/A	↓ 2.4% knee extension strength at $0°·s^{-1}$ ↓ 9.9% knee extension strength at $60°·s^{-1}$ ↓ 15.2% knee extension strength at $300°·s^{-1}$ ↓ 6.3% knee flexion strength at $60°·s^{-1}$ ↓ 11.1% knee flexion strength at $300°·s^{-1}$ ↓ 2.8% elbow flexion strength at $0°·s^{-1}$ ↓ 2.5% elbow flexion strength at $60°·s^{-1}$ ↓ 4.9% elbow flexion strength at $300°·s^{-1}$ ↓ 8.3% elbow extension strength at $60°·s^{-1}$ ↓ 4.5% elbow extension strength at $300°·s^{-1}$ ↓ 11.4% grip strength
Ftaiti et al. (2001)	N = 6 trained male runners 26 ± 3 yr	Treadmill 40 min at 65% maximal aerobic velocity Whore clothes to avoid sweat evaporation (induce hyperthermia)	2%	+2.2º C tympanic temperature	**Relative peak torque** ↓ 12% knee extension at $0°·s^{-1}$ ↓ 17% knee extension strength at $60°·s^{-1}$ ↓ 3.4% knee extension strength at $240°·s^{-1}$ ↓ 10% knee flexion strength at $0°·s^{-1}$ ↓ 9% knee flexion strength at $60°·s^{-1}$ ↓ 8.4% knee flexion strength at $240°·s^{-1}$
Montain et al. (1998)	N = 10 physically active volunteers (5 male and 5 female) 21 to 40 yr	Treadmill and cycling Moderate-intensity 40°C and 20% RH 2–3 hours (Sauna if necessary) Three to eight hours of recovery	A) –4.0% B) –0.6%	N/A	**Hypohydration (A) vs. euhydration (B)** ↑ 4.4% knee extension ↑ 16% maximal isometric force 30 s after exhaustive exercise ↓ 8% coefficient of variation for time to fatigue
Moore et al. (1992)	N = 7	Light exercise and thermal dehydration	–4.8%	N/A	↓ 8.9% knee extension strength at $180°·s^{-1}$

	national caliber collegiate wrestlers				
Webster et al. (1990)	N = 7 intercollegiate wrestlers	36-h weight loss period combination techniques	–4.9%	N/A	↓ 6.9% R knee extension strength – fast ↑ 10.2% R knee extension strength – low ↑ 7.4% R knee flexion strength – fast ↑ 11.4% R knee flexion strength – slow ↑ 5.5% L knee extension strength – fast ↑ 2.7% L knee extension strength – slow ↓ 0.7% L knee flexion strength – fast ↑ 6.9% L knee flexion strength – slow
Bijlani and Sharma (1980)	N = 14 young male	Stepping up and down a stool 30-cm high at a frequency of 20 min^{-1}, for 10 min at a time with an interval of 10 min between two consecutive bouts and heat exposition 40.8 ºC. dry and 29.6 ºC wet bulbs (2-2.5 hrs) Rested for 0, 15, or 30 min	–3.0%	<1º C the oral temperature	0 % Δ L elbow extensor strength
Greenleaf et al. (1967)	N = 12 women (divided in 2 groups) 22- to 33 yr	Rest and walking at 4.8 $km \cdot h^{-1}$, average time in the chamber (49 ºC) Average time was 326 min	A) –3.3% B) 0%	N/A	**Hypohydrate (A) vs. Control (B) groups** ↓ 7.5 % R knee flexion isometric strength ↓ 10 % L knee flexion isometric strength ↓ 7% R elbow flexion isometric strength ↓ 9.5 % L elbow flexion isometric strength
Saltin (1964)	N= 10	Exercise at 56% VO_{2max} at 36-38.5 ºC or Sauna at 80 ºC 2.5 to 4 hours Rest about 1 hour	A) –4.0% B) –3.8%	< 37.2 ºC	**Exercise (A)** ↑ 0.5% R knee extension strength ↑ 2.9% L knee extension strength ↑2.7% R elbow flexion strength ↓0.7% L elbow flexion strength **Sauna (B)** ↑ 0.2% R knee extension strength

					↓ 0.4% L knee extension strength ↑ 2.3% R elbow flexion strength ↑ 2.8% L elbow flexion strength

Data are showed as percentage change from baseline. N: sample size. ↑: improved. ↓: decreased. =: indicates no changes. L: left member. R: right member. T_{re}: rectal temperature. $VO_{2ma:}$ maximal oxygen output. N/A: nothing added. CHO: carbohydrate.

APPENDIX B

Table 1.2 Exogenous carbohydrate oxidation and substrate utilization during exercise in the heat.

Study	Participant's characteristics	Protocol	Drink characteristics	Marker/analysis	Results
Jentjens et al. (2002b)	N = 9 nonacclimated male cyclists or triathletes 24.4 ±2.6 yr	90 min of cycling 55% W_{max} 35.4 ºC and 27% RH (heat) vs. 16.4 ºC and 60% RH (cool)	8% GLU	Enriched U-^{13}C tracer, 13C/12C ration in the expired air Substrate utilization during 60-to 90 min period	**ΔT$_{re}$ (ºC)** 2.0 (heat) ↓ 30%; 1.4 (cool) **CHO total (g·min^{-1})** 3.18 (heat) ↓ 10.4%; 2.85 (cool) **Fat total (g·min^{-1})** 0.36 (heat) ↑ 33.3%; 0.48 (cool) **Exogenous glucose (g·min^{-1})** 0.76 (heat) ↑ 10.5%; 0.84 (cool) **Muscle glycogen (g·min^{-1})** 1.66 (heat) ↑24.7%; 2.07 (cool) **Plasma glucose (g·min^{-1})** 1.12 (heat) ↑ 6.3%; 1.19 (cool) **Liver-derived glucose (g·min^{-1})** 0.36 (heat) = 0%; 0.36 (cool)

Jetjens 2006	N = 9 nonacclimated male cyclists or triathletes 27.8 ±2.9 yr	120 min of cycling at 50% W_{max} 31.9 °C and 30% RH	1) 1.0 g/min of 9% GLU+ 0.5 g/min FRU 2) 1.5 g/min of 9% GLU 3) Water (Wa) Initial 600 ml bolus and 200 ml every 15 min	Corn-derived glucose monohydrate and crystalline fructose (^{13}C isotope) Substrate utilization during the 90-120 minutes	**CHO total (g·min^{-1})** 1.52 (Wa) ↑ 42.1% - 2.16 (GLU+FRU) ↑ 38.8% - 2.11 (GLU) **Fat total (g·min^{-1})** 0.97 (Wa) ↓ 25.8% - 0.72 (GLU+FRU) ↓ 24.8% - 0.73 (GLU) **CHO_{exo} oxidation (g·min^{-1})** 1.12 (GLU + FRU) ↓ 33.9% - 0.74 (GLU) **CHO_{end} oxidation (g·min^{-1})** 1.52 (Wa) ↓ 31.6% - 1.04 (GLU+FRU) ↓ 10.5% - 1.36 (GLU)
Carter et al. (2005)	N = 8	Cycled to exhaustion at	1) Sweet 6.4% CHO solution	CHO oxidation rates	**CHO oxidation (g·min^{-1})** 2.1 (Wa)

	endurance-trained men 24 ± 4 yr	55% peak power output ($60\%VO_{2max}$) 35 °C and 30% RH	(maltodextrin) (SC) 2) Nonsweet 6.4% CHO solution (maltodextrin, 0.2% aspartame) (NSC) 3) Water (Wa) Initial bolus of 8 ml·kg^{-1} and 3 ml·kg^{-1} every 15 min	were calculated from indirect calorimetry CHO oxidation at 75 min	↑ 30.9 % - 2.75 (SC) ↑ 42.9 % - 3.0 (NSC) **Exercise capacity (time to fatigue min)** 128 (Wa) ↑ 15.8% - 152 (SC) ↑11.8% - 145 (NSC)
Dumke et al. (2013)	N = 8 recreationally active males 23.5 ± 1.4 yr	1 hr cycling at 70% W_{max} and 3 hr of recovery 38°C and 40% RH	1) 6% CHO (maltodextrin) 2) placebo (P) Initial bolus of 8 ml.kg-1, aliquots of of 4 ml·kg^{-1}·30min^{-1} (~37 g·hr^{-1}) during exercise and 4 ml·kg^{-1}·hr^{-1} (~18.4 g·hr^{-1}) during recovery	Muscle glycogen (biopsies *vastus lateralis*)	**CHO oxidation (g·min^{-1})** 2.6 (P) ↑ 3.8% - 2.7 (CHO) **Fat total (g·min^{-1})** 0.8 (P) ↓ 25% - 0.6 (CHO) **Muscle glycogen (mmol·kg^{-1}) pre and post exercise** ↓44% (~121 to 81) (P) ↓35% (~118 to 65) (CHO)

Data are showed as percentage change from baseline. N: sample size. RH: relative humidity. ↑: improved. ↓: decreased. =: indicates no changes. T_{re}: rectal temperature. VO_{2ma}: maximal oxygen output. CHO: carbohydrate. CHO_{exo}: exogenous carbohydrate. CHO_{endo}: endogenous carbohydrate. P: placebo. W_{max}: maximal workload. GLU: glucose. FRU: Frutose. Wa: water.

APPENDIX C

Table 1.3 Comparison in substrate utilization during exercise in thermoneutral conditions between different biological maturation.

Study	Participant's characteristics	Protocol	Drink characteristics	Results
Timmons et al. (2003a)	N = 22 pre and early pubertal boys (9.8 ± 0.1 yr) and men (22.1 ± 0.5 yr)	Cycling for 60 min at 70% VO_{2max}.	1) Flavoured water (placebo trial) 2) ^{13}C-enriched 6% CHO (4% sucrose + 2% glucose) (carbohydrate trial) Initial bolus of 4 ml·kg^{-1}·BW^{-1} and 5 aliquots every 15-20 min.	**Fat (mg·kg^{-1}·min^{-1})** *Placebo trial* 3.7 ± 0.5 (Men) ↑67.6 % - 6.2 ± 0.6 (Boys) *Carbohydrate trial* 3.0 ± 0.4 (Men) ↑70% - 5.4 ± 0.5 (Boys) **CHO (mg·kg^{-1}·min^{-1})** *Placebo trial* 33.0 ± 0.9 (Men) ↓23.3% - 25.3 ± 1.4 (Boys) *Carbohydrate trial* 34.8 ± 1.2 (Men) ↓23% - 27.4 ± 1.5 (Boys) **CHO_{exo} (mg·kg^{-1}·min^{-1})** *Carbohydrate trial* 6.2 ± 0.5 (Men) ↑ 8.8 ± 0.5 (Boys)

				Relative proportion of total energy CHO_{exo} (%) *Carbohydrate trial* 14.6 ± 0.9% (Men) 21.8 ± 1.4% (Boys)
Timmons et al. (2007b)	N = 20 12-yr-old boys: 7 prepubertal (PP), 7 early pubertal (EP) and 6 mid- to late pubertal (M-LP) and 9 14-yr-old boys (older boys, OB)	Cycling for 60 min at 70% of theirVO_{2max}	1) Flavoured water (placebo trial) 2)^{13}C-enriched 6% CHO (4% sucrose + 2% glucose) (carbohydrate trial) Initial bolus of 12 ml·kg^{-1} and aliquots of 4 ml·kg^{-1} every 15 min	**Fat (mg·kg^{-1}·min^{-1})** *Placebo trial* 4.2 ± 0.7 (PP) 6.0 ± 1.1 (EP) 3.3 ± 1.3 (M-LP) 2.1 ± 0.9 (OB) *Carbohydrate trial* 2.0 ± 0.8 (PP) 3.3 ± 1.4 (EP) 2.0 ± 0.8 (M-LP) 0.6 ± 0.3 (OB) **Total CHO (mg·kg^{-1}·min^{-1})** *Placebo trial* 29.2 ± 2.9 (PP) 26.8 ± 2.3 (EP) 33.3 ± 3.6 (M-LP) 36.2 ± 2.5 (OB) *Carbohydrate trial* 37.1 ± 4.5 (PP) 34.2 ± 2.2 (EP) 38.8 ± 4.1 (M-LP) 40.8 ± 2.9 (OB)

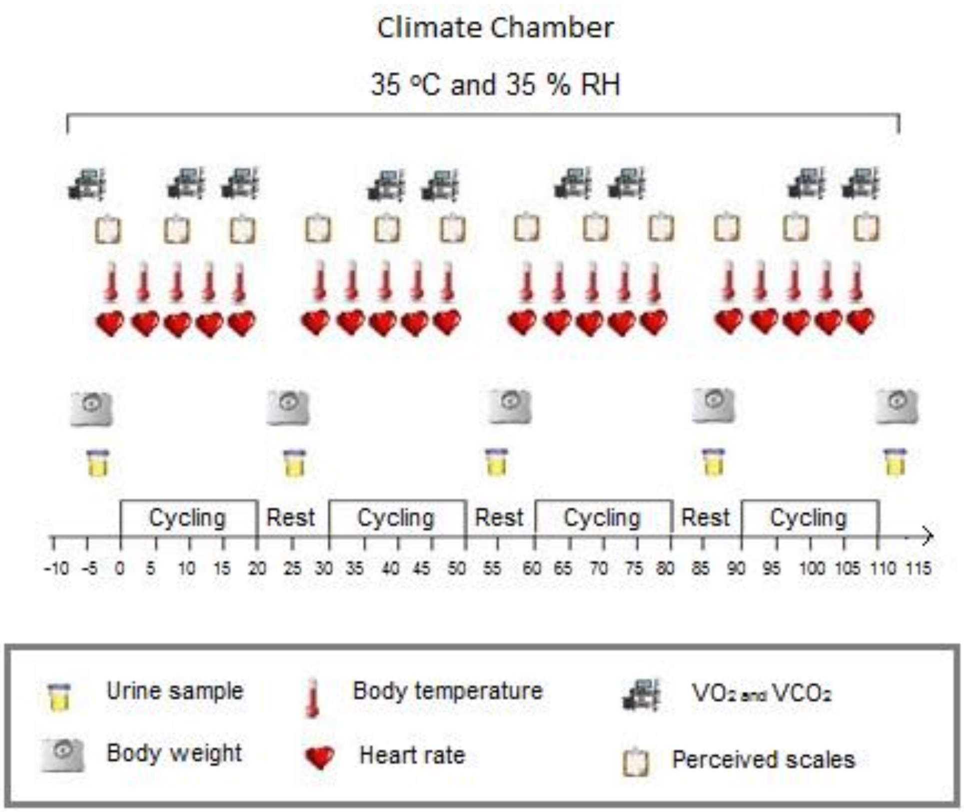
				CHO_{endo} (mg·kg^{-1}·min^{-1}) *Placebo trial* 29.2 ± 2.9 (PP) 26.8 ± 2.3 (EP) 33.3 ± 3.6 (M-LP) 36.1 ± 2.5 (OB) *Carbohydrate trial* 24.2 ± 3.0 (PP) 21.1 ± 1.9 (EP) 28.3 ± 3.8 (M-LP) 30.6 ± 2.1 (OB) **CHO_{exo} (mg·kg^{-1}·min^{-1})** *Carbohydrate trial* 13.4 ± 1.3 (PP) 13.0 ± 1.1 (EP) 10.4 ± 1.2 (M-LP) 10.2 ± 0.9 (OB) **Relative proportion of total energy CHO_{exo}** 30% PP an EP 24% M-LP and OB
Timmons et al. (2007a)	N = 22 12-yr-old and 14-yr-old girls	Cycling for 60 min at 70% VO_{2max}	1) Flavoured water (placebo trial) 2)^{13}C-enriched 6% CHO (4% sucrose + 2% glucose) (carbohydrate trial) Initial bolus of 12 ml·kg^{-1} and aliquots of 4 ml·kg^{-1} every 15 min	**Fat (mg·kg^{-1}·min^{-1})** *Placebo trial* 3.6 ± 0.7 (young) ↓ 58.3% - 1.5 ± 0.4* (older) *Carbohydrate trial* 1.8 ± 0.4 (young) ↓ 38.8% - 1.1 ± 0.6 (older) **CHO (mg·kg^{-1}·min^{-1})** *Placebo trial* 22.6 ± 2.9 (young) ↑ 43.4% - 32.4 ± 2.8*(older) *Carbohydrate trial* 31.3 ± 2.0 (young)

				↑ 9.9% - 34.4 ± 2.8 (older) **CHO_{endo} (mg·kg^{-1}·min^{-1})** *Placebo trial* 22.6 ± 2.9 (young) ↑ 43.4% - 32.4 ± 2.8*(older) *Carbohydrate trial* 24.1 ± 1.8 (young) ↑ 14.5% - 27.6 ± 2.6* (older) **CHO_{exo} (mg·kg^{-1}·min^{-1})** *Carbohydrate trial* 7.1 ± 0.5 (young) ↓ 4.2% - 6.8 ± 0.4 (older)

Data are showed as percentage change from baseline. N: sample size. ↑: improved. ↓: decreased. =: indicates no changes. T_{re}: rectal temperature. VO2ma: maximal oxygen output. CHO: carbohydrate. CHO_{exo}: exogenous carbohydrate. CHO_{endo}: endogenous carbohydrate. P: placebo. Wmax: maximal workload. PP: prepubertal. EP: early pubertal. M-LP: M-LP. OB: older boys.

APPENDIX D. Copyright Permission Form

APPENDIX E. Experimental design Chapter 2.



APPENDIX F. Consent forms Chapter 2.

Parent Information and Consent Form

Title of Study:	*Thermoregulatory Responses of Boys and Men during Exercise in the Heat*
Principal Investigator:	*Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*
Local Principal Investigator:	*Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*
Sponsor:	*Gatorade Sports Science Institute*

INTRODUCTION

Your child is being invited to participate in a research study under the supervision of Dr. Brian Timmons because your child is healthy and active. In order to decide whether or not you want to be a part of this research study, you should understand what is involved and the potential risks and benefits. This form gives detailed information about the research, which will be discussed with you. Once you understand the study, you will be asked to sign this form if you wish to participate. Your child will be asked to sign another form to confirm that he agree to participate. Feel free to discuss it with your family.

WHY IS THIS RESEARCH BEING DONE?

Children often perform physical activity in an outdoor environment, and studies suggest that children may not handle the heat as well as adults. Apparently children do not adapt to extremes of temperature as effectively as adults, due to a differences in their body size and metabolism. Some studies have suggested that children actually heat up during exercise faster than adults and this may be related to how much they dehydrate (how much body water they lose during exercise). These older studies have been used by organizations such as the American Academy of Pediatrics to develop safety guidelines, but these older studies didn’t actually directly compare children with adults. We are interested in understanding the relationship between dehydration and a child’s ability to cool their body (called thermoregulation) and to determine if there are really differences between children and adults.

WHAT IS THE PURPOSE OF THIS STUDY?
Our main goal is to compare thermoregulation between boys and men during cycling in the heat. We want to verify the relationship (if any) between dehydration and body core temperature of boys and men during cycling in the heat.

WHO IS GOING TO PARTICIPATE IN THIS STUDY?
For this study we are recruiting a total of 10 male children aged 10-12 years old, and 10 male adults aged 19-25 years old, from the Hamilton area.

WHAT WILL MY CHILD'S RESPONSIBILITIES BE IF THEY TAKE PART IN THE STUDY?
If your child volunteers to participate in this study, we will ask you to bring your child to the McMaster University Medical Centre in Hamilton on 2 occasions 4-7 days apart.

Visit	What to expect	Approximate time required
1	Your child will have a maximal exercise test conducted on a stationary bicycle. This exercise test (called a "VO_{2max} test") lasts 8 to 12 minutes and is used to determine aerobic fitness by breathing through a mouthpiece. Your child's body composition will also be determined using a machine called a dual-energy X-ray absorptiometry scanner. This scanner machine is like an X-ray. But this test delivers only 1/100th of the amount of radiation that is delivered from a typical chest X-ray. This is less than the amount of radiation your child is exposed to on a daily basis from sources of natural background radiation, such as simply walking around outside. Scanning time is about 5 minutes and is painless. Your child needs to be lying on the table during the testing and the machine will scan his body. At this visit, a questionnaire about health will also be completed.	1 hour
2	Your child will perform in our special climate room. In this room, we can control the temperature and humidity and we will make it feel like a hot summer's day for this study. Children will cycle on a stationary bicycle for 80 minutes performed as 4, 20-minute bouts, with 10 minutes rest between each bout. We will use a mouthpiece to collect breath and verify exercise intensity. We will measure your child's weight and collect any urine that he can produce before or during the exercise. We will also attach special thermometers that are small and attach to his skin so we can record skin temperature	3 hours

	and a flexible rectal thermometer will be used to record core body temperature. We will also monitor his heart rate.	

WHAT ARE THE POSSIBLE RISKS AND DISCOMFORTS?
Exercise testing: The VO_{2max} test requires your child to give a maximal effort. This means that he will feel tired after he is done the test. The other exercise your child is asked to complete is very similar to what they might normally do as part of their daily lives, especially when playing soccer or participating in summer camp or an all-day tournament outside. The exercise to be done on Visit 2 will make your child sweat and they will feel tired when they are done.

Exercising in the heat: The exercise your child will do in the heat will be similar to playing sports outdoors on a hot summer's day. Your child will probably feel tired after this exercise session in the heat just as they would after playing hard outside. In this study, we expect your child will lose about 2% of their body weight in sweat. This is not unlike what they might experience on a typical summer day. There is the possibility that when he becomes dehydrated he might feel dizzy or feel like stopping the exercise. If he has these feelings, he can stop at any time. We will closely monitor his body temperature and stop the test if he gets too hot. Once he is done his exercise – whether he stops because of these feelings or whether he completes the entire exercise session, we will provide fluids to drink right away.

Rectal thermometer: To measure body temperature for accuracy and safety, your child will use a small, flexible rectal temperature sensor. Body temperature is a main outcome in this study and is required to be measured. Placing this sensor may make your child feel embarrassed, but it is used for safety and there is really no good substitute. We will ask that you help your child place the sensor in the correct position (if necessary) and we will explain exactly how to do this. While positioning the thermometer is a delicate process, there is no risk of injury to your child if you follow our instructions. We have used this method in numerous studies with children the same age as your child.

WHAT ARE THE POSSIBLE BENEFITS FOR MY CHILD?
We cannot promise any personal benefits to you or your child from his participation in this study. However, we will share the results of the study and of your son with you and talk to you about appropriate fluid intake during exercise when your child is active. We will also make each exercise session fun and enjoyable. He will also learn his fitness level and his body composition.

WHAT INFORMATION WILL BE KEPT PRIVATE?
All of your child's information will be stored in locked filing cabinets for 10 years under the supervision of Dr. Brian Timmons. We will supervise access to your

child's information by other people in our group, only if necessary. Your child will be assigned a participant number, and this number will be used to identify his records. These records will be kept confidential. If the results of the study are published, your child's identity will remain confidential.

CAN PARTICIPATION IN THE STUDY END EARLY?

If you and your child volunteer to be in this study, you or your child may withdraw at any time with no prejudice. The investigator may also withdraw you from this research if circumstances arise which warrant doing so.

WHAT COSTS ARE THERE FOR PARTICIPATING IN THIS STUDY?

There will be no costs to you for participating in this study. We will reimburse you any parking expenses occurred. We will also give your child a small honorarium for his time and effort equal to $10 for every hour spent in the laboratory.

IF I HAVE ANY QUESTIONS OR PROBLEMS, WHOM CAN I CALL?

If you have any questions about the research now or later, or if you think you have a research-related injury, you can contact Gabriela at our research office at 905-521-2100, extension 73567 or you can contact Dr. Brian Timmons directly at 905-521-2100 extension 77615.

If you have any questions regarding your rights as a research participant, you may contact Office of the Chair of the Hamilton Integrated Research Ethics Board at 905-521-2100 extension 42013.

CONSENT STATEMENT

I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction and to the satisfaction of my son. I agree to allow my son to participate in this study entitled *"Thermoregulatory Responses of Boys and Men during Exercise in the Heat"*. I understand that I will receive a signed copy of this form.

____
Name of Participant (child's name)

____
Name of Legally Authorized Representative

____
Signature of Legally Authorized Representative/Date

Consent form administered and explained in person by:

__________________________ ____________________________
Name and title Signature/Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent for their son to participate in this research study.

____
Signature of Investigator/Date

FUTURE RESEARCH
At the end of the study, we may wish to store leftover sample for use in a future study. We will not store your child's sample longer than 10 years. All records identifying your child will remain confidential. Information about your child will not be released. If the results of the study are published, your child's identity will remain confidential.

CONSENT STATEMENT FOR STORAGE OF SAMPLES
I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to have my child's sweat and urine stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name of Participant (child's name)

____
Name of Legally Authorized Representative

____
Signature of Legally Authorized Representative /Date

Consent form administered and explained in person by:

____
Name and title

____
Signature/Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to have their child's urine and sweat stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

Child Assent Form

Title of Study: *Thermoregulatory Responses of Boys and Men during Exercise in the Heat*

Principal Investigator: *Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*

Local Principal Investigator: *Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*

***Sponsor**:* *Gatorade Sports Science Institute*

Why are we doing this study?
A research study is a way to learn more about people. We are doing a research study about what are the differences between children and adults during exercise in heat. When you exercise in warm weather, your body sweats and loses water and this affects the temperature of your body. We want to see if exercise affects body temperature the same way in boys and men.

Why am I being asked to be in the study?
We are inviting you to be in the study because you are a healthy and active boy and between the ages of 10 to 12.

What if I have questions?
You can ask questions if do you do not understand any part of the study. If you have questions later that you don't think of now, you can talk to me (Gabriela) again or ask your mom or dad to call me (905-521-2100 extension 73567).

If I am in the study what will happen to me?
If you decide that you want to be part of this study, you will come visit us 2 times. When you visit us, we will do a few tests. On the first day, you will have your weight measured, your height taken and you will also be asked to ride a special bicycle to see how fit you are.

In the second visit you will get to ride the special bike in our special room that we will make feel like a hot day. We will write down how fast your heart is beating and how much air you are breathing when you are exercising. You will wear specials thermometers to see how hot your body gets. We will measure how hot your body gets by having you put a very thin and flexible plastic wire in your bum. We will collect a pee sample before and after you do the exercise.

Will I be hurt if I am in the study?
You will not be hurt if you are in the study. The exercise you will do might make you feel a little tired, but it will be like playing outside on a hot summer day. The plastic wire in your bum is used to measure how hot your body gets. This is the main reason for doing the study. Plus, this is done to make sure you stay safe while you are exercising.

Will the study help me?
If you are in the study it may not benefit you directly, but we will learn about how kids might be different than adults in how they keep their body cool during exercise.

Do I have to be in this study?
You do not have to be in this study, if you do not want to be. If you decide that you don't want to be in the study after we begin, that's OK too. Nobody will be angry or upset. It is your choice. We are discussing the study with your parents and you should talk to them about it too.

What happens after the study?
When we are finished this study we will write a report about what was learned. This report will not include your name or that you were in the study.

Assent:

If you decide you want to be in this study, please print your name. If you decide that you don't want to be in the study, even if you have started in the study, then all you have to do is tell Gabriela or Dr. Timmons that you don't want to be in the study anymore.

I,______________________ (Print your name) would like to be in this research study.

______________________ (Date of assent)

______________________ ______________________

Investigator name Signature and Date

Adult Information and Consent Form

Title of Study: *Thermoregulatory Responses of Boys and Men during Exercise in the Heat*

Principal Investigator: *Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*

Local Principal Investigator: *Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*

Sponsor: *Gatorade Sports Science Institute*

INTRODUCTION

You are being invited to participate in a research study under the supervision of Dr. Brian Timmons because you are healthy and active. In order to decide whether or not you want to be a part of this research study, you should understand what is involved and the potential risks and benefits. This form gives detailed information about the research, which will be discussed with you. Once you understand the study, you will be asked to sign this form if you wish to participate. Feel free to discuss it with your family.

WHY IS THIS RESEARCH BEING DONE?

In this study, we will compare children and adults. Children often perform physical activity in an outdoor environment, and studies suggest that children may not handle the heat as well as adults. Apparently children do not adapt to extremes of temperature as effectively as adults, due to a differences in their body size and metabolism. Some studies have suggested that children actually heat up during exercise faster than adults and this may be related to how much they dehydrate (how much body water they lose during exercise). These older studies have been used by organizations such as the American Academy of Pediatrics to develop safety guidelines, but these older studies didn’t actually directly compare children with adults. We are interested in understanding the relationship between dehydration and a child’s ability to cool their body (called thermoregulation) and to determine if there are really differences between children and adults.

WHAT IS THE PURPOSE OF THIS STUDY?
Our main goal is to compare thermoregulation between boys and men during cycling in the heat. We want to verify the relationship (if any) between dehydration and body core temperature of boys and men during cycling in the heat.

WHO IS GOING TO PARTICIPATE IN THIS STUDY?
For this study we are recruiting a total of 15 male children aged 10-12 years old, and 15 male adults aged 19-25 years old, from the Hamilton area.

WHAT WILL MY RESPONSIBILITY BE IF I TAKE PART IN THE STUDY?
If you volunteer to participate in this study, we will ask you to visit the McMaster University Medical Centre in Hamilton on 3 occasions 4-7 days apart.

Visit	What to expect	Approximate time required
1	You will have a maximal exercise test conducted on a stationary bicycle. This exercise test (called a "VO_{2max} test") lasts 8 to 12 minutes and is used to determine aerobic fitness by breathing through a mouthpiece. Your body composition will also be determined using a machine called a dual-energy X-ray absorptiometry scanner. This scanner machine is like an X-ray. But this test delivers only 1/100th of the amount of radiation that is delivered from a typical chest X-ray. This is less than the amount of radiation you are exposed to on a daily basis from sources of natural background radiation, such as simply walking around outside. Scanning time is about 5 minutes and is painless. At this visit, a questionnaire about health will also be completed.	1 hour
2	You will perform exercise in our special climate room that we will make feel like a hot summer's day. You will cycle on a stationary bicycle for 80 minutes performed as 4, 20-minute bouts, with 10 minutes rest between each bout. We will use a mouthpiece to collect breath and verify exercise intensity. We will measure your weight and collect any urine that you can produce before or during the exercise. We will also attach special thermometers to verify your skin temperature and a flexible rectal thermometer will be used to record core body temperature. We will also monitor your heart rate.	3.0 hours
3	The same procedures as in Visit 2 will be repeated, with the only difference being the intensity of exercise that you perform. On one	3.0 hours

	occasion, you will exercise at moderate intensity and on another you will exercise at the same intensity as one of the child participants.	

WHAT ARE THE POSSIBLE RISKS AND DISCOMFORTS?

Exercise testing: The VO_{2max} test requires you to give a maximal effort. This means that you will feel tired after you are done the test. The other exercise you are asked to complete is very similar to what you might normally do as part of your daily life, especially when playing soccer or participating in all-day tournaments outside in the summer. The exercise to be done on Visits 2 and 3 will make you sweat and you will feel tired when you are done.

Exercising in the heat: The exercise in the heat will be similar to playing sports outdoors on a hot summer's day. You will probably feel tired after your 2 exercise sessions in the heat just as you would after exercising hard outside. In this study, we expect you will lose about 2% of your body weight in sweat. This is not unlike what you might experience on a typical summer day. There is the possibility that when you become dehydrated you might feel dizzy or feel like stopping the exercise. If you have these feelings, you can stop at any time. We will closely monitor your body temperature and stop the test if you get too hot. Once you are done the exercise – whether you stop because of these feelings or whether you complete the entire exercise session, we will provide fluids to drink right away.

Rectal thermometer: To measure body temperature for accuracy and safety, you will use a small, flexible rectal temperature sensor. Body temperature is a main outcome in this study and is required to be measured. Placing this sensor may make you feel embarrassed, but it is used for safety and there is really no good substitute. While positioning the thermometer is a delicate process, there is no risk of injury to you if you follow our instructions. We have used this method in numerous studies with children and young adults.

WHAT ARE THE POSSIBLE BENEFITS FOR ME?

We cannot promise any personal benefits to you from participating in this study. However, we will share the results of the study with you and talk to you about appropriate fluid intake during exercise when you are active. We will also make each exercise session fun and enjoyable. You will also learn your fitness level and body composition.

WHAT INFORMATION WILL BE KEPT PRIVATE?

All of your information will be stored in locked filing cabinets for 10 years under the supervision of Dr. Brian Timmons. We will supervise access to your information by other people in our group, only if necessary. You will be assigned a participant number, and this number will be used to identify your records. These records will be kept confidential. If the results of the study are published, your identity will remain confidential.

CAN PARTICIPATION IN THE STUDY END EARLY?
If you volunteer to be in this study, you may withdraw at any time with no prejudice. The investigator may also withdraw you from this research if circumstances arise which warrant doing so.

WHAT COSTS ARE THERE FOR PARTICIPATING IN THIS STUDY?
There will be no costs to you for participating in this study. We will give you a small honorarium for your time and effort equal to $10 for every hour spent in the laboratory.

IF I HAVE ANY QUESTIONS OR PROBLEMS, WHOM CAN I CALL?
If you have any questions about the research now or later, or if you think you have a research-related injury, you can contact Gabriela at our research office at 905-521-2100, extension 73567 or you can contact Dr. Brian Timmons directly at 905-521-2100 extension 77615.

If you have any questions regarding your rights as a research participant, you may contact Office of the Chair of the Hamilton Integrated Research Ethics Board at 905-521-2100 extension 42013.

CONSENT STATEMENT
I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to participate in this study entitled *"Thermoregulatory Responses of Boys and Men during Exercise in the Heat"*. I understand that I will receive a signed copy of this form.

____
Name of Participant

____
Signature of Participant/Date

Consent form administered and explained in person by:

________________________ __________________________
Name and title Signature/Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to participate in this research study.

____
Signature of Investigator/Date

FUTURE RESEARCH

At the end of the study, we may wish to store leftover sample for use in a future study. We will not store your sample longer than 10 years. All records identifying you will remain confidential. Information about you will not be released. If the results of the study are published, your identity will remain confidential.

CONSENT STATEMENT FOR STORAGE OF SAMPLES

I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to have my sweat and urine stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name of Participant

____
Signature of Participant/Date

Consent form administered and explained in person by:

____
Name and title

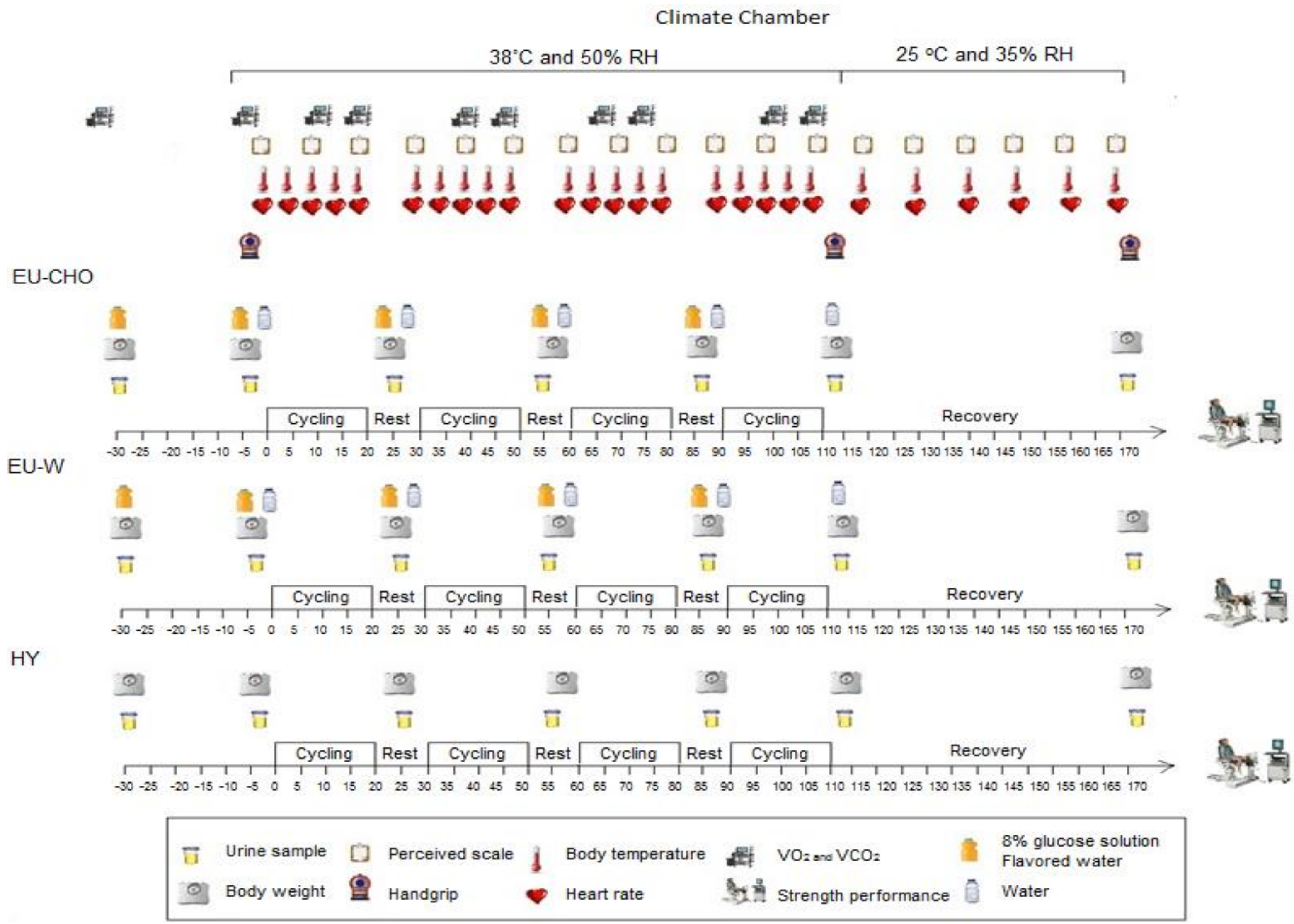
____
Signature/Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to have their urine and sweat stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name and title

____
Signature of Investigator/Date

APPENDIX G. Experimental Design


Climate Chamber
38°C and 50% RH
25 °C and 35% RH
EU-CHO
EU-W
HY
Cycling
Rest
Recovery
-30 -25 -20 -15 -10 -5 0 5 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115 120 125 130 135 140 145 150 155 160 165 170
Urine sample
Body weight
Perceived scale
Handgrip
Body temperature
Heart rate
VO2 and VCO2
Strength performance
8% glucose solution
Flavored water
Water

APPENDIX H. Consent forms Chapters 3 and 4.

Parent Information and Consent Form

Title of Study: *Effects of dehydration and substrate utilization during exercise on subsequent strength performance in boys and men*

Principal Investigator: *Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*

Local Principal Investigator: *Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*

INTRODUCTION

Your child is being invited to participate in a research study under the supervision of Dr. Brian Timmons because your child is healthy and active. In order to decide whether or not you want to be a part of this research study, you should understand what is involved and the potential risks and benefits. This form gives detailed information about the research, which will be discussed with you. Once you understand the study, you will be asked to sign this form if you wish to participate. Your child will be asked to sign another form to confirm that he agree to participate. Feel free to discuss it with your family.

WHY IS THIS RESEARCH BEING DONE?

Children often perform physical activity in an outdoor environment, and it seems children do not adapt to extremes of temperature as effectively as adults, due to a differences in their body size and metabolism. Children usually do not replace all water that they lose, mainly through sweating, during exercise, and this leads to dehydration. There are a small number of studies, with conflicting results, that investigated the impact of dehydration (how much body water they lose during exercise) on strength. We are interested in understand whether and how dehydration status affect strength performance in children and adults.

Also, we want to see how much energy children and adults are burning and how much of that energy is made up of sugar calories during exercise in the heat and how it affects performance. Therefore, participants will exercise in the heat in three identical session, except for drink availability: 1) no drink will be provide in order to induce dehydration (~2% of total body weight), 2) hydration will be maintained by providing flavoured water 2) hydration will be maintained by providing glucose drink. Sessions will happen in a random order. After the exercise in the heat participants will perform a strength performance test in equipment called isokinetic dynamometer. These results would be used in the future to provide information about the importance of hydration and glucose drinks for children and adults, and suggest mechanisms that lead differences between them.

WHAT IS THE PURPOSE OF THIS STUDY?
Our main goal is to compare the effect of dehydration (0% and 2%) on physical performance (muscle strength) in boys and men after exercise in the heat. Also, we want see how much energy they are burning and how much of that energy is made up of sugar calories during exercise in the heat.

WHO WILL BE INVITED TO PARTICIPATE IN THIS STUDY**?**
For this study we are recruiting a total of 9 male children aged 10-12 years old, and 9 male adults aged 19-25 years old, from the Hamilton area.

WHAT WILL MY CHILD'S RESPONSIBILITIES BE IF THEY TAKE PART IN THE STUDY?
If your child volunteers to participate in this study, we will ask you to bring your child to the McMaster University Medical Centre in Hamilton on 2 occasions 4-7 days apart.

Visit	What to expect	Approximate time required
1	Your child will have a maximal exercise test conducted on a stationary bicycle. This exercise test (called a "VO_{2max} test") lasts 8 to 12 minutes and is used to determine aerobic fitness by breathing through a mouthpiece. Your child's body composition will also be determined using a machine called a dual-energy X-ray absorptiometry scanner. This scanner machine is like an X-ray. But this test delivers only 1/100th of the amount of radiation that is delivered from a typical chest X-ray. This is less than the amount of radiation your child is exposed to on a daily basis from sources of natural background radiation, such as simply walking around outside. Scanning time is about 5 minutes and is painless. Your child	1.5 hour

	needs to be lying on the table during the testing and the machine will scan his body. At this visit, a questionnaire about health will also be completed. A familiarization with a strength test will be performed.	
2	Your child will perform in our special climate room. In this room, we can control the temperature and humidity and we will make it feel like a hot summer's day for this study. Children will cycle on a stationary bicycle for 80 minutes performed as 4, 20-minute bouts, with 10 minutes rest between each bout. While your child is riding our bicycle, we will have them breathe through a mouthpiece apparatus.We will measure your child's weight and collect any urine that he can produce before or during the exercise. We will also attach special thermometers that are small and attach to his skin so we can record skin temperature and a flexible rectal thermometer will be used to record core body temperature. We will also monitor his heart rate. One hour after exercise, after resting in a thermoneutral room, we will measure the leg and arm strength in equipment called dynamometer.	4.5 hours
3	The same procedures as in Visit 2 will be repeated, with the only difference being the water availability. On one occasion, we will replace fluid losses during exercise and on another no fluid will be available.	4.5 hours
4	The same procedures as in Visit 3 will be repeated, with the only difference being the availability of a sweetened beverage before and during exercise.	4.5 hours

WHAT ARE THE POSSIBLE RISKS AND DISCOMFORTS?
Exercise testing: The VO_{2max} test requires your child to give a maximal effort. This means that he will feel tired after he is done the test. The other exercise your child is asked to complete is very similar to what they might normally do as part of their daily lives, especially when playing soccer or participating in summer camp or an all-day tournament outside. The exercise and the muscle strength performance test to be done on visit 2, 3 and 4 will make your child sweat and they will feel tired when they are done and muscle ache is possible in the day after the test.

Exercising in the heat: The exercise your child will do in the heat will be similar to playing sports outdoors on a hot summer's day. Your child will probably feel tired after this exercise session in the heat just as they would after playing hard outside. In this study, we expect your child will lose about 2-3% of their body weight in sweat. This is not unlike what they might experience on a typical summer day.

There is the possibility that when he becomes dehydrated he might feel dizzy or feel like stopping the exercise. If he has these feelings, he can stop at any time. We will closely monitor his body temperature and stop the test if he gets too hot. Once he is done his exercise – whether he stops because of these feelings or whether he completes the entire exercise session, we will provide fluids to drink right away.

Rectal thermometer: To measure body temperature for accuracy and safety, your child will use a small, flexible rectal temperature sensor. Body temperature is a main outcome in this study and is required to be measured. Placing this sensor may make your child feel embarrassed, but it is used for safety and there is really no good substitute. We will ask that you help your child place the sensor in the correct position (if necessary) and we will explain exactly how to do this. While positioning the thermometer is a delicate process, there is no risk of injury to your child if you follow our instructions. We have used this method in numerous studies with children the same age as your child.

Sweetened beverage: In the sweetened beverage we will add a special marker called $_{13}C$. This marker is found naturally in foods like corn so it is completely safe. In fact, it is in all of our bodies at a concentration of about 1%. By adding a little bit of this special marker to the drink we will be able to see how much of the sugar in the drink is actually being burned for energy. We have used this method in numerous studies with children the same age as your child.

WHAT ARE THE POSSIBLE BENEFITS FOR MY CHILD?
We cannot promise any personal benefits to you or your child from his participation in this study. However, we will share the results of the study and of your son with you and talk to you about appropriate fluid intake during exercise when your child is active. We will also make each exercise session fun and enjoyable. He will also learn his fitness level and his body composition.

WHAT INFORMATION WILL BE KEPT PRIVATE?
All of your child's information will be stored in locked filing cabinets for 10 years under the supervision of Dr. Brian Timmons. We will supervise access to your child's information by other people in our group, only if necessary. Your child will be assigned a participant number, and this number will be used to identify his records. These records will be kept confidential. If the results of the study are published, your child's identity will remain confidential.

CAN PARTICIPATION IN THE STUDY END EARLY?
If you and your child volunteer to be in this study, you or your child may withdraw at any time with no prejudice. The investigator may also withdraw you from this research if circumstances arise which warrant doing so.

WHAT COSTS ARE THERE FOR PARTICIPATING IN THIS STUDY?
There will be no costs to you for participating in this study. We will reimburse you any parking expenses occurred. We will also give your child a small honorarium for his time and effort equal to $10 for every hour spent in the laboratory.

IF I HAVE ANY QUESTIONS OR PROBLEMS, WHOM CAN I CALL?
If you have any questions about the research now or later, or if you think you have a research-related injury, you can contact Gabriela at our research office at 905-521-2100, extension 73567 or you can contact Dr. Brian Timmons directly at 905-521-2100 extension 77615.

If you have any questions regarding your rights as a research participant, you may contact Office of the Chair of the Hamilton Integrated Research Ethics Board at 905-521-2100 extension 42013.

CONSENT STATEMENT
I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction and to the satisfaction of my son. I agree to allow my son to participate in this study entitled *"Effects of dehydration on strength performance in boys and men"*. I understand that I will receive a signed copy of this form.

____
Name of Participant (child's name)

____
Name of Legally Authorized Representative

____
Signature of Legally Authorized Representative/Date

Consent form administered and explained in person by:

__________________________ ____________________________
Name and title Signature/Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent for their son to participate in this research study.

____
Signature of Investigator/Date

FUTURE RESEARCH
At the end of the study, we may wish to store leftover sample (urine sample) for use in a future study. We will not store your child's sample longer than 10 years. All records identifying your child will remain confidential. Information about your child will not be released. If the results of the study are published, your child's identity will remain confidential.

CONSENT STATEMENT FOR STORAGE OF SAMPLES
I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to have my child's urine stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name of Participant (child's name)

____
Name of Legally Authorized Representative

____
Signature of Legally Authorized Representative /Date

SIGNATURE OF INVESTIGATOR:
In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to have their child's urine and sweat stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Signature of Investigator/Date

Child Assent Form

Title of Study: *Effects of dehydration and substrate utilization during exercise on subsequent strength performance in boys and men*

Principal Investigator: *Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*

Local Principal Investigator: *Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*

Why are we doing this study?
A research study is a way to learn more about people. When you exercise in warm weather, your body sweats and loses water and this can affects the muscle strength. We want to understand if after you drink different beverages or not drink at all during an exercise in the heat your muscle strength will change (increase or decrease). We want to see if exercise and hydration affects performance the same way in boys and men. Also, we want to see how the sugar in a sweetened beverage is used by your body to produce energy during exercise in the heat.

Why am I being asked to be in the study?
We are inviting you to be in the study because you are a healthy and active boy and between the ages of 10 to 12.

What if I have questions?

You can ask questions if do you do not understand any part of the study. If you have questions later that you don't think of now, you can talk to me (Gabriela) again or ask your mom or dad to call me (905-521-2100 extension 73567).

If I am in the study what will happen to me?

If you decide that you want to be part of this study, you will come visit us 4 times. When you visit us, we will do a few tests. On the first day, you will have your weight measured, your height taken and you will also be asked to ride a special bicycle to see how fit you are.

In the second, third and fourth visit you will get to ride the special bike in our special room that we will make feel like a hot day. We will write down how fast your heart is beating and how much air you are breathing when you are exercising. You will wear specials thermometers to see how hot your body gets. We will measure how hot your body gets by having you put a very thin and flexible plastic wire in your bum. We will collect a pee sample before and after you do the exercise. After exercise we will see how strong your arm and leg are.

Will I be hurt if I am in the study?

You will not be hurt if you are in the study. The exercise you will do might make you feel a little tired, but it will be like playing outside on a hot summer day. The strength performance you will do can make you feel muscle ache in the day after test. The plastic wire in your bum is used to measure how hot your body gets. This is done to make sure you stay safe while you are exercising.

Will the study help me?

If you are in the study it may not benefit you directly, but we will learn about how kids might be different than adults in how they keep their body cool during exercise. The study may help you learn how to get the most out of exercise. It will tell you and your parents how much energy you use during exercise and how fit you are.

Do I have to be in this study?

You do not have to be in this study, if you do not want to be. If you decide that you don't want to be in the study after we begin, that's OK too. Nobody will be angry or upset. It is your choice. We are discussing the study with your parents and you should talk to them about it too.

What happens after the study?

When we are finished this study we will write a report about what was learned. This report will not include your name or that you were in the study.

Assent:

If you decide you want to be in this study, please print your name. If you decide that you don't want to be in the study, even if you have started in the study, then all you have to do is tell Gabriela or Dr. Timmons that you don't want to be in the study anymore.

I,_____________________ (Print your name) would like to be in this research study.

_______________________ (Date of assent)

_______________________ _______________________

Investigator name Signature and Date

Adult Information and Consent Form

Title of Study: *Effects of dehydration and substrate utilization during exercise on subsequent strength performance in boys and men*

Principal Investigator: *Gabriela Tomedi Leites, PhD Candidate, Department of Pediatrics, McMaster University*

Local Principal Investigator: *Brian W. Timmons (PhD), Department of Pediatrics, McMaster University*

INTRODUCTION

You are being invited to participate in a research study under the supervision of Dr. Brian Timmons because you are healthy and active. In order to decide whether or not you want to be a part of this research study, you should understand what is involved and the potential risks and benefits. This form gives detailed information about the research, which will be discussed with you. Once you understand the study, you will be asked to sign this form if you wish to participate. Feel free to discuss it with your family.

WHY IS THIS RESEARCH BEING DONE?

In this study, we will compare children and adults. Children often perform physical activity in an outdoor environment, and it seems children do not adapt to extremes of temperature as effectively as adults. Children usually do not replace all water that they lose, mainly through sweating, during exercise, and it leads to dehydration. There are a small number of studies, with conflicting results, that investigated the impact of dehydration (how much body water they lose during exercise) on strength. We are interested in understand whether and how dehydration status affect strength performance in children and adults. Also, we want to see how much energy children and adults are burning and how much of that energy is made up of sugar calories during exercise in the heat and how it affects performance. Therefore, participants will exercise in the heat in three identical session, except for drink availability: 1) no drink will be provide in order to induce dehydration (~2% of total body weight), 2) hydration will be maintained by providing flavoured water 2) hydration will be maintained by providing glucose drink. Sessions will happen in a random order. After the exercise in the heat participants will perform a strength performance test in equipment called isokinetic dynamometer. These results would be used in the future to provide information about the importance of hydration and glucose drinks for children and adults, and suggest mechanisms that lead differences between them.

WHAT IS THE PURPOSE OF THIS STUDY?
Our main goal is to compare the effect of dehydration (0% and 2-3%) on physical performance (muscle strength) in boys and men after exercise in the heat. We want to verify the impact of dehydration and glucose drink in strength performance of boys and men. Also, we want see how much energy they are burning and how much of that energy is made up of sugar calories during exercise in the heat.

WHO WILL BE INVITED TO PARTICIPATE IN THIS STUDY?
For this study we are recruiting a total of 9 male children aged 10-12 years old, and 9 male adults aged 19-25 years old, from the Hamilton area.

WHAT WILL MY RESPONSIBILITY BE IF I TAKE PART IN THE STUDY?
If you volunteer to participate in this study, we will ask you to visit the McMaster University Medical Centre in Hamilton on 3 occasions 4-7 days apart.

Visit	What to expect	Approximate time required
1	You will have a maximal exercise test conducted on a stationary bicycle. This exercise test (called a “VO_{2max} test”) lasts 8 to 12 minutes and is used to determine aerobic fitness by breathing through a mouthpiece. Your body composition will also be determined using a machine called a dual-energy X-ray absorptiometry scanner. This scanner machine is like an X-ray. But this test delivers only 1/100th of the amount of radiation that is delivered from a typical chest X-ray. This is less than the amount of radiation you are exposed to on a daily basis from sources of natural background radiation, such as simply walking around outside. Scanning time is about 5 minutes and is painless. At this visit, a questionnaire about health will also be completed. A familiarization with a strength test will be performed.	1.5 hours
2	You will perform in our special climate room. In this room, we can control the temperature and humidity and we will make it feel like a hot summer’s day for this study. You will cycle on a stationary bicycle for 80 minutes performed as 4, 20-minute bouts, with 10 minutes rest between each bout. While you are riding our bicycle, we will	4.5 hours

	have you breathe through a mouthpiece apparatus We will measure your weight and collect any urine that he can produce before or during the exercise. We will also attach special thermometers that are small and attach to his skin so we can record skin temperature and a flexible rectal thermometer will be used to record core body temperature. We will also monitor his heart rate. One hour after exercise, after resting in a thermoneutral room, we will measure the leg and arm strength in equipment called dynamometer	
3	The same procedures as in Visit 2 will be repeated, with the only difference being the water availability. On one occasion, we will replace fluid losses during exercise and on another no fluid will be available.	4.5 hours
4	The same procedures as in Visit 3 will be repeated, with the only difference being the availability of a sweetened beverage before and during exercise.	4.5 hours

WHAT ARE THE POSSIBLE RISKS AND DISCOMFORTS?
Exercise testing: The VO_{2max} test requires you to give a maximal effort. This means that you will feel tired after you are done the test. The other exercise you are asked to complete is very similar to what you might normally do as part of your daily life, especially when playing soccer or participating in all-day tournaments outside in the summer. The exercise and the muscle strength performance test to be done on visit 2 and 3 will make you sweat and they will feel tired when they are done, and muscle ache is possible in the day after the test.

Exercising in the heat: The exercise in the heat will be similar to playing sports outdoors on a hot summer's day. You will probably feel tired after your 2 exercise sessions in the heat just as you would after exercising hard outside. In this study, we expect you will lose about 2% of your body weight in sweat. This is not unlike what you might experience on a typical summer day. There is the possibility that when you become dehydrated you might feel dizzy or feel like stopping the exercise. If you have these feelings, you can stop at any time. We will closely monitor your body temperature and stop the test if you get too hot. Once you are done the exercise – whether you stop because of these feelings or whether you complete the entire exercise session, we will provide fluids to drink right away.

Rectal thermometer: To measure body temperature for accuracy and safety, you will use a small, flexible rectal temperature sensor. Body temperature is a main outcome in this study and is required to be measured. Placing this sensor may make you feel embarrassed, but it is used for safety and there is really no good substitute. While positioning the thermometer is a delicate process, there is no risk of injury to you if you follow our instructions. We have used this method in numerous studies with children and young adults.

Sweetened beverage: In the sweetened beverage we will add a special marker called ^{13}C. This marker is found naturally in foods like corn so it is completely safe. In fact, it is in all of our bodies at a concentration of about 1%. By adding a little bit of this special marker to the drink we will be able to see how much of the sugar in the drink is actually being burned for energy. We have used this method in numerous studies with children and adults.

WHAT ARE THE POSSIBLE BENEFITS FOR ME?

We cannot promise any personal benefits to you from participating in this study. However, we will share the results of the study with you and talk to you about appropriate fluid intake during exercise when you are active. We will also make each exercise session fun and enjoyable. You will also learn your fitness level and body composition.

WHAT INFORMATION WILL BE KEPT PRIVATE AND CONFIDENTIAL?

E-mail is an efficient way of communication, but not completely secure. We will not to use your email information for any purpose other than communication about this research study. All of your information about this study will be stored in locked filing cabinets for 10 years under the supervision of Dr. Brian Timmons. We will supervise access to your information by other people in our group, only if necessary. You will be assigned a participant number, and this number will be used to identify your records. These records will be kept confidential. If the results of the study are published, your identity will remain confidential.

CAN PARTICIPATION IN THE STUDY END EARLY?

If you volunteer to be in this study, you may withdraw at any time with no prejudice. The investigator may also withdraw you from this research if circumstances arise which warrant doing so.

WHAT COSTS ARE THERE FOR PARTICIPATING IN THIS STUDY?

There will be no costs to you for participating in this study. We will give you a small honorarium for your time and effort equal to $10 for every hour spent in the laboratory.

IF I HAVE ANY QUESTIONS OR PROBLEMS, WHOM CAN I CALL?

If you have any questions about the research now or later, or if you think you have a research-related injury, you can contact Gabriela at our research office at 905-

521-2100, extension 73567 or you can contact Dr. Brian Timmons directly at 905-521-2100 extension 77615.

If you have any questions regarding your rights as a research participant, you may contact Office of the Chair of the Hamilton Integrated Research Ethics Board at 905-521-2100 extension 42013.

CONSENT STATEMENT

I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to participate in this study entitled *"Effects of dehydration on strength performance in boys and men"*. I understand that I will receive a signed copy of this form.

____

Name of Participant

____

Signature of Participant/Date

Consent form administered and explained in person by:

__________________________ ____________________________

Name and title Signature/Date

SIGNATURE OF INVESTIGATOR:

In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to participate in this research study.

____

Signature of Investigator/Date

FUTURE RESEARCH

At the end of the study, we may wish to store leftover sample (urine samples) for use in a future study. We will not store your sample longer than 10 years. All records identifying you will remain confidential. Information about you will not be released. If the results of the study are published, your identity will remain confidential.

CONSENT STATEMENT FOR STORAGE OF SAMPLES

I have read the preceding information thoroughly. I have had the opportunity to ask questions, and all of my questions have been answered to my satisfaction. I agree to have urine stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name of Participant

____
Signature of Participant/Date

SIGNATURE OF INVESTIGATOR:

In my judgement, the participant is voluntarily and knowingly giving informed consent and possesses the legal capacity to give informed consent to have their urine and sweat stored so it can be used in future research studies approved by the Research Ethics Board other than the one described in this information form.

____
Name and title

____
Signature of Investigator/Date

APPENDIX I. Supplementary figures Chapter 3

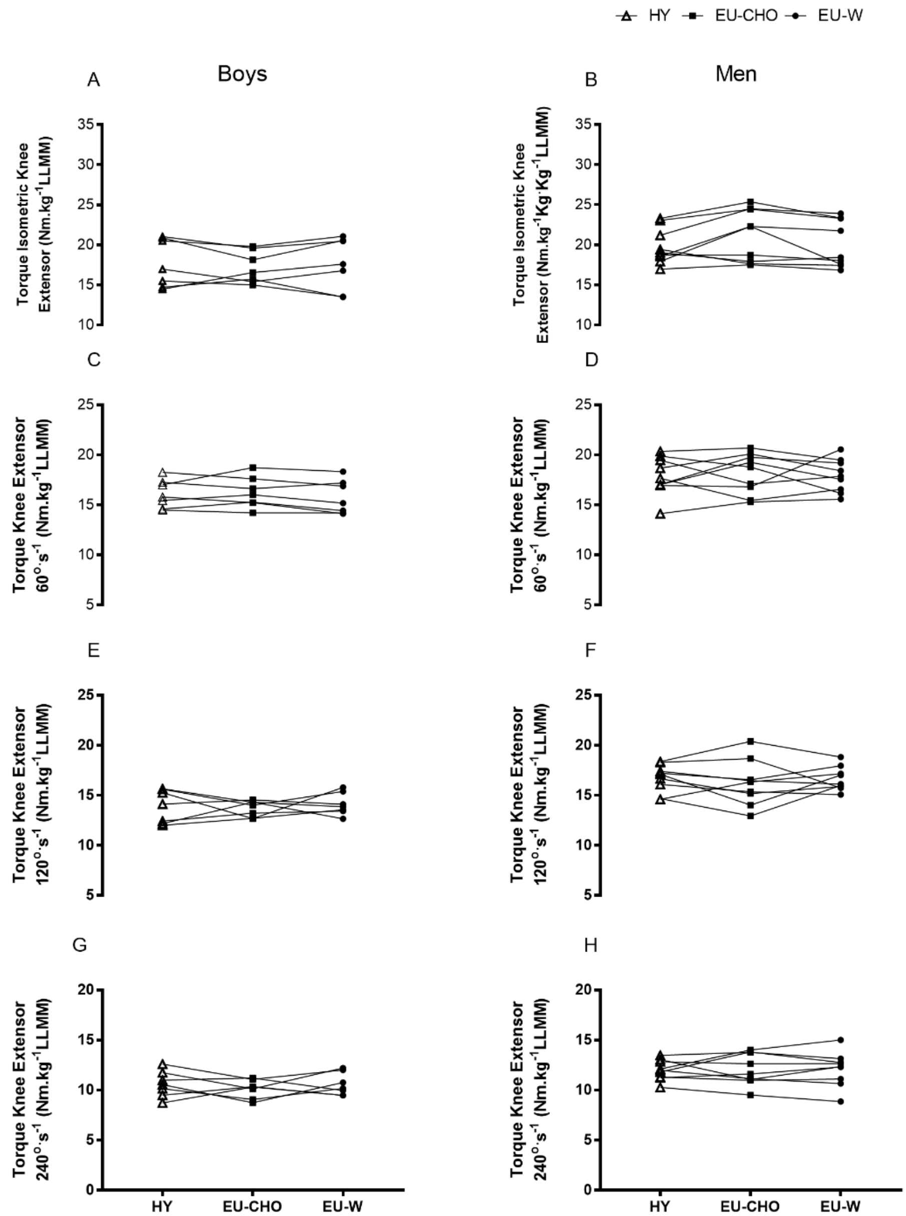


Figure F.1 Isometric and isokinetic muscle strength performance of knee extensors in boys and men during HY (∆), EU-CHO (■) and EU-W trials (●).

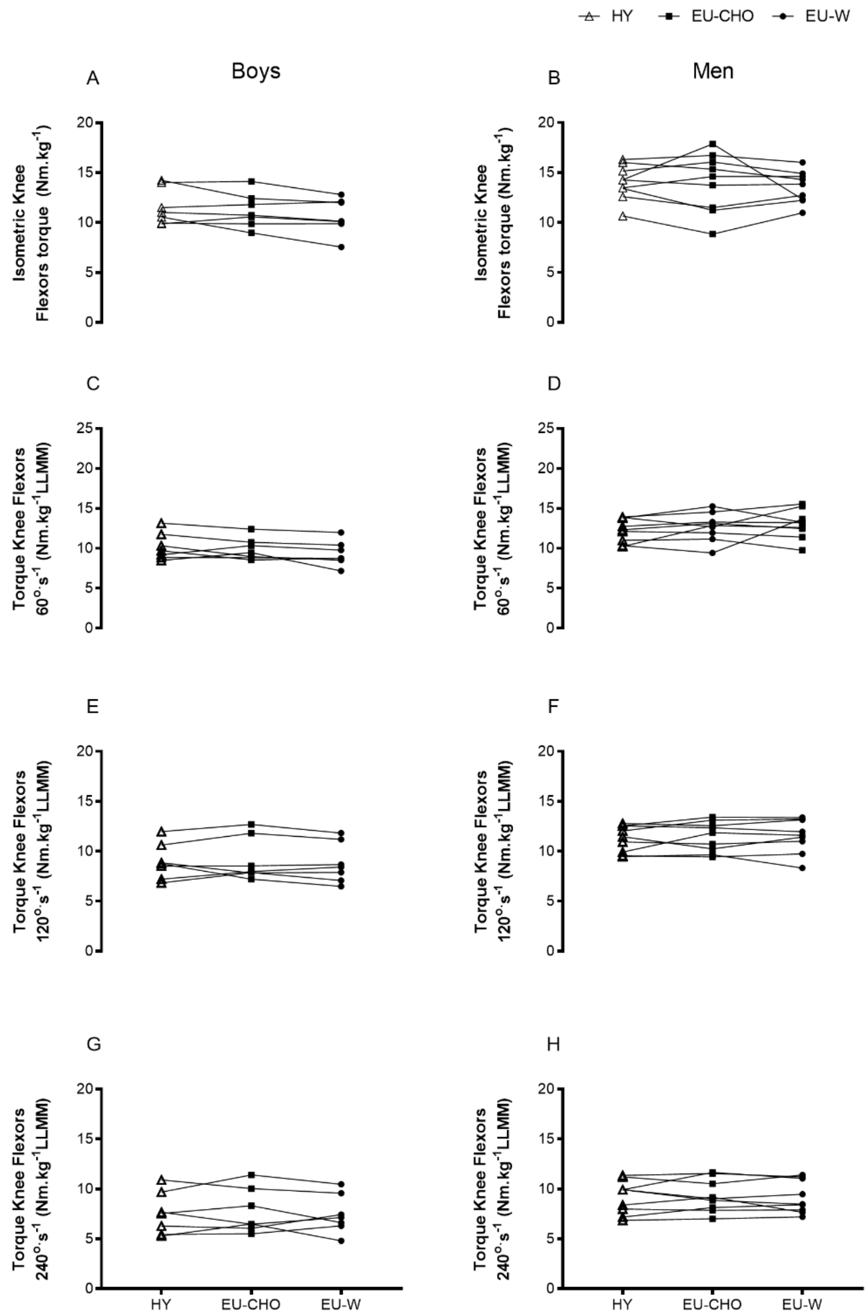


Figure F.2. Isometric and isokinetic muscle strength performance of knee flexors in boys and men during HY (∆), EU-CHO (■) and EU-W trials (●).

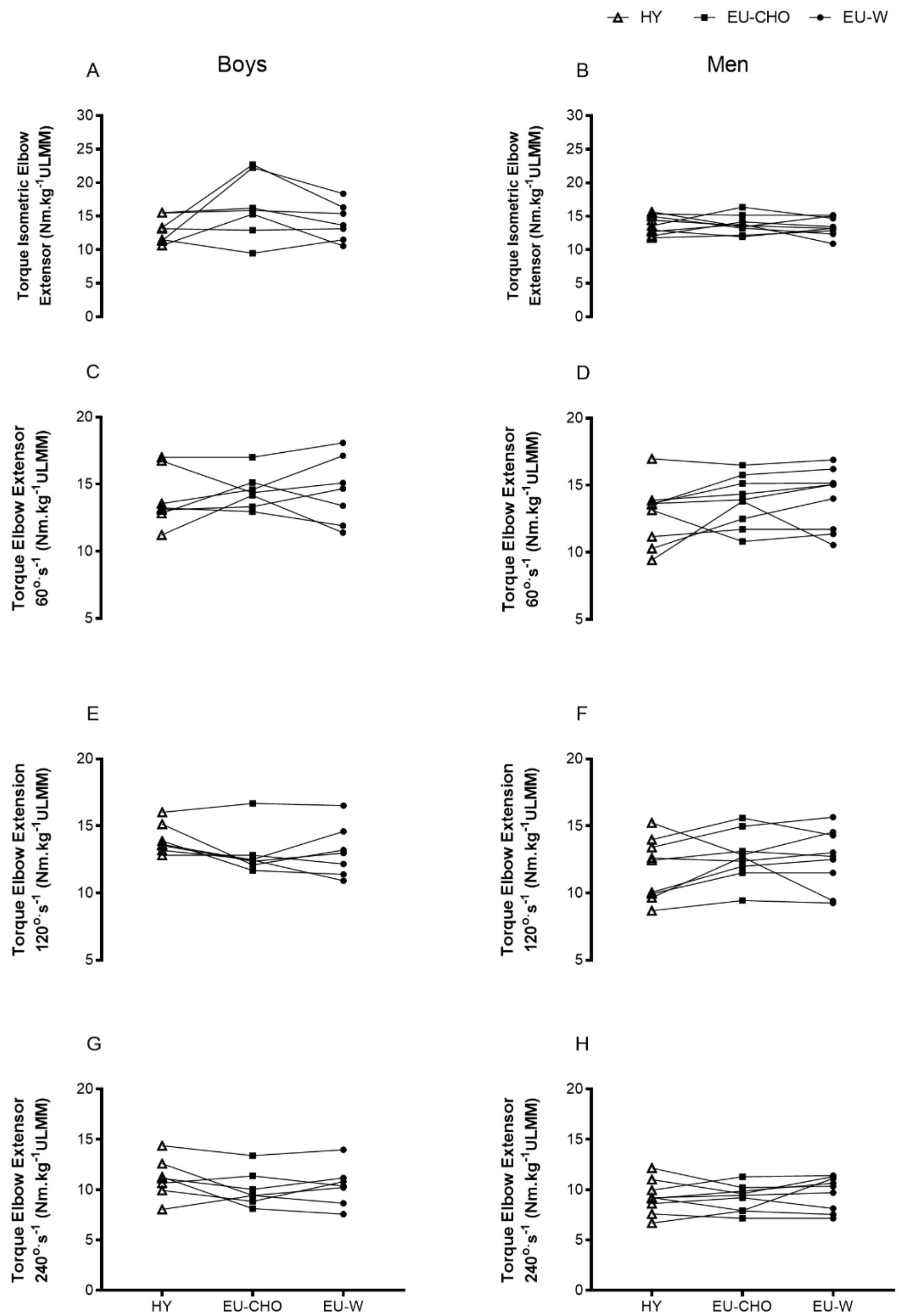


Figure F.3. Isometric and isokinetic muscle strength performance of elbow extensors in boys and men during HY (∆), EU-CHO (■) and EU-W trials (●).

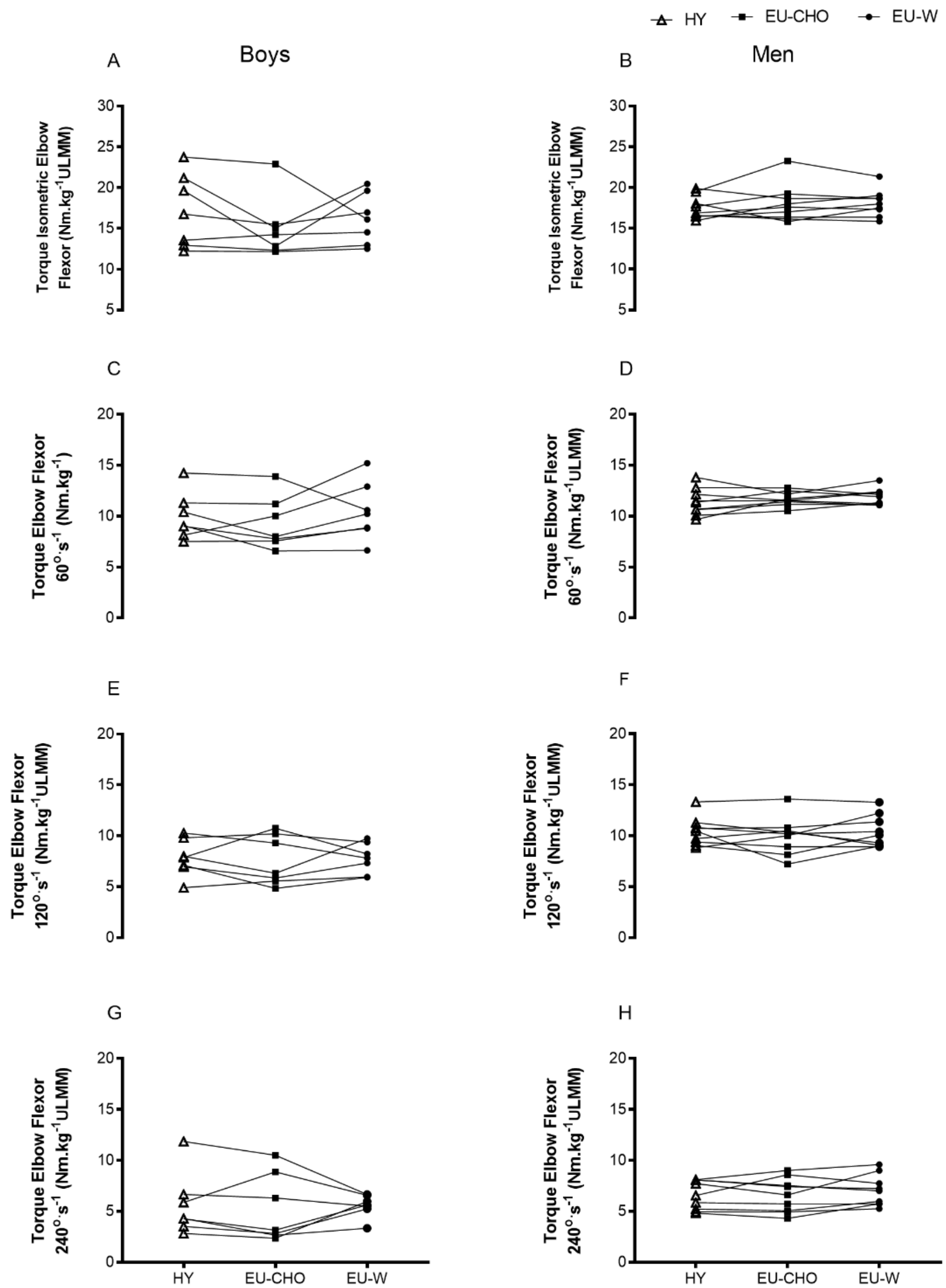


Figure F.4 Isometric and isokinetic muscle strength performance of elbow extensors in boys and men during HY (∆), EU-CHO (■) and EU-W trials (●).

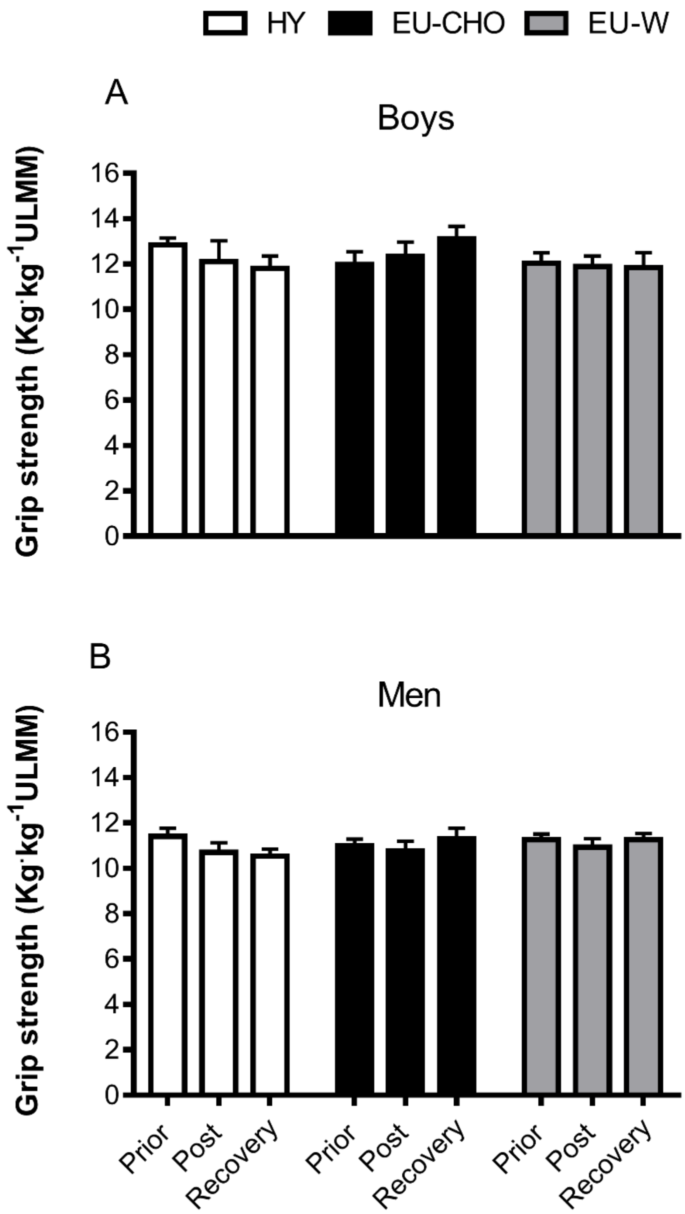


Figure F.5 Handgrip strength ($kg{\cdot}kg^{-1}ULMM^{-1}$) in boys and men prior to exercise, post exercise in the heat and post recovery in EU-CHO (black), EU-W (grey), and HY (white).

APPENDIX J. Experimental Design Chapter 4.