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EXOGENOUS FLUID DELIVERY SCHEDULE AND COMPOSITION ON FLUID  
BALANCE, PHYSIOLOGIC STRAIN, AND SUBSTRATE USE IN THE HEAT

By  
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B.S. North Central College  
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Thesis Paper

presented in partial fulfillment of the requirements for the degree of:

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## CHAPTER ONE: INTRODUCTION

### Introduction

#### *Dehydration and Physiologic Outcomes*

Fluid turnover is an unavoidable outcome across a spectrum of exercise modalities. Water turnover is dictated by self-selected behaviors of exogenous fluid ingestion and work rate, which in turn are influenced by ambient temperature and sweat rate (20). The maintenance of euhydration during exercise is a balance between exogenous fluid consumption and its availability, fluid turnover, and environmental conditions. Simultaneous exercise and fluid intake should result in relatively little to no fluctuations in body mass, replenishing only what is lost in the sweat (50). Ideally, concurrent exercise and fluid consumption will avoid dehydration by  $\geq 2\%$  of body weight (11,41,42,48,76).

Collectively, past research has shown that compromised hydration has led to both impaired (11,42,46) and unaffected exercise performance (11,23,42). Kenefick et al. (42) demonstrated that when individuals were dehydrated by 4% of body weight, there was a 5% performance decrement during a cycling time trial in a temperate environment when compared to a euhydrated time trial. However, when individuals are kept cool or exercise in a cool environment, performance decrements often seen when hypohydrated are attenuated (11,42)

Evaporative heat loss from the skin, which relies on sweat, is the primary driver of heat dissipation during exercise at ambient temperatures above approximately 20°C, overtaking dry heat loss mechanisms (conduction, convection, and radiation) (57). Due to the reliance on sweating to dissipate heat, a large proportion of lost fluids during

exercise (50,76). As exercise intensity or duration increases, the need for heat dissipation increases or is sustained, thereby requiring an increase in sweat production, leading to subsequent fluid loss (57). Additionally, graded hypohydration levels have been shown to decrease sweat rates (77), wherein the ability to off load heat is attenuated which can contribute to increases in physiologic strain.

Fluid loss without adequate exogenous fluid consumption during exercise can contribute to increases in physiologic strain when compared to euhydrated states. Sweat production greater than exogenous fluid intake is associated with decreases in plasma volume (29,32,36,42,44,46,77,78,83). The plasma volume decreases incurred may lead to increases in cardiovascular strain, causing reductions in stroke volume (29,31,32,42), blood pressure (29,32) and systemic blood flow (31) when compared to euhydrated states. González-Alonso et al. (32) demonstrated that dehydrating an individual by 4% of their body weight during a 3-hour cycling intervention generated a 7-8% reduction in stroke volume and a 5  $\text{beat}\cdot\text{min}^{-1}$  increase in heart rate (HR) when compared to a euhydrated control trial. Markers of physiologic strain index (PSI), HR (2,11,29,31,42,46) and core temperature ( $T_c$ ) (2,29,42,46), are exacerbated when dehydrated and working at constant workloads. Adams et al. (2) displayed augmented PSI in both temperate and hot environments when subjects hypohydrated by 1-2% of their body weight walked on a treadmill with a 45 lb. rucksack for 90 minutes at 50%  $\text{VO}_2$  max when compared to a euhydrated state.

In general, close maintenance of baseline euhydration status can help serve to attenuate unfavorable physiologic alterations that would otherwise exacerbate PSI and blunt heat offloading during exercise. Consumption of fluids to closely match individual

losses then becomes a component of prudent practice during extended exercise in the heat.

### *Common Fluid Intake Recommendations*

Exogenous fluid intake functions on the premise that fluids are available, accessible, and actively ingested. *Ad libitum* intake strategies are not always effective in the maintenance of body weight over an extended exercise bout, often leading to decreases in body weight upwards of 4% (22,23,35). When *ad libitum* fluid consumption was compared to programmed fluid consumption during a treadmill half marathon, individuals consumed 3.5 times more fluid with a programmed amount, maintaining body weight within 1% of pre-exercise weight (23). Greenleaf and Sargent II (35) showed that heat and hypohydration had larger influences on *ad libitum* fluid consumption than exercise, indicating a decrease in thirst sensitivity while exercising. Decreased thirst sensitivity during extended exercise could contribute to lesser amounts of fluids consumed *ad libitum*, despite availability and accessibility, showcasing that more programmed delivery schedules could aid in euhydration maintenance.

Exogenous fluid intake recommendations vary as a function of heat exchange factors in reference to the environment (humidity, temperature, wind), work rate, and exposed surface area (1,47,50,76). These recommendations also vary further based upon sport, occupation, and individual sweat rates (50,76). Collectively, the American College of Sports Medicine (ACSM), National Athletic Trainers' Association (NATA), and the United States Army training doctrine (TRADOC) regulation 350-29 recommend consuming exogenous fluids to prevent dehydration  $\geq 2\%$  of body weight (1,50,76).

The ACSM's suggested strategy advises consuming fluids *ad libitum*, amounting to volumes between 400 and 800 ml·h<sup>-1</sup>, dependent upon work rate, environment, clothing, and body weight (76). The NATA suggests that athletes should be aware of their individual sweat rates as a function of their sport, suggesting 100% replacement of estimated fluid loss for less than a 2% body weight decrease (50). Military fluid intake guidelines are based upon a matrix of 15 conditions in reference to 5 colored flag temperatures (white, green, yellow, red, and black) and 3 work-rate categories (easy, moderate, and hard), with fluid consumption assigned in reference to the corresponding environment, work task, and equipment worn (1,47). 15 conditions yield a wide range of acceptable fluid intake volumes, ranging from 0.5 to 1 quart·h<sup>-1</sup> (470 to 940 ml·h<sup>-1</sup>) with the stipulation as not to exceed either 1.5 quarts·h<sup>-1</sup> (1400 ml·h<sup>-1</sup>) or a total of 12 quarts·day<sup>-1</sup> (11,300 ml·day<sup>-1</sup>) (1,47). Fluid intake recommendations from these agencies are designed to limit dehydration to no more than 2% of body weight, precipitating a wide range of acceptable fluid intake volumes. However, the interval between doses that complies with total hourly intake is unclear.

#### *Fluid Volume and Administration Manipulation*

Varying intake volumes may have differing effects on urinary output and subsequent net fluid balance (41,75,84). Shirreffs et al. (84) found that a bout of cycling in the heat paired with immediate fluid intake of assorted volumes yielded proportional urine outputs during a 6-hour recovery period, while net fluid balance did not consistently improve with higher volumes of consumed fluid. Jones et al. (41) evaluated post-exercise rehydration efficacy of a metered (2 doses·h<sup>-1</sup> for 4 hours) and bolus dosing (1 dose at the beginning of the 4 hour recovery) of plain water equal to 100% of

fluid lost during exercise in the heat, finding a 20% increase in net fluid balance in the metered dosing over an 8-hour recovery period. In both the Shirreffs et al. (84) and Jones et al. (41) studies it is clear that inappropriate volumes or administration technique of exogenous fluid (e.g. too little volume, too much volume, and or rapid administration) can still leave individuals in a hypohydrated state following a bout of exercise. Together, these studies suggest that manipulating intake volume and timing of administration can elicit independent effects on fluid balance, underscoring the importance of exogenous fluid delivery methods.

### *Gastric Emptying and Hormonal Response*

The volume and timing of the fluid consumption can manipulate gastric emptying rates (15,53,69), hormonal responses (84,86), and subsequent renal function (86). Costill and Saltin (14) showed proportional increases in gastric emptying following consumption of  $\geq 600$  ml of fluid, wherein greater volumes demonstrate comparable gastric emptying rates. Larger volumes of ingested fluids contribute more to the stomach's total volume (inclusive of gastric secretions and saliva), provoking volume dependent rates of gastric emptying (14,53), resulting from intragastric pressure and stomach distension increases (61). Volume dependent gastric emptying persists during exercise, where 2 hours of cycling at 70%  $\text{VO}_2$  max displayed corresponding emptying rates of high (23  $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ), medium (17.1  $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ), and low (11.5  $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ) fluid volumes administered at 15-minute intervals (53).

Since little to no fluid is absorbed in the stomach, the majority of fluid absorption occurs in the small intestine (70), indicating that absorption processes are delayed until gastric emptying has occurred (79). The ensuing net fluid movement across intestinal



walls into circulation passively occurs via aquaporins as a function of osmotic, hydrostatic, and filtration pressures (37). Once gastric emptying has occurred, absorption can relatively quickly in the small intestine. Reitemeier et al. (70), using isotopically labeled water, found that a mean time of 7 minutes was required for 67% of water to be absorbed from the small intestine into the arterial blood.

Absorption of fluid and solutes into the arterial blood subsequently influences hormonal release, responding to changes in blood volume (84), blood pressure (33,34), and plasma osmolality (13,55). Release of hormones vasopressin, angiotensin II, and aldosterone influence fluid retention at the renal level (4). Shirreffs et al. (84) found that blood volume increased as function of the fluid volume consumed following exercise, where vasopressin (2 hours post exercise), angiotensin II (2 and 6 hours post exercise), and aldosterone (2 and 6 hours post exercise) were elevated during low fluid consumption trials when compared to high fluid trials. Gonzalez-Villalobos et al. (33,34) demonstrated in angiotensin-converting enzyme knockout mice that blood pressure changes and ensuing renal response is associated with the formation of angiotensin II. Montain et al. (55) showed across a variety of treadmill exercise intensities (25%, 45%, and 65% of  $VO_2$  max) and hypohydration levels (0%, 3%, and 5% of body weight) that increases in vasopressin were closely related with rises in plasma osmolality, while Thompson et al. (92,93) quantifies a threshold of approximately 285 mOsm as the point in which vasopressin begins to rise.

#### *Fluid Composition Manipulation*

Varying exogenous fluid compositions may have additional effects on fluid retention and subsequent net fluid balance (18,81,83-85). Gastric emptying is further

dependent upon fluid composition, where plain water (hypotonic) empties at faster rates than both energy dense and hypertonic solutions (14,69). In general, ingesting a hypotonic solution in a large volume would serve to increase urine production (83,84,86), which would be counter to fluid retention. Seery and Jakeman (81), following dehydration by 2% of body weight, showed that administration of a carbohydrate-electrolyte solution at a volume equivalent to 150% of fluids lost from exercise can help bring post exercise hydration status towards baseline more effectively than plain water. Similarly, Shirreffs et al. (83), evaluated the rehydration efficacy of 4 commonly consumed beverages (carbohydrate-electrolyte solution, carbonated apple beverage, mineral water, and plain water), finding that net fluid balance was more closely restored to baseline levels following consumption of a carbohydrate-electrolyte solution. Shirreffs et al. (84), following exercise induced dehydration by 2% of body weight, displayed proportional increases in urine output as low sodium solutions were administered in incrementally higher fluid loss by body weight percentages (50%, 100%, 150%, and 200%). In general, beverages with higher sodium concentrations tend to decrease urine output and provoke more favorable hydration statuses (75). The other beverages, with their inadequate carbohydrate and sodium compositions, did not effectively restore baseline hydration status.

### *Carbohydrates and Performance*

Carbohydrate-electrolyte solutions, formulated with higher energy densities and electrolytes, in addition to helping restore fluid balance (81,84), also help maintain carbohydrate oxidation (10,12,15,49,53,62,78), promoting subsequent endurance exercise performance (12,49,80). Carbohydrate is often administered at 10-15-minute

intervals during exercise (10,15,39), at a recommended rate between 30-60 g·h<sup>-1</sup> (91) to mitigate sodium-glucose transporter saturation (27). Curvilinear dose-response relationships have been elucidated, where a carbohydrate delivery rate > 78 g·h<sup>-1</sup> produces diminishing performance returns (89).

However, manipulating the administration frequency of identical amounts of a carbohydrate-electrolyte solution does not appear to impact total work output. Schweitzer et al. (80) administered equal total dosage volumes of a carbohydrate-electrolyte solution (75 g) at differing intervals during 2 hours of cycling, providing a total of 4 doses·h<sup>-1</sup> for 2 hours. Trials differed by providing fluids continuously, front-loaded (4 doses·h<sup>-1</sup> for the first hour, placebo second hour), or end-loaded (placebo first hour, 4 doses·h<sup>-1</sup> for the second hour). Despite differences in the timing of the carbohydrate-electrolyte solution, total work output did not differ during a 15-minute cycling time trial.

### *Renal Response to Exercise*

The volume and solutes of liquids crossing the intestinal walls into circulation manipulate the glomerular hydrostatic pressure at the renal level, dictating how much fluid and solute may cross the filtration membrane (4). At rest, glomerular filtration rate (GFR) is approximately 120 ml·min<sup>-1</sup> (26,68), incrementally decreasing as a function of increasing exercise intensity. Freund et al. (26) demonstrated a GFR decrease to less than 80 ml·min<sup>-1</sup> when subjects cycled at 80% of VO<sub>2</sub> max for 20 minutes. Distribution of cardiac output during exercise similarly mirrors GFR perturbations. At rest, cardiac output is approximately 5 L·min<sup>-1</sup> where 20% is distributed to the kidneys, while heavy exercise warrants blood flow redistribution to working tissue with a cardiac output increase to > 20 L·min<sup>-1</sup> and approximately 3% distribution to the kidneys (5). Decreases

in GFR (26,68), redistribution of blood flow (5), and exercise dependent hormonal increases (26,55,64,90) work to preserve net fluid balance.

### *Fluid Demands in the Field*

Fluid turnover and hydration demands have been documented during field type scenarios using stable isotope tracer methodologies (19,20,54,72-74). Field type research studies are able to capture the relationship that the ambient environment and workload/type have on fluid turnover. These studies showcase large fluid demands during wildland firefighter work shifts ( $6.7 \pm 1.4$  (73) ,  $7.8 \pm 1.6$  (71) , and  $9.5 \pm 1.7$  L·day<sup>-1</sup> (20)), the Kona Ironman World Championships (16.6 L over 10.7 hours) (19), the Western States 100 Ultramarathon (18.2 L over 26.8 hours) (72), and the Badwater 135 Ultramarathon (35.2 L over 45.1 hours) (72). These field studies, while excellent in quantifying overall fluid demands, are unable to confidently assert the relationship between those fluid demands, euhydration status, and administration strategies that make up the hourly intake. Together, the need is outlined for a comprehensive lab-based intervention.

### **Problem**

The hydration position stands from ACSM, NATA, and the military highlight a range of potentially acceptable hourly or individualized fluid volumes (1,47,50,76). Yet, the large spectrum of recommendation from these stances may make it difficult to adopt a more optimal strategy. Individual interpretation of these recommendations could result in a spectrum of hydration strategies, and more often, end user confusion or misinterpretation (48,96,97). Fluid recommendations are often prescribed on the basis of temperature, humidity, activity, work rate, calculated or perceived sweat rate, and

clothing worn. The volumes are outlined as an amount per hour or percentage of body weight lost, but the particularities of what strategy to use when consuming the fluids are unclear.

Hydration and rehydration strategies function on the premise that exogenous fluids are available, consumed, and adequately absorbed. *Ad libitum* fluid intake lends itself to the possibility of inadequate maintenance of net fluid balance during exercise (22,23,35). For fluid administration to be advantageous, manipulation of volume, frequency, and composition appears necessary to promote euhydration as opposed to high urine output resulting from rapid gastric emptying rates (41,81,83-86).

Manipulating the rate at which fluid exits the stomach into the small intestine could be advantageous in euhydration maintenance during exercise. Moreover, while volume, frequency, and composition likely combine to alter fluid availability, each factor may also exert effects separately. An intervention manipulating fluid volume intake has the potential to alter gastric emptying, renal filtration processes, and hormonal responses, allowing for perhaps a more efficient approach to fluid delivery during extended exercise bouts by minimizing expected urinary outputs.

Prior research on hydration and rehydration strategies have examined the effects of fluid composition (18,78,81,83-85), fluid temperature (36), and fluid administration frequency (41,44,81) both during and following exercise. However, fluid recovery strategies that combine elements of volume consumption and composition appear most advantageous. Jones et al. (41), dehydrated subjects by 2% of body weight with intermittent treadmill exercise and then rehydrated subjects with plain water using a frequent metered or an infrequent bolus dosing strategy, both equaling 100% of weight

lost. Post exercise rehydration using a frequent metered dosing elicited a 20% greater net fluid balance than the opposing bolus dosing strategy. Similarly, Shirreffs et al. (83), using a combination of beverages with different carbohydrate and sodium contents demonstrated that urinary output was minimized to improve net fluid balance was more when solutions with higher sodium and energy densities were included in the rehydration plan. Implementation of post-rehydration strategies of administration interval and composition to an exercise-based scenario could be advantageous for euhydration maintenance. It is presently unclear how altered fluid delivery protocols (frequent micro-dosing versus infrequent bolus-dosing) may influence physiologic responses (fluid retention, PSI, blood glucose, and serum electrolytes) during extended exercise in the heat.

### **Purpose & Hypothesis**

The primary focus of this study was to target administration frequency of equal total volumes as the sole strategy for altering hydration status during and following exercise. The purpose of the present study was to determine the effects of altered fluid delivery schedules (frequent micro-dosing vs. infrequent bolus-dosing) on fluid retention, and heat stress during extended exercise. A secondary focus was to evaluate the combination of micro-dosing a carbohydrate-electrolyte solution during an identical exercise condition. Steady state expired gas provided insight into substrate utilization as a function of fluid dose interval and composition. Further calculation of estimated glomerular filtration rate (eGFR) may give further insight into renal hemodynamics as they are theoretically altered by exercise, fluid volume and composition dosages.

We hypothesized that micro-dosing fluids in more frequent amounts would improve overall fluid retention, regardless of fluid composition, while the inclusion of a carbohydrate-electrolyte solution would further improve fluid retention both during and following exercise. We also hypothesized that micro-dosing a carbohydrate-electrolyte solution would maintain a higher carbohydrate oxidation rate.

### **Primary Null Hypotheses**

#### *Water: Micro vs Bolus Dosing*

1. There will no difference in body weight loss during or post exercise between the trials.
2. There will be no difference in urine production over time between the trials.
3. There will be no difference in the exercise sweat rate between the trials.
4. There will be no difference in urine specific gravity during exercise and recovery between the trials.
5. There will be no change in plasma osmolality before or following exercise between trials.
6. There will be no difference in core temperature response to exercise trials over time.
7. There will be no difference in skin temperature response to exercise between trials.
8. There will be no difference in heart rate response to exercise between trials.

#### *Carbohydrate-Electrolyte: Micro vs Bolus Dosing*

1. There will no difference in body weight loss during or post exercise between the trials.

2. There will be no difference in urine production over time between the trials.
3. There will be no difference in the exercise sweat rate between the trials.
4. There will be no difference in urine specific gravity during exercise and recovery between the trials.
5. There will be no change in plasma osmolality before or following exercise between trials.
6. There will be no difference in core temperature response to exercise trials over time.
7. There will be no difference in skin temperature response to exercise between trials.
8. There will be no difference in heart rate response to exercise between trials.

### **Secondary Null Hypotheses**

#### *Water: Micro vs Bolus Dosing*

1. There will be no difference in calculated carbohydrate oxidation during exercise between trials.
2. There will be no difference in estimated glomerular filtration rate between trials.

#### *Carbohydrate-Electrolyte: Micro vs Bolus Dosing*

1. There will be no difference in calculated carbohydrate oxidation during exercise between trials.
2. There will be no difference in estimated glomerular filtration rate between trials.

### **Significance and Rationale**

Quantifying the efficacy of a novel micro-dosing hydration strategy opens the possibility for making current fluid intake recommendations while exercising



unambiguous. Distinctly identifying what strategy to use when consuming fluids during exercise can use gastric emptying, renal filtration, and hormonal responses advantageously.

Micro-dosing may additionally give way to carrying less fluid volume while exercising, eliminating dependence on access dependent *ad libitum* drinking strategies that are not always effective in keeping athletes euhydrated during exercise. The end goal from this study would be to develop recommendations, systems, or applications that make fluid intake more intuitive and euhydration easily maintainable over a prolonged bout of exercise.

### **Delimitations**

1. Subjects drank 500 ml of water 1 hour before going to bed the night before and 30 minutes before all trials.
2. Subjects kept and were instructed to follow their food and fluid consumption log.
3. Standardized temperature (33°C) and relative humidity (30%) across trials.
4. Standardized clothing and pack weight across trials.
5. Individualized fluid intake predicted from familiarization trial, equaling 100% of fluid lost.
6. Standardized fluid temperature across trials (33°C).
7. Absolute treadmill intensity of  $1.4 \text{ m}\cdot\text{s}^{-1}$  ( $3.0 \text{ miles}\cdot\text{hour}^{-1}$ ) at a 5% grade.
8. Counterbalanced trial order.
9. Trials of the same composition will be separated by 7 days and there will be a 2-week wash out period between fluid compositions to eliminate heat acclimation and trial interaction.

10. All trials will begin at approximately the same time.

### **Limitations**

1. Subjects' activity, food intake, fluid intake, and behavior cannot be controlled outside of the laboratory.
2. Convenience sampling from the university and surrounding community.
3. Insensible water loss.
4. "Salty" v "unsalty" sweaters
5. Subject experience walking with a loaded pack.
6. Total climate control in the heat chamber will not reflect unpredictable environmental changes that free-range humans can experience.
7. Constant work rate does not mimic real life work:rest ratios.

### **Assumptions**

1. Consumption of 500 ml of water the night before and the morning prior to each trial will ensure subject euhydration (2,29-31,36,41,42,46,81,83,85), where Reitemeier et al. (70) demonstrated that 67% of water was absorbed into the arterial blood after 7 minutes.
2. Euhydration prior to each trial assumes that plasma osmolality is within a range (approximately 280 mOsm) that does not excessively provoke antidiuretic hormone activity (55,86).
3. Expected metabolic load of 22-25 ml·kg<sup>-1</sup>·min<sup>-1</sup>, predicted from the Pimental and Pandolf (65) equation. A relative VO<sub>2</sub> max of 45 ml·kg<sup>-1</sup>·min<sup>-1</sup> yields an approximate intensity of 48-55% VO<sub>2</sub>, wherein Freund et al. (26) demonstrated an approximate GFR of 100 ml·min<sup>-1</sup>. This intensity is well below the 80% VO<sub>2</sub>

max limitation in the Chronic Kidney Disease-Epidemiology Equation (CKD-EPI) demonstrated by Poortmans et al. (66) and within the low bias range outlined by Levey et al. (45).

4. Metabolic load ( $22\text{-}25 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ) and chamber temperature ( $33^{\circ}\text{C}$  and 30% RH) combination is not expected to promote excessive hormonal activity (26,55,90).

## **CHAPTER TWO: REVIEW OF LITERATURE**

### **Dehydration**

#### *Defined, Hydration Position Stands, and Measurement Techniques*

The ACSM and NATA refer to dehydration as a loss of body water, hypohydration as body water deficits resulting from dehydration, and euhydration as “normal” body water content (50,76). Fluid loss can manifest in the form of sweat (50,76), obligatory urine production (50,85), feces (50), vomiting (50), and respiratory water loss (52,76). ACSM, NATA, and the United States Army TRADOC regulation 350-29 collectively recommend consuming exogenous fluids to prevent dehydration  $\geq 2\%$  of body weight (1,50,76).

Hydration status can be quantified through a variety of physiologic and bodily measures, giving insight into water turnover, water loss, and electrolyte loss. The literature often uses plasma volume (36,78,83), plasma osmolality (38,44,81), hematocrit (38,44,78), hemoglobin (38,44,78), urine specific gravity (USG) (2,18,36,48), urine osmolality (2,38,73,83,85), urine color (2), and body weight (2,18,36,41,44,78,81,83,85) to objectively measure hydration status. Stable isotope tracer methodologies have also

been employed during field-based scenarios to quantify water turnover and fluid needs assessments (19,20,54,72-74).

### *Negative Physiologic Outcomes Associated with Dehydration and Hypohydration*

Evaporative heat loss from the skin, which relies on sweat, is the primary driver of heat dissipation during exercise at ambient temperatures above approximately 20°C, overtaking dry heat loss mechanisms (conduction, convection, and radiation) (57). Due to the reliance on sweating to dissipate heat, a large proportion of lost fluids during exercise (50,76) and compounded with inadequate exogenous fluid intake leads to decreases in plasma volume (29,32,36,42,44,46,77,78,83). There is a linear relationship between the change in extracellular fluid space and the change in plasma volume, which occurs as a function of free water loss (sweat, urine, etc.) (59).

Dehydration can lead to increases in cardiovascular strain through decreases in plasma volume (29,32,36,42,44,46,77,78,83), reducing stroke volume (29,31,32) and blood pressure (29,31,32) in comparison to euhydrated states during exercise at constant workloads. A blood volume drop of approximately 200 ml has been identified as a threshold value at which stroke volume decreases begin to occur (32), while Sawka et al. (77) found approximate plasma volume decreases of 150-300 ml in their hypohydrated by 3% of body weight trials.

During heavy exercise, a large proportion of cardiac output (80-85%) typically gets diverted to working tissue (5). However, decreases in plasma volume that lead to drops in perfusion pressure and cardiac output due to stroke volume decreases may lead to a decline in blood flow to working muscles (31). González-Alonso et al. (29,32) demonstrated that dehydrating an individual by  $\geq 4\%$  of their body weight during a

heated cycling intervention can generate reductions in stroke volume that contribute to cardiac output decrements up to 18%. During a cycling intervention to volitional exhaustion, subjects became dehydrated by approximately 4% of body weight, wherein blood flow to the exercising leg muscles decreased by 13% towards the end of the protocol when compared to a euhydrated trial of identical length (31). Hypohydrated exercise in temperate environments (20°C) has also been shown to induce cardiovascular strains similar to exercise in the heat that become further amplified when heat and hypohydration are superimposed on one another (2,32).

PSI metrics of HR (2,11,29,31,42,46) and  $T_c$  (2,29,42,46) have shown to be exacerbated when exercising hypohydrated or achieving incremental dehydration resulting during prolonged exercise. Beginning exercise hypohydrated in both temperate (32,42) and cold (42) environments can increase HR and  $T_c$  when compared to an identical exercise modality in a euhydrated state. Adams et al. (2) displayed increased PSI metrics when subjects walked on a treadmill with a 45 lb. rucksack hypohydrated in a temperate environment when compared to a euhydrated state. Kenefick et al. (42) dehydrated individuals by 4% of body weight and displayed increased HR and  $T_c$  across a range of ambient temperatures (10°C, 20°C, 30°C, and 40°C) during 30 minutes of cycling at 50%  $\dot{V}O_2$  max when compared to identical euhydrated trials. Similarly, Chevront et al. (11), following 3-hours of passive dehydration in a hot (45°C) environment, utilizing a similar protocol as in the Kenefick et al. (42) study, also displayed HR and  $T_c$  potentiation when hypohydrated in cold (1°C) and temperate (20°C) environments compared to euhydrated trials in identical environments. PSI may additionally be elevated at rest prior to commencing exercise when hypohydrated (2,42).

### *Effects of Dehydration and Hypohydration on Performance*

Past research has shown that compromised hydration status has led to impaired (11,42,46) and unaffected exercise performance (11,23,42). Dehydration statuses  $\geq 2\%$  of body weight has frequently been cited as a threshold wherein athletic performance can be negatively impacted (11,42,46,48,50,76). However, alterations in ambient temperature (11) or skin temperature (42) can help maintain or attenuate decreases in exercise performance.

Logan-Sprenger et al. (46) found that beginning a cycling time trial (intensity of 6 kJ·kg BM<sup>-1</sup>) hypohydrated by 2% of body weight displayed a 13% performance decrement, producing an elevated T<sub>c</sub>, HR, and plasma osmolality compared to a euhydrated trial, despite cycling at a lower total power output. Cheuvront et al. (11) passively dehydrated subjects by 3% of body weight prior to administering a 30-minute cycling performance trial, finding that performance was impaired in a temperate environment (20°C) by 7.6% when compared to a euhydrated baseline trial. Similarly, Kenefick et al. (42) found that when subjects began cycling hypohydrated by 4% of body weight there was a graded performance decrement (3%, 5%, 12%, and 23%) across a variety of ambient temperatures (10°C, 20°C, 30°C, and 40°C) when each trial was compared to a euhydrated trial.

Cheuvront et al. (11) and Kenefick et al. (42) demonstrated that performance decrements associated with hypohydration (3-4% of body weight) were attenuated to approximately 3%, remaining closer to euhydrated baseline trials when ambient temperature (2°C) and or skin temperature were kept cool (<29°C). Cuddy et al. (17) found that when ambient temperature was kept cooler (18°C and 26°C), time to

volitional exhaustion during a treadmill intervention was 13% and 22% longer in comparison to a warmer temperature (42°C) as a result of a more favorable core to skin temperature gradient. Dion et al. (23), during a treadmill half marathon, showed no performance advantage in maintaining euhydration when subjects consumed exogenous water following a drinking programmed designed to minimize fluid loss to < 2% of body weight when compared to *ad libitum* drinking. During the *ad libitum* drinking trial subjects finished dehydrated by 3% of body weight, wherein mean finish time, pace, and speed did not differ between *ad libitum* drinking and programmed drinking strategies. Contrarily, incremental dehydration by 1.8%-3% of body weight post marathon has been associated with faster finishing times, where those who finished under 3 hours were more hypohydrated than the individuals who required greater than 4 hours to finish (98). At least in the marathon, some mild degree of body weight loss from dehydration during the event may have a positive performance benefit.

## **Fluid Volume**

### *Effects on Gastric Emptying*

The volume and timing of fluid consumption can manipulate gastric emptying rates (15,53,69). A fast phase is followed by a slow gastric emptying phase (69). Larger volumes of ingested fluids contribute more to the stomach's total volume (inclusive of gastric secretions and saliva), provoking volume dependent rates of gastric emptying (14,53,69). Increases in intragastric pressure are sensed by stretch receptors in the gastric mucosa, as evidenced by a linear relationship in the amount of stomach distension and the number of impulses detected from afferent nerve fibers (61). Rehrer

et al. (69) demonstrated that a fast phase of gastric emptying can be maintained if more frequent boli (~146 ml every 20 minutes) are administered.

Relatively little fluid is absorbed in the stomach, meaning absorption processes are delayed until gastric emptying has occurred (79), wherein absorption occurs via the small intestine (70). The ensuing net fluid movement across intestinal walls into circulation passively occurs via aquaporins as a function of osmotic, hydrostatic, and filtration pressures (37). Reitemeier et al. (70), using isotopically labeled water, found that a mean time of 7 minutes was required for 67% of water to be absorbed from the small intestine into the arterial blood. Costill and Saltin (14) showed proportional increases in gastric emptying 15 minutes following consumption of single boli up to fluid  $\leq 600$  ml, with the maximal rate approximated at  $380 \text{ ml} \cdot 15 \text{ min}^{-1}$ .

Costill and Saltin (14) and Costill et al. (15) demonstrated that exercise intensity minimally alters gastric emptying until  $> 70\%$  of  $\text{VO}_2$  max, wherein gastric emptying is blunted and gastric residue is increased. However, Mitchell and Voss (53), examining approximate volumes of 800 ml, 1200 ml, and 1600 ml of fluid ingestion were able to demonstrate unaltered volume dependent gastric emptying during 2-hours of cycling at  $70\% \text{ VO}_2$  max. Fluids volumes delivered at 15-minute intervals, high ( $23 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{hr}^{-1}$ ), medium ( $17.1 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{hr}^{-1}$ ), and low ( $11.5 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{hr}^{-1}$  every 15 minutes) volumes were proportionally emptied.

### *Urinary Output and Fluid Balance*

Fluid intake volume may have proportional effects on expected urinary output and net fluid balance. High urinary output as a function of a large fluid intake volume (41,44) may manifest as a result of the fast first phase of gastric emptying (69).



Jones et al. (41), administering a large bolus of water equivalent to 100% of fluid loss by body weight, found that urinary output was higher than a trial administering an equivalent metered dose of water (12.5% of fluid loss by body weight every 30 minutes over 4 hours). The bolus trial produced a urinary output of 700 ml, while the metered dosing trial yielded 420 ml. Post-exercise net fluid balance was 20% greater in the metered trial when compared to the bolus trial. Kovacs et al. (44) examined a high rate of fluid consumption totaling 120% of fluid lost (60%, 40%, and 20% of body weight loss over 3 hours) and a low rate of fluid consumption (24% of body weight loss over 5 hours) finding that the high rate produced acute increases in urinary output and plasma volume.

Shirreffs et al. (84) found that a bout of cycling in the heat paired with immediate fluid intake of assorted volumes yielded proportional urine outputs during a 6-hour recovery period. Net fluid balance did not consistently improve with higher volumes of consumed fluid. A high fluid volume consumption (200% of fluid loss by body weight) provoked greater than 1,360 ml of urine output. Similarly, Savoie et al. (75) during 3 hours of water induced hyperhydration ( $30 \text{ ml}\cdot\text{kg FFM}^{-1}$ ) provoked a urine output of 1829 ml, where only 90 ml of fluid were retained. Taken together, these studies suggest that high volumes of exogenous fluid ingestion post-exercise could lead to bolus-induced diuresis and net negative fluid balance.

## **Fluid Composition**

### *Effects on Gastric Emptying*

Composition of ingested fluids has been shown to additionally alter the rate of gastric emptying, wherein plain water empties at faster rates than both energy dense

and hypertonic solutions (14,16,60,69,95). Costill and Saltin (14) showed that glucose concentrations above 139 mM yielded concurrent increases in gastric residue. Both Vist and Maughan (95) and Costill and Saltin (14) showed that osmolality can alter the rate of gastric emptying, where increases in osmolality above 200-300 mOsm can slow gastric emptying. However, carbohydrates appear to have a more robust impact than osmolality when these factors are taken together (95). Further, substrates with identical energy densities, regardless of solid or liquid composition, have been shown to empty into the small intestine at nearly identical rates (60).

#### *Rehydration Solutions and Rehydration Efficacy*

Inadequate fluid composition coupled with high fluid volume can increase urine production (75,83,84,86) and lower net fluid balance. The literature often pits differing compositions of fluid against one another following exercise (18,41,43,51,81,83-85). There are a host of drink compositions with differing contents of carbohydrate, sodium, and potassium (82). Fluid composition can differentially alter net fluid balance following a bout of exercise.

Shirreffs et al. (83) examined the post-exercise rehydration efficacy of 4 beverages delivered equivalent to 150% of fluid lost during exercise (carbohydrate-electrolyte, carbonated water/apple juice, mineral water, and plain water), finding that the carbohydrate-electrolyte solution, with its more favorable composition of sodium, moved individuals closer to their baseline net fluid balance. In a similar study conducted by Seery and Jakeman (81), milk was found to be a more effective rehydration solution than both a carbohydrate-electrolyte solution and plain water when administered at a volume of 150% of fluid lost following thermal exercise dehydration by 2%. Shirreffs et

al. (85) also evaluated the hydration efficacy of milk compared to a carbohydrate electrolyte-solution and plain water. However, in one trial, 20 mmol·L<sup>-1</sup> of sodium chloride was added to the milk. When evaluating cumulative urine outputs between the solutions, milk and the sodium chloride infused milk trials led to smaller outputs, moving individuals into net fluid balances similar to baseline.

In general, when sodium is added into post-exercise rehydration solutions it helps move individuals closer to their original net fluid balance (51,75,84) and can even hyperhydrate individuals (75). Both Merson et al. (51) and Shirreffs et al. (84) following exercise induced hypohydration found that sodium content was inversely proportional to urine output, meaning that the more sodium a solution had, the less the subjects urinated. During the higher sodium content trials individuals were in higher net fluid balance, indicating the importance of appropriate fluid composition for post-exercise ingestion.

#### *Carbohydrate-Electrolyte Solutions and Exercise*

Previous research examining fluid retention has elucidated that drinks other than water are more effective at recovering fluid (51,81,83-85). Similarly, sports drink solutions are often used during extended exercise in lieu of water alone in order to provide both fuel and fluid balance maintenance. The ACSM recommends delivering carbohydrate during extended exercise lasting longer than 60 minutes at rates of 30-60 g·h<sup>-1</sup> (91), so as to avoid sodium-glucose transporter saturation (27). Curvilinear carbohydrate dose responses have outlined diminishing performance returns at hourly ingestion rates that exceed approximately 80 g·h<sup>-1</sup> (89). Sports drink solutions contain a formulation of carbohydrates and electrolytes, where concentrations of 6-8% carbohydrate are

generally accepted to be a more optimal formulation for carbohydrate-electrolyte beverages (76). Ingesting carbohydrate beverages that use a combination of fuel sources, such as glucose and fructose have been shown to achieve peak carbohydrate oxidation rates during exercise (39). Ingesting carbohydrate beverages during exercise has been shown to maintain carbohydrate oxidation (10,12,15,49,53,62,78) and blood glucose levels during exercise (10,39), positively influencing performance (10,12,49,80).

Campbell et al. (10) examined the efficacy of different carbohydrate-supplement forms (sports drinks, sports beans, and gels) consumed during 80 minutes of cycling at 75%  $\text{VO}_2$  max on a post-ride 10 km cycling time-trial and found no difference in performance. Schweitzer et al. (80) administered equal total dosage volumes of a carbohydrate-electrolyte solution at differing intervals during 2 hours of cycling, providing a total of 75 g of carbohydrate in 4 doses $\cdot\text{h}^{-1}$  over 2 hours. Trials differed by providing fluids continuously, front-loaded (4 doses $\cdot\text{h}^{-1}$  for the first hour, placebo second hour), or end-loaded (placebo first hour, 4 doses $\cdot\text{h}^{-1}$  for the second hour). Despite differences in the timing of the carbohydrate-electrolyte solution, total work output did not differ during a 15-minute cycling time trial. McConnell et al. (49) found similar results when altering carbohydrate ingestion interval. In general, it appears that as long as carbohydrates are ingested, positive downstream performance influences occur.

## **Renal and Hormonal Response to Exogenous Fluid and Exercise**

### *Glomerular Filtration*

The volume and solutes of liquids crossing the intestinal walls into circulation manipulate the glomerular hydrostatic pressure at the renal level, dictating how much fluid and solute may cross the filtration membrane (4). Renal blood flow and GFR at rest

have been determined to be approximately  $1200 \text{ ml}\cdot\text{min}^{-1}$  and  $120 \text{ ml}\cdot\text{min}^{-1}$  (26,68), respectively. GFR incrementally decreases as a function of increasing exercise intensity (26). Radigan and Robinson (68) found that walking at 3.0 mph at a 5% grade in the heat ( $50^{\circ}\text{C}$ ) decreased GFR to approximately  $80 \text{ ml}\cdot\text{min}^{-1}$ . Freund et al. (26) also demonstrated a GFR decrease to less than  $80 \text{ ml}\cdot\text{min}^{-1}$  when subjects cycled at 80% of  $\text{VO}_2$  max for 20 minutes. Distribution of cardiac output during exercise mirrors GFR perturbations. At rest, approximately 20% of cardiac output is distributed to the kidneys, while heavy exercise warrants blood flow redistribution to working tissue, allowing for approximately 3% of cardiac output to be distributed to the kidneys, despite cardiac output increases from  $5 \text{ L}\cdot\text{min}^{-1}$  to greater than  $20 \text{ L}\cdot\text{min}^{-1}$  (5).

GFR can be measured from renal clearance of exogenous substances that are filterable at the glomerulus, unaltered (synthesized, destroyed, reabsorbed, or excreted) by the tubules, and possess inert physiological effects, one such substance is inulin (88). GFR can also be measured from the clearance of creatinine or estimated from serum creatinine or cystatin C using standardized equations (45,66). The Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation has been proposed as a more accurate measure of eGFR when compared to the previously accepted Modification of Diet in Renal Disease (MDRD) equation (45). The CKD-EPI equation's accuracy is especially prevalent at higher GFRs, indicating a low bias range at  $> 60 \text{ ml}\cdot\text{min}^{-1}$ . However, Poortmans et al. (66) examined the accuracy of the CKD-EPI equation following 30 minutes of treadmill exercise at 80%  $\text{VO}_2$  max finding that the CKD-EPI equation underestimated GFR when compared to clearance creatinine using timed urine collections. Underestimation of GFR using the CKD-EPI equation persisted

into the recovery period. Freund et al. (26) indicated that exercise at > 80% of  $\text{VO}_2$  max yields a  $\text{GFR} < 80 \text{ ml}\cdot\text{min}^{-1}$ , putting the  $\text{GFR}$  resulting from the exercise intensity outlined in the Poortmans et al. (66) study within the high bias range of the CKD-EPI equation determined by Levey et al. (45).

### *Hormone Response*

Hormones acting at the renal level, angiotensin II, aldosterone, and vasopressin influence fluid retention, while atrial natriuretic peptide influences fluid excretion (4). Initial hydration status alters baseline plasma hormonal concentration (8), where graded levels of hypohydration warrant higher concentrations of fluid retaining hormones (26,55). Hormonal release also responds to changes in blood volume (84), blood pressure (24,33,34,94), and plasma osmolality (13,55). Plasma renin activity is positively correlated with increases in plasma angiotensin II, wherein angiotensin II synthesis is associated with aldosterone secretion (3,90). Angiotensin II is a rapid and potent vasoconstrictor (3,21) and aldosterone works at the renal level to retain sodium (3,13,67). Angiotensin II and aldosterone work concurrently to raise systemic blood pressure and blood volume. Vasopressin promotes water reabsorption into the blood from renal collecting ducts (58), allowing for antidiuresis. Atrial natriuretic peptide increases sodium excretion (67), promoting diuresis. Angiotensin II, aldosterone, and vasopressin are additionally influenced by exercise intensity (13,26,55,90), exercise duration (13,64), fluid volume (84), and fluid composition (8,90).

Staessen et al. (90) demonstrated during a graded cycling protocol (30%, 60%, and 100% of  $\text{VO}_2$  max) that angiotensin II increased in an intensity dependent manner and in proportion to plasma renin activity during light and moderate intensities. Freund

et al. (26) administered varying intensities of 20-minute cycle ergometry (25%, 40%, 60%, and 80% of  $\text{VO}_2$  max), determining that plasma renin activity, aldosterone, and vasopressin were also intensity dependent. Similarly, Montain et al. (55) determined that 20 minutes of treadmill exercise at 65% of  $\text{VO}_2$  max elicited higher levels of aldosterone and vasopressin when compared to 25% and 45% of  $\text{VO}_2$  max. 60% of  $\text{VO}_2$  max has been listed as a threshold at which fluid retaining hormones significantly increase (26).

Convertino et al. (13) administered 8 days of prolonged cycle ergometer training for 2 h-day<sup>-1</sup> at 65% of  $\text{VO}_2$  max, producing nine-fold increases in plasma renin activity and vasopressin during exercise when compared to baseline. Comparably, 90 minutes of cycling at 60%-70% of  $\text{VO}_2$  max promoted progressive increases in plasma renin activity, aldosterone, and vasopressin (64). Progressive increases in plasma hormonal concentration has also been observed over 3 hours of low to moderate intensity cycling (70 watts) (8). The threshold for increases in plasma renin activity and aldosterone during prolonged exercise occurs at approximately 30 minutes, while vasopressin significantly increases after approximately 60 minutes (64).

Shirreffs et al. (84), examining exogenous fluid volume and composition administration, found that blood volume increased as function of the fluid volume consumed following exercise. Low fluid volume trials, with subsequently decreased blood volume, displayed elevated plasma levels of vasopressin (2 hours post-exercise), angiotensin II (2 and 6 hours post-exercise), and aldosterone (2 and 6 hours post-exercise) when compared to high fluid volume trials. Concurrent rehydration during exercise can attenuate hormonal response (8,90). Staessen et al. (90) demonstrated that higher sodium intake during an incremental cycling protocol to 60%  $\text{VO}_2$  max

decreased angiotensin II's stimulatory effect on aldosterone, where urinary sodium excretion was inversely related to plasma aldosterone concentration. Brandenberger et al. (8), hydrating subjects with a glucose-electrolyte drink or water during a 3-hour cycling protocol, displayed suppressed plasma renin activity, aldosterone, and vasopressin plasma concentration levels both during exercise and rest.

Increases in plasma osmolality are sequentially coupled with increased vasopressin (55,92,93). Montain et al. (55) showed across a variety of treadmill exercise intensities (25%, 45%, and 65% of  $\text{VO}_2$  max) and hypohydration levels (0%, 3%, and 5% of body weight) that increases in vasopressin were closely related with rises in plasma osmolality, while Thompson et al. (92,93) quantifies a threshold of approximately 285 mOsm as the point in which vasopressin begins to rise.

## **Hydration Strategies**

### *During Exercise*

Fluid intake recommendations function on the premise that fluids are available, accessible, and actively ingested by individuals. *Ad libitum* intake strategies are not always effective in the maintenance of body weight over extended exercise bouts and can lead to decreases in body weight upwards of 4% (22,23,35). When *ad libitum* fluid consumption was compared to programmed fluid consumption during a treadmill half marathon, individuals consumed 3.5 times more fluid with a programmed amount, maintaining body weight within 1% of pre-exercise weight (23). Greenleaf and Sargent II (35) showed that heat and hypohydration had larger influences on *ad libitum* fluid consumption than exercise, due differences in thirst sensitivity when compared to heat and hypohydration alone. Exercise related decreases in thirst sensitivity may contribute



to lesser amounts of fluids consumed *ad libitum*, despite adequate availability and accessibility.

The multifactorial nature of sweat loss is reflected in the spectrum of fluid volume intake stances. Exogenous fluid intake recommendations vary as a function of heat exchange factors in reference to the environment (humidity, temperature, wind), work rate, and exposed surface area (1,47,50,76). Recommendations vary further based upon sport, occupation, and individual sweat rates (50,76). Governing bodies seem to stray away from blanket fluid intake recommendations, recognizing that individuals competing within the same event or sport can have variable sweat rates (6). The ACSM, NATA, and the United States Army TRADOC regulation 350-29 collectively recommend consuming exogenous fluids to prevent dehydration  $\geq 2\%$  of body weight (1,50,76).

The ACSM's suggested strategy advises consuming fluids *ad libitum*, amounting to 400 to 800 ml·h<sup>-1</sup>, which is dependent upon work rate, environment, clothing, and body weight (76). The NATA suggests that athletes should be aware of their individual sweat rates as a function of their sport, suggesting 100% replacement of estimated fluid loss for < 2% body weight decrease with the stipulation as not increase in body weight (50). Military fluid intake guidelines are based upon a matrix of 15 conditions in reference to 5 colored flag temperatures (white, green, yellow, red, and black) and 3 work-rate categories (easy, moderate, and hard), with fluid consumption assigned in reference to the corresponding environment, work task, and equipment worn (1,47). 15 conditions yield a wide range of acceptable fluid intake volumes, ranging from 470 to 940 ml·h<sup>-1</sup> (0.5 to 1 quart·h<sup>-1</sup>) with the stipulation as not to exceed either 1400 ml·h<sup>-1</sup> (1.5

quarts·h<sup>-1</sup>) or a total of 11,300 ml·day<sup>-1</sup> (12 quarts·day<sup>-1</sup>). The strategy outlined in the TRADOC 350-29 is the only strategy to take into account work:rest ratios.

Collectively, fluid intake recommendations from these agencies attempt to limit dehydration to no more than 2% of body weight, precipitating a wide range of acceptable fluid intake volumes (1,50,76). The ACSM and NATA underscore the importance of the individual's role in assessing their own sweat rates so that they may accommodate fluids appropriately tailored for their specific event and individual response (50,76). These recommendations outline total hourly rates, but do not specify dose intervals that accommodate the total hourly intake, illustrating subtle ambiguities associated with fluid intake recommendations.

### *Post Exercise*

Composition and volume have been identified as key post exercise rehydration components (41,50,51,76,81,83-85). The ACSM recommends consuming ~1.5 L of fluid for every kilogram lost during exercise (76), while NATA advises consumption of 150% of fluid lost by body weight (50). Generally, both the ACSM and NATA advise including some facet of sodium to aid in fluid retention. Both recommendations require diligent individual assessment of weight lost during exercise and awareness of sweat loss constituents. Post exercise fluid replacement research studies have examined the relationship between fluid volume and composition finding that inappropriate volume or composition results in unfavorable net fluid balance (81,83-85). High fluid volumes (>100% of fluid lost by body weight) can acutely provoke positive net fluid balance, however, inadequate composition of ingested fluids only increases urine output during extended recovery periods (51,83,84). Merson et al. (51), following post exercise thermal

dehydration by 2% of body weight consumed 150% of fluids lost and found that urine output incrementally decreased with higher sodium concentrations (1, 31, 40, and 50 mmol·L<sup>-1</sup>) in each fluid. More favorable net fluid balance was achieved when subjects ingested fluids with 40 or 50 mmol·L<sup>-1</sup>. Even fluids administered with low sodium and a volume of 200% fluids lost during exercise leads to unfavorable net fluid balances as a result of greater urinary outputs (84). Plain water, with an unfavorable composition, has been shown to provoke high urine outputs when used as a post exercise rehydration solution (81,83,85). Broadly, post exercise rehydration strategies should include a volume slightly above what was lost during the preceding bout of exercise with the addition of sodium to aid in fluid retention.

#### *Fluid Administration Frequency*

Several studies have taken the evidence-based influence that fluid composition and fluid volume have on post exercise rehydration by taking them a step further and altering administration strategy (41,44,63,81). Pérez-Idárraga and Aragón-Vargas (63) examined 3 different administration styles of a carbohydrate-electrolyte solution (40% of fluids lost every 30 minutes, ascending 25%, 40%, 55% once every 30 minutes, or descending 55%, 40%, or 25% every 30 minutes for 90 minutes) following thermal exercise dehydration by 2% of body weight, finding that there was no difference in urine volume or fluid retention between the trials. However, with a similar dosing style, Kovacs et al. (44) examined a high (60%, 40%, 20% of body weight loss over 3 hours) and low rate (24% of body weight loss over 5 hours) of a carbohydrate-electrolyte solution totaling 120% of fluid lost during exercise. High rates of fluid consumption produced acute increases in urinary output, plasma volume, and net fluid balance.

Jones et al. (41) evaluated post-exercise rehydration efficacy of metered (2 doses·h<sup>-1</sup> for 4 hours) and bolus dosing (1 dose at the beginning of the 4 hour recovery) of plain water equal to 100% of fluid lost during exercise in the heat, finding a 20% increase in net fluid balance in the metered dosing over an 8-hour recovery period. Similarly, Seery and Jakeman (81) evaluated a metered dosing (1000 ml and then 500 ml every 30 minutes until individualized replenishment) of milk, carbohydrate-electrolyte, and plain water wherein superimposition of a metered dosing style with a higher energy density composition improved post exercise net fluid balance. Taken together, these studies suggest that dosage volume and fluid composition interact to improve post-exercise rehydration status.

## **Conclusion**

Fluid intake recommendations for preserving euhydration status during exercise are relatively ambiguous, leaving administration style open to end user interpretation. Governing bodies outline the importance of individual assessment for adequate exogenous fluid provision. Research has suggested that fluid volume, composition, and administration style may interact to alter gastric emptying rates and hormonal balance that in turn influence fluid retention. However, implementing evidence based post-exercise rehydration strategies during about of extended exercise in the heat to maintain euhydration is unclear.

## **CHAPTER THREE: METHODOLOGY**

### **Subjects**

12 healthy males (age  $25 \pm 4$  years, mass  $78 \pm 12$  kg,  $14.6 \pm 4.4$  % body fat,  $\text{VO}_2$  peak  $55.4 \pm 6.6$  ml·kg<sup>-1</sup>·min<sup>-1</sup>,  $n = 12$ ) completed the study. Subjects were deemed

fit for activity and injury free via the 2019 Physical Activity Readiness Questionnaire (PAR-Q). The University's institutional review board approved all methodologies.

Subjects were informed of the procedures and risks associated with participating in the study and provided both informed and written consent.

## **Preliminary Testing**

### *Body Composition and VO<sub>2</sub> Peak Testing*

Each subject refrained from alcohol consumption and exercise during the 24 hours leading up to the initial visit and came in overnight fasted. After recording mass (CW-11, Ohaus, Pine Brook, NJ) and measuring height, body composition was obtained through hydrodensitometry using residual volume estimates (28). Underwater mass was collected using an electronic scale (Exertech, Dresbach, MN) and used to calculate body density. The Siri equation was used to convert body density to percent body fat (87).

A motorized treadmill (4Front, Woodway USA Inc., Waukesha, WI) was used to determine peak oxygen uptake (VO<sub>2</sub> Peak) via a graded exercise test until volitional exhaustion and the achievement of at least two of the following criteria: respiratory exchange ratio (RER) >1.10, plateau in VO<sub>2</sub>, heart rate (HR) within 10 beats of age predicted max, or a rating of perceived exertion (RPE) >17. The Bruce graded exercise test began with a 10-minute warm up at 0.76 m·s<sup>-1</sup> (1.7 mph) at a 0% grade, remaining test stages were 3 minutes in length (9). The stages are outlined in table 1. During the test, expired gas samples were collected using a calibrated metabolic cart (ParvoMedics Inc., Salt Lake City, Utah). During the final 30-seconds of each stage, HR from a watch and chest strap (Polar Electro, Kempele, FL) and RPE using the Borg

Scale (7) were recorded.  $\text{VO}_2$  peak was calculated as the highest 15-second average oxygen uptake achieved before volitional exhaustion.

**Table 1:**  $\text{VO}_2$  peak test: Bruce protocol stages. Stage 1 is a 10-minute warm up, while each subsequent stage is 3 minutes long.

STAGE	Speed ( $\text{m}\cdot\text{s}^{-1}$ )	% Grade
1	0.76 (1.7 mph)	10
2	1.1 (2.5 mph)	12
3	1.5 (3.4 mph)	14
4	1.9 (4.2 mph)	16
5	2.2 (5.0 mph)	18
6	2.5 (5.5 mph)	20
7	2.7 (6.0 mph)	22
8	2.9 (6.5 mph)	24
9	3.1 (7.0 mph)	26

#### *Metabolic Rate Prediction*

An equation outlined by Pimental and Pandolf (65) was used to approximate the appropriate speed, grade, and load combination to elicit a  $\text{VO}_2$  approximating  $22.2 \text{ ml}\cdot\text{kg}\cdot\text{min}^{-1}$  during the experimental trials. The equation is outlined below, where  $W$  is the subject mass in kg,  $L$  is the external load in kg,  $\eta$  is the terrain factor,  $V$  is the velocity in  $\text{m}\cdot\text{s}^{-1}$ , and  $G$  is the grade in percent. A terrain factor of 1 was used to represent the surface of the treadmill.

$$\text{VO}_2(\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}) = 1.5W + 2.0(W + L)(L/W)^2 + \eta(W + L)(1.5V^2 + 0.35VG)$$

#### *Familiarization Trial*

All subjects were required to complete a 60-minute familiarization trial at least 24 hours post  $\text{VO}_2$  peak test and 7 days before beginning the experimental protocol. Each subject refrained from the consumption of alcohol and caffeine, while at the same time abstaining from exercise for the 24 hours leading up to all trials. Subjects were

additionally instructed to consume 500 ml of water 60 minutes before bed the night before, fast for the 12 hours prior, and refrain from consuming any fluids the morning of the familiarization trial. For accountability and standardization across each trial, each subject kept a food and fluid consumption log for the 24 hours prior.

Upon entering the lab, subjects voided their bladder, drank 500 ml of water, and sat for 30 minutes in a temperate environment before providing a baseline void and nude body weight. The familiarization trial mirrored the trial protocol. Subjects walked for 60 minutes on a motorized treadmill with a 15 kg pack (Fireline Pack NFES# 0674, FedMall, Fort Belvoir, VA) at  $1.4 \text{ m}\cdot\text{s}^{-1}$  with a 5% grade in an environmental chamber (Tesco Inc., Warminster, PA) set at  $33^{\circ}\text{C}$  with a relative humidity of 30%. Each subject wore a standard WLFF Nomex shirt (Wildland Firefighter's Shirt, FedMall, Fort Belvoir, VA), flame resistant pants (Flame Resistant Pants, FedMall, Fort Belvoir, VA), a cotton base layer t-shirt (Hanes, Winston-Salem, NC), and their own choice of footwear. No fluid was administered to subjects during this trial. Following the hour, subjects immediately voided their bladder, towed off, and had nude body weight recorded.

The purpose of the 60-minute familiarization trial was to predict total water loss during the 120-minute experimental protocols and to minimize changes in body weight. Post-exercise nude body weight was subtracted from pre-exercise nude body weight to calculate the rate of fluid loss ( $\text{ml}\cdot\text{h}^{-1}$ ) in order to accommodate 100% of predicted fluid loss during the experimental trials.

## **Experimental Trial**

### *Overview*

Each subject completed 4 trials in a repeated-measures cross over design. Subjects were instructed to abide by the same parameters and restrictions as outlined in the familiarization trial, while adhering to their food and fluid consumption logs. There were 7 days between the first 2 trials, a 2-week washout period, and 2 more experimental trials separated by 7 more days. Trials were separated in this manner to control for potential heat acclimation.

Subjects were assigned to a composition of plain water or carbohydrate-electrolyte solution before being further assigned into completing either a micro-dosing or bolus-dosing trial first. Following the 2-week washout period, subjects switched solutions and administration strategy order, making 4 groups of 4 intervention orders (Table 2). The fluid interventions included bolus-dosed carbohydrate-electrolyte (BCE), bolus-dosed water (BW), micro-dosed carbohydrate-electrolyte (MCE), and micro-dosed water (MW).

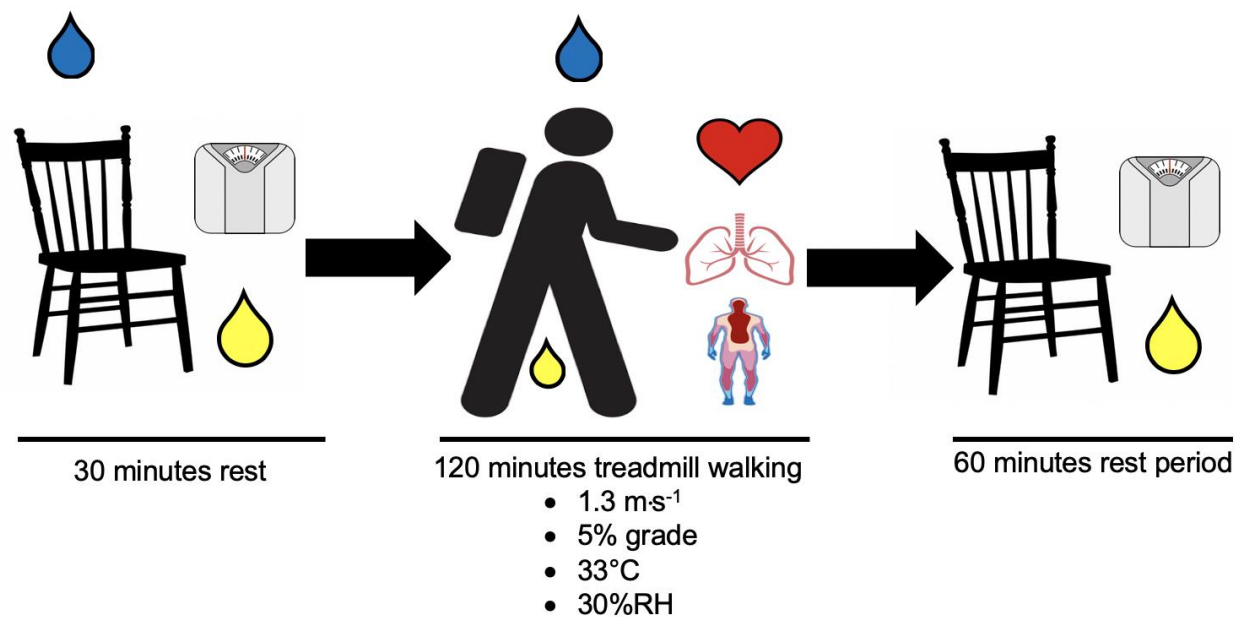
**Table 2:** Subject assignments by group ( $n=12$ ): micro-dosed water (MW), bolus-dosed water (BW), micro-dosed carbohydrate-electrolyte (MCE), or bolus-dosed carbohydrate-electrolyte (BCE).

<b>GROUP (<math>n=12</math>)</b>	<b>WEEK 1</b>	<b>WEEK 2</b>	<b>WEEK 3</b>	<b>WEEK 4</b>
A ( $n=3$ )	MCE	BCE	BW	MW
B ( $n=3$ )	BCE	MCE	MW	BW
C ( $n=3$ )	MW	BW	BCE	MCE
D ( $n=3$ )	BW	MW	MCE	BCE

Identical to the familiarization trial, subjects voided their bladder, drank 500 ml of water, and sat for 30 minutes in a temperate environment before providing a baseline void and nude body weight. Following baseline nude body weight, subjects dressed in the above-mentioned standardized WLFF uniform and pack. The second voided urine was measured for volume and urine specific gravity (USG) (URC-NE, Atago, Cohasset, MA). Subjects then completed a 120-minute treadmill protocol in the above-mentioned



conditions while consuming either BCE, BW, MCE, or MW. Heart rate (HR), rectal temperature ( $T_c$ ) (RET-1, PhysiTemp, Clifton, NJ), and skin temperature ( $T_s$ , sensor placed 5 cm above the left nipple on pectoralis major muscle) (SST-1, PhysiTemp, Clifton, NJ) were continuously measured during the treadmill protocol and recorded via a portable data logger (SDL200, Extech Instruments, Nashua, NH). Total urine output was recorded twice during the 120-minute intervention (60 and 120 minutes) and 60 minutes following exercise. Each void was measured for USG and volume. Steady state expired gas was collected near the end of each hour during exercise. Nude body weight was measured pre, post, and 60 minutes post exercise. The overview is outlined below in figure 1.



**Figure 1:** Overview of the experimental protocol.

### *Treadmill Protocol*

Each subject completed an intermittent treadmill walking protocol, 2 x 55-minutes with 5-minutes rest to complete the hour and accommodate urine collection. The

treadmill and environmental chamber were set at a speed, grade, temperature, and relative humidity as mentioned above.

### *Fluid Administration*

All fluids were kept in the heat chamber for 12 hours prior to each trial and served at the temperature of the chamber (33°C) to ascertain fluid delivery temperature across trials. Fluid composition consisted of either water or a carbohydrate-electrolyte solution (Gatorade Endurance Thirst Quencher, PepsiCo, Chicago, IL) (Table 3).

**Table 3:** Carbohydrate-Electrolyte Solution Composition.

<b>Sodium (mmol·L<sup>-1</sup>)</b>	<b>Potassium (mmol·L<sup>-1</sup>)</b>	<b>Carbohydrates (g·L<sup>-1</sup>)</b>	<b>Calories (Kcal·L<sup>-1</sup>)</b>
37.4	9.9	61.1	388.8

Micro-dosed intake was administered for 120 minutes every 2 minutes from 5-25 minutes, 28-50 minutes, and a final time during the 53rd minute in equal increments (22 doses·h<sup>-1</sup>, 46 ± 11 ml·dose<sup>-1</sup>). Micro-dosed administration was done using 60 ml syringes (BD, Franklin Lakes, NJ). Bolus-dosed intake was administered near the top of each hour (minute 5 and 65) in 2 equal amounts (1005 ± 245 ml·h<sup>-1</sup>). Subjects were given 5 minutes to consume the bolus volume. The total fluid volume administered in each trial was identical within subjects, matching the fluid loss predicted from the pre-experiment familiarization trial.

**Table 4:** Fluid administration technique and frequency.

<b>Water</b>		<b>Carbohydrate-Electrolyte</b>	
Micro-Dosing	Bolus-Dosing	Micro-Dosing	Bolus-Dosing
22 doses·h <sup>-1</sup>	1 dose·h <sup>-1</sup>	22 doses·h <sup>-1</sup>	1 dose·h <sup>-1</sup>

### *Heart Rate, Core Temperature, and Skin Temperature*

HR,  $T_c$ , and  $T_s$  were measured throughout the 120-minute duration of exercise and recorded within the last 30-seconds of minutes 5 (first hour), 53, and 113, totaling 3 recordings over 120 minutes. HR,  $T_c$ , and  $T_s$  were measured using the above-mentioned equipment. PSI was calculated with an equation developed by Moran et al. (56), where  $T_{ct}$  and  $HR_t$  are simultaneous measurements at the time point of interest and  $T_{c0}$  and  $HR_0$  are initial measurements from the beginning of each trial:

$$PSI = 5(T_{ct} - T_{c0}) \cdot (39.5 - T_{c0})^{-1} + 5(HR_t - HR_0) \cdot (180 - HR_0)^{-1}$$

#### *Body Weight, Urine Output, and Sweat Rate*

Nude body weight was measured pre, immediately post, and 60 minutes post exercise. Urine was additionally collected at the end of each hour during exercise. Urine void volume was recorded and measured for USG. Sweat rate was calculated using the change in pre- and post-exercise nude body weight in kg (BW), corrected for urine output in kg (U), fluid consumption in kg (FC), and respiratory water loss in kg (RWL). RWL was calculated per Mitchell et al. (52) and water vapor pressure as determined by Fox et al. (25).

$$SR (L \cdot h^{-1}) = \frac{[(BW_{PRE} - BW_{POST} + FC) - (U + RWL)]}{h}$$

$$RWL (g \cdot min^{-1}) = (0.0019)(VO_2 L \cdot min^{-1})(44 - \text{Water Vapor Pressure})$$

$$\text{Water Vapor Pressure} = 13.955 - .6584 (\text{Temperature}) + 0.0419 (\text{Temperature})^2$$

#### *Substrate Utilization*

Gas exchange was measured using a calibrated metabolic cart via indirect calorimetry as indicated above. Steady state expired gas samples were collected at 0-5 minutes during the first hour to act as baseline. Subsequent expired gas samples were recorded during minutes 50-53 and 110-113, totaling 3 recordings over 120 minutes.

Steady state values of  $VO_2$  ( $L \cdot \text{min}^{-1}$ ) and  $VCO_2$  ( $L \cdot \text{min}^{-1}$ ) were used to calculate total carbohydrate oxidation as demonstrated by Jeukendrup and Wallis (40), with the assumption that protein oxidation was negligible:

$$\text{Carbohydrate Oxidation (g} \cdot \text{min}^{-1}\text{)} = (4.344 \cdot VCO_2) - (3.061 \cdot VO_2)$$

#### *Blood Sampling, Preservation, and Analysis*

Venipuncture technique was used to collect 5 ml of blood from each subject pre, post, and 60 minutes post exercise from an antecubital vein into a serum separator tube (BD, Franklin Lakes, NJ). Samples were inverted 5 times and allowed to sit for 20 minutes before being spun in a benchtop centrifuge (MR22i, Jouan Inc., Waltham, MA) 15 minutes. Following centrifugation, aliquots of serum plasma were placed into 2 secondary tubes and stored at  $-30^\circ\text{C}$  for less than 14 days before analysis. The blood metrics measured were glucose, sodium, potassium, chloride, and estimated glomerular filtration rate (eGFR). All assays were completed by LabCorp in Missoula, MT (Laboratory Corporation of America, Burlington, NC).

#### *Statistical Analysis*

A two-way ANOVA (time x trial) with repeated measures was used to evaluate body weight (pre-, post-, and 60 minutes post exercise), HR (5, 60, and 120 minutes during exercise),  $T_c$  (5, 60, and 120 minutes during exercise),  $T_s$  (5, 60, and 120 minutes during exercise), PSI (5, 60, and 120 minutes during exercise), urine by void (60 and 120 minutes during exercise and 60 minutes post exercise), and substrate utilization (5, 60, and 120 minutes during exercise). A one-way ANOVA (trial) with repeated measures was used for RWL during exercise, relative exercise intensity, exercise sweat rate, post-exercise sweat rate, cumulative urine output, exercise urine output, and post

exercise urine output. A 2-tailed paired t-test was used to evaluate oxygen consumption ( $\text{VO}_2$ ) measured during exercise and estimated from the Pimental and Pandolf equation (65). Statistical significance was set at a type I probability error of less than 5% ( $p < 0.05$ ), with data expressed as *mean  $\pm$  SD*. Statistical analysis was done using SPSS (IBM, Chicago, IL) and Excel (Microsoft Corporation, Redmond, WA).

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**Exogenous fluid delivery schedule and composition on fluid balance, physiologic strain, and substrate use in the heat**

Short running head: Fluid delivery schedule and composition

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**Abstract**

**Introduction:** Wildfire suppression is characterized by high total energy expenditures and water turnover rates. Hydration position stands outline hourly fluid intake rates. However, dose interval remains ambiguous. We aimed to determine the effects of micro- and bolus-dosing water (MW, BW) or carbohydrate-electrolyte (MCE, BCE) solutions on fluid balance, heat stress, and carbohydrate oxidation during extended thermal exercise. **Methods:** Males ( $n=12$ ), in a repeated measures cross-over design, completed 4, 120-minute treadmill trials ( $1.3 \text{ m}\cdot\text{s}^{-1}$ , 5% grade,  $33^\circ\text{C}$ , 30%RH) wearing a USFS uniform and 15kg pack. Fluid delivery approximated losses calculated from a pre-experiment familiarization trial, providing 22 doses $\cdot\text{h}^{-1}$  or 1 dose $\cdot\text{h}^{-1}$  ( $46\pm 11, 1005\pm 245 \text{ ml}\cdot\text{dose}^{-1}$ ). Body weight (pre-, post-exercise) and urine volume (pre-, during, post-exercise) were recorded. Heart rate, rectal temperature, skin temperature, and steady state expired air samples were recorded throughout exercise. Differences were determined via repeated measures ANOVA's with statistical significance set at  $p<0.05$ . **Results:** Total body weight loss ( $n=11$ ,  $-0.6\pm 0.3 \text{ kg}$ ,  $p>0.05$ ) and cumulative urine output ( $n=11$ ,  $677\pm 440 \text{ ml}$ ,  $p>0.05$ ) were not different across trials. Exercise sweat rate was lower in the MCE trial than the BCE, BW, and MW trials ( $n=11$ ,  $0.86\pm 0.22$ ,  $0.93\pm 0.21$ ,  $0.91\pm 0.23$ ,  $0.89\pm 0.22 \text{ ml}$ , respectively,  $p<0.05$ ). PSI was lower at minute 60 than 120 ( $3.6\pm 0.7$ ,  $4.5\pm 0.9$ ,  $p<0.05$ ), with no differences across trials. Carbohydrate oxidation was higher in the CE trials than the W trials ( $1.5\pm 0.3$ ,  $0.8\pm 0.2$ ,  $\text{g}\cdot\text{min}^{-1}$ ,  $p<0.05$ ), with no differences between identical composition dosing styles. **Conclusions:** Varied fluid delivery schedules of equal volume did not affect fluid balance, PSI, and carbohydrate oxidation during extended thermal work.

**Key Words:** hydration, thermal stress, carbohydrate oxidation

## Introduction

Wildland firefighters (WLFF) commonly complete 14-day assignments with 12-18 hour workshifts under arduous conditions <sup>1</sup>. Total energy expenditure ranges between 12 and 26 MJ·day<sup>-1</sup> (2868 to 6214 kcal·day<sup>-1</sup>) <sup>1-3</sup>. The mean metabolic cost of load carriage hiking in WLFF (via the Pimental and Pandolf equation <sup>4</sup>) is  $22 \pm 12$  ml·kg<sup>-1</sup>·min<sup>-1</sup>, and varies between 19 and 34 ml·kg<sup>-1</sup>·min<sup>-1</sup> depending on hike classification (ingress, shift, egress, or more intense training hikes) <sup>5</sup>. Measurements of water turnover from multiple investigations demonstrate aggressive hydration demands ( $6.7 \pm 1.4$  L·d<sup>-1</sup> <sup>6</sup>,  $7.8 \pm 1.6$  L·d<sup>-1</sup> <sup>3</sup>, and  $9.5 \pm 1.7$  L·d<sup>-1</sup> <sup>2</sup>). Digital flowmeter drinking technology has revealed varied hourly volume and intake intervals that are proportionate to changes in environmental stress during a workshift <sup>7</sup>.

Inadequate exogenous fluid ingestion during extended exercise at a constant work rate can lead to reductions in plasma volume and thermoregulatory strain from increases in heart rate (HR) and core temperature <sup>8,9</sup>. Appropriately administered fluid volume has been shown to alleviate HR and core temperature increases otherwise resulting from hypohydration <sup>10-13</sup>. However, WLFF appear to modify their behavior in response environmental stimuli by self-selecting sustainable work rates and avoiding unnecessary heat to attenuate thermal strain <sup>2,14</sup>.

The American College of Sports Medicine (ACSM) <sup>15</sup>, the National Athletic Trainers' Association (NATA) <sup>16</sup>, and U.S Army training doctrine regulation 350-29 <sup>17</sup> have outlined total hourly exogenous fluid intake recommendations designed to limit dehydration to < 2% loss of initial body weight. These recommendations propose that minimizing dehydration ensures that thermoregulatory strain can be attenuated. However, recommendations remain ambiguous for the fluid ingestion interval between doses to comply with suggested hourly intake needs.

Research employing different fluid intake methodologies following exercise<sup>18-22</sup> have elucidated effective fluid recovery strategies that appear to manipulate gastric emptying. These rehydration strategies have demonstrated that volume<sup>20</sup>, composition<sup>20-22</sup>, and administration interval<sup>18,22</sup> collectively influence rates of rehydration and fluid retention by reducing urinary fluid loss. Prior research has demonstrated that intermittent fluid administration was 20% more effective than a bolus dose at recovering net fluid balance post-exercise<sup>18</sup>.

Individual bolus doses  $\leq 600$  ml empty more rapidly from the stomach, while gastric emptying appears minimally impaired until  $\geq 70\%$  of  $\text{VO}_2$  peak<sup>23</sup>. The majority of WLFF tasks occur at intensities  $\leq 50\%$  of  $\text{VO}_2$  max<sup>5</sup>, therefore minimally altering the volume dependent gastric emptying relationship. However, changing the administration interval of different fluid volumes and/or fluid compositions could optimize the movement of fluid out of the stomach, contributing to an enhanced ability to maintain euhydration during work, while improving post-work fluid retention.

Sports drink solutions are often formulated with a combination of carbohydrate (CHO) and electrolytes for use during extended exercise in lieu of water alone to accommodate exogenous CHO oxidation and improve fluid balance. The contribution of exogenous CHO oxidation to total CHO oxidation during exercise is improved when CHO sources are delivered at 10-15-minute intervals<sup>24-26</sup>. Large amounts of ingested exogenous CHO ( $70\text{-}100\text{ g}\cdot\text{h}^{-1}$ ) have been shown to shift the proportion of exogenous fuel use rates towards peak values<sup>26,27</sup>. However, it is unclear if the provision of frequent small oral doses will alter total CHO oxidation.

The primary purpose of this study was to determine the influence of altered fluid delivery schedules, micro-dosing versus bolus-dosing, of identical hourly volumes on fluid

retention and physiologic strain index (PSI) while exercising in the heat. A secondary purpose was to examine the influence of superimposing a carbohydrate-electrolyte (CE) solution with an altered fluid delivery schedule on fluid retention and substrate use during an identical exercise environment. We hypothesized that 1) micro-dosing fluids would improve overall fluid retention regardless of composition, while 2) inclusion of a CE solution would further improve fluid retention. We additionally hypothesized 3) that micro-dosing a CE solution would maintain a higher CHO oxidation rate during exercise.

## **Methods**

### **SUBJECTS**

12 healthy males (age  $25 \pm 4$  years, mass  $78 \pm 12$  kg,  $14.6 \pm 4.4$  % body fat,  $\text{VO}_2$  peak  $55.4 \pm 6.6$  ml·kg<sup>-1</sup>·min<sup>-1</sup>,  $n = 12$ ) completed the study. Subjects were deemed fit for activity and injury free via the 2019 Physical Activity Readiness Questionnaire (PAR-Q). The University's institutional review board approved all methodologies. Subjects were informed of the procedures and risks associated with participating in the study and provided both informed and written consent.

### **PRELIMINARY TESTING**

#### *Body composition and $\text{Vo}_2$ peak testing*

Each subject came in overnight fasted and refrained from alcohol consumption and exercise during the 24 hours leading up to the first visit. After recording mass (CW-11, Ohaus, Pine Brook, NJ) and measuring height, body composition was obtained via hydrodensitometry (Exertech, Dresbach, MN) using residual volume estimates<sup>28</sup>. The Siri equation was used to convert body density to percent body fat<sup>29</sup>.

Peak oxygen uptake ( $\text{VO}_2$  peak) was determined via a Bruce graded exercise test<sup>30</sup> on a motorized treadmill (4Front, Woodway USA Inc., Waukesha, WI) until volitional exhaustion

and the achievement of at least two of the following criteria: respiratory exchange ratio (RER) > 1.10, plateau in  $\text{VO}_2$ , heart rate (HR) within 10 beats of age predicted max, or a rating of perceived exertion (RPE) > 17. A calibrated metabolic cart (ParvoMedics Inc., Salt Lake City, Utah) was used to collect expired gas samples throughout the test. During the final 30 seconds of each stage, HR from a watch and chest strap (Polar Electro, Kempele, FL) and RPE using the Borg Scale<sup>31</sup> were recorded.  $\text{VO}_2$  peak was calculated as the highest 15-second average oxygen uptake achieved before volitional exhaustion.

#### *Metabolic Rate Prediction*

An equation outlined by Pimental and Pandolf<sup>4</sup> was used to approximate the appropriate treadmill speed, grade, and load combination to elicit a  $\text{VO}_2$  approximating  $22.5 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$  during all trials. The equation is outlined below, where W is the subject mass in kg, L is the external load in kg,  $\eta$  is the terrain factor, V is the velocity in  $\text{m}\cdot\text{s}^{-1}$ , and G is the grade in percent. A terrain factor of 1 was selected to represent the surface of the treadmill.

$$\text{VO}_2(\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}) = 1.5W + 2.0(W + L)(L/W)^2 + \eta(W + L)(1.5V^2 + 0.35VG)$$

#### *Familiarization trial*

All subjects were required to complete a 60-minute familiarization trial at least 24 hours post  $\text{VO}_2$  peak test and 7 days before beginning the experimental protocol. Each subject refrained from the consumption of alcohol and caffeine, while at the same time abstaining from exercise for the 24 hours leading up to all trials. Subjects were additionally instructed to fast for 12 hours, consume 500 ml of water 60 minutes before bed the evening before, and refrain from consuming any fluids the morning of all trials. For accountability and standardization across trials, each subject kept a food and fluid consumption log for the 24 hours prior.

Upon entering the lab, subjects voided their bladder, drank 500 ml of water, and sat for 30 minutes in a temperate environment before providing a baseline urine void and nude body weight. Subjects walked for 60 minutes on a motorized treadmill with a 15 kg pack (Fireline Pack NFES# 0674, FedMall, Fort Belvoir, VA) at  $1.3 \text{ m}\cdot\text{s}^{-1}$  with a 5 % grade in an environmental chamber (Tesco Inc., Warminster, PA) set at  $33^{\circ}\text{C}$  with a relative humidity of 30 %. Each subject wore a standard WLFF Nomex shirt (Wildland Firefighter's Shirt, FedMall, Fort Belvoir, VA), flame resistant pants (Flame Resistant Pants, FedMall, Fort Belvoir, VA), a cotton base layer t-shirt (Hanes, Winston-Salem, NC), and their own footwear. No fluid was administered to subjects during this trial. Following the familiarization trial, subjects immediately voided andtoweled off before providing a nude body weight.

The purpose of the 60-minute familiarization trial was to use the change in body weight to predict hourly water loss during the 120-minute experimental trials. Hourly water loss estimates allowed exogenous fluid administration to minimize body weight loss during the experimental trials. Post-exercise nude body weight was subtracted from pre-exercise nude body weight to calculate the rate of fluid loss ( $\text{ml}\cdot\text{h}^{-1}$ ).

## EXPERIMENTAL TRIAL

### *Overview*

Each subject completed 4 trials in a repeated-measures cross over design. Subjects were instructed to abide by the same parameters and restrictions as outlined in the familiarization trial, while adhering to their food and fluid consumption logs. There were 7 days between the first 2 consecutive experimental trials, a 2-week washout period, and 2 more experimental trials separated by 7 more days. Trials were separated in this manner to control for potential heat acclimation.



Subjects were assigned to a fluid composition of plain water or CE solution before being further assigned into completing either a micro-dosing or bolus-dosing trial first. Following the 2-week washout period, subjects switched solutions and administration strategy order, making 4 groups of 4 intervention orders. The fluid interventions included bolus-dosed carbohydrate-electrolyte (BCE), bolus-dosed water (BW), micro-dosed carbohydrate-electrolyte (MCE), and micro-dosed water (MW).

Identical to the familiarization trial, subjects urinated, drank 500 ml of water, and sat for 30 minutes in a temperate environment before providing a baseline urine void and nude body weight. Subjects then dressed in the above-mentioned standardized WLFF uniform and pack before entering the heat chamber. The experimental 120-minute treadmill protocols took place in the above-mentioned conditions while consuming either BCE, BW, MCE, or MW. Heart rate (HR), rectal temperature ( $T_C$ ) (RET-1, PhysiTemp, Clifton, NJ), and skin temperature ( $T_S$ , sensor placed 5 cm above the left nipple on pectoralis major muscle) (SST-1, PhysiTemp, Clifton, NJ) were continuously measured via a portable data logger (SDL200, Extech Instruments, Nashua, NH). Urine was additionally collected during and following exercise. Steady state expired gas was collected during exercise. Nude body weight was additionally recorded following exercise.

#### *Treadmill and fluid administration protocol*

Subjects walked for 2 sets of 55 minutes with 5 minutes standing rest to complete the hour and accommodate urine collection. The treadmill and environmental chamber were set at the above-mentioned speed, grade, temperature, and relative humidity.

All fluids were kept in the heat chamber for 12 hours prior to each trial and served at the temperature of the chamber (33°C) to ascertain identical fluid temperature across trials and mimic fluid ingestion temperature while on the fireline. Fluid composition consisted of either

water or a CE solution (Gatorade Endurance Thirst Quencher, PepsiCo, Chicago, IL). Micro-dosed intake was administered for 120 minutes every 2 minutes from 5-25 minutes, 28-50 minutes, and a final time during the 53rd minute in equal increments ( $22 \text{ doses} \cdot \text{h}^{-1}$ ,  $46 \pm 11 \text{ ml} \cdot \text{dose}^{-1}$ ) with 60 ml syringes (BD, Franklin Lakes, NJ). Bolus-dosed intake was administered near the top of each hour (minute 5 and 65) in 2 equal amounts ( $1005 \pm 245 \text{ ml} \cdot \text{h}^{-1}$ ). Subjects were given 5 minutes to consume the bolus volume. The total fluid volume administered in each trial was identical within subjects, matching the fluid loss predicted from the pre-experiment familiarization trial.

*Heart rate, core temperature, skin temperature, and physiologic strain index*

HR,  $T_c$ , and  $T_s$  were measured throughout the 120-minute duration of exercise and recorded within the last 30 seconds of minutes 5, 53, and 113. HR,  $T_c$ , and  $T_s$  were measured using the above-mentioned equipment. PSI was calculated with an equation developed by Moran et al.<sup>32</sup> where  $T_{ct}$  and  $HR_t$  are simultaneous measurements at the time point of interest and  $T_{c0}$  and  $HR_0$  are initial measurements from the beginning of each trial:

$$\text{PSI} = 5(T_{ct} - T_{c0})(39.5 - T_{c0})^{-1} + 5(HR_t - HR_0)(180 - HR_0)^{-1}$$

*Body Weight, Urine Output, and Sweat Rate*

Nude body weight was measured pre, post, and 60 minutes post exercise. Urine was collected pre, during exercise (60 and 120 minutes), and 60 minutes following exercise. Each urine void was measured for volume and urine specific gravity (USG) (URC-NE, Atago, Cohasset, MA). Sweat rate was calculated using the changes in nude body weight in kg (BW), corrected for urine output in kg (U), fluid consumption in kg (FC), and respiratory water loss in kg (RWL). RWL was calculated per Mitchell et al.<sup>33</sup> and water vapor pressure as determined by Fox et al.<sup>34</sup>:

$$\text{SR} (\text{L} \cdot \text{h}^{-1}) = \frac{[(\text{BW}_{\text{PRE}} - \text{BW}_{\text{POST}} + \text{FC}) - (\text{U} + \text{RWL})]}{h}$$

$$\text{RWL (g} \cdot \text{min}^{-1}) = (0.0019)(\text{VO}_2 \text{ L} \cdot \text{min}^{-1})(44 - \text{Water Vapor Pressure})$$

$$\text{Water Vapor Pressure} = 13.955 - 0.6584 (\text{Temperature}) + 0.0419 (\text{Temperature})^2$$

### *Substrate utilization*

Gas exchange was measured using a calibrated metabolic cart via indirect calorimetry as mentioned above. Steady state expired gas samples were collected at 0-5 minutes during the first hour to act as baseline. Subsequent expired gas samples were recorded during minutes 50-53 and 110-113. Steady state values of  $\text{VO}_2$  ( $\text{L} \cdot \text{min}^{-1}$ ) and  $\text{VCO}_2$  ( $\text{L} \cdot \text{min}^{-1}$ ) were used to calculate total carbohydrate oxidation as demonstrated by Jeukendrup and Wallis<sup>35</sup> with the assumption that protein oxidation was negligible:

$$\text{Carbohydrate Oxidation (g} \cdot \text{min}^{-1}) = (4.344 \cdot \text{VCO}_2) - (3.061 \cdot \text{VO}_2)$$

### STATISTICAL ANALYSIS

A two-way ANOVA (time x trial) with repeated measures was used to evaluate body weight (pre-, post-, and 60 minutes post exercise), HR (5, 60, and 120 minutes during exercise),  $T_C$  (5, 60, and 120 minutes during exercise),  $T_s$  (5, 60, and 120 minutes during exercise), PSI (5, 60, and 120 minutes during exercise), urine by void (60 and 120 minutes during exercise and 60 minutes post exercise), and substrate utilization (5, 60, and 120 minutes during exercise). A one-way ANOVA (trial) with repeated measures was used for RWL during exercise, relative exercise intensity, exercise sweat rate, post-exercise sweat rate, cumulative urine output, exercise urine output, and post exercise urine output. A 2-tailed paired t-test was used to evaluate oxygen consumption ( $\text{VO}_2$ ) measured during exercise and estimated from the Pimental and Pandolf equation<sup>4</sup>. Statistical significance was set at a type I probability error of less than 5 % ( $p < 0.05$ ), with data expressed as *mean*  $\pm$  *SD*. Statistical analysis was done using SPSS (IBM, Chicago, IL) and Excel (Microsoft Corporation, Redmond, WA).

## Results

12 subjects completed all trials. However, 1 subject vomited following one of the trials and was removed from analyses for RWL, body weight, and urine oriented metrics. Another subject was removed from the USG analysis due to an inability to provide a baseline urine void. RWL, body weight, cumulative urine output, urine output by void, and post exercise urine output, and post exercise sweat rate were analyzed with 11 subjects, while USG was analyzed with 10 subjects. Each sample size is appropriately denoted below, otherwise all of other reported data include 12 subjects.

### EXERCISE INTENSITY AND RESPIRATORY WATER LOSS

Relative intensity estimated from the Pimental and Pandolf<sup>4</sup> equation and measured during the experimental trials were not significantly different ( $21.8 \pm 0.6$  and  $22.0 \pm 1.3 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ , respectively,  $p > 0.05$ ). Similarly, relative intensity was not significantly different across trials ( $p > 0.05$ ) (Table 1). Calculated RWL was also not significantly different across trials ( $n = 11$ ,  $p > 0.05$ ) (Table 1).

### BODY WEIGHT AND SWEAT RATE

Body weight immediately post and 60 minutes post exercise was significantly lower than pre-exercise ( $n = 11$ ,  $p < 0.05$ ) (Table 1), and body weight 60 minutes post exercise was significantly lower than post exercise ( $n = 11$ ,  $p < 0.05$ ) (Table 1). However, total body weight loss at the end of the 60-minute post exercise rest period was not significantly different across trials ( $n = 11$ ,  $p > 0.05$ ) (Table 1). Sweat rate during the MCE trial was significantly lower than the BCE, BW, and MW trials ( $n = 11$ ,  $p < 0.05$ ) (Table 1). Post exercise sweat rates were not significantly different across trials ( $n = 11$ ,  $p > 0.05$ ) (Table 1).

### URINE OUTPUT AND URINE SPECIFIC GRAVITY

Cumulative urine output across trials during exercise and following 60 minutes of recovery was not significantly different across trials ( $n = 11$ ,  $p > 0.05$ ) (Table 1). Similarly, urine void by time was not significantly different across trials ( $n = 11$ ,  $242 \pm 169$ ,  $231 \pm 174$ ,  $205 \pm 147$  ml, for 60, 120, and 180 minutes, respectively,  $p > 0.05$ ). USG was significantly lower than baseline at 60, 120, and 180 minutes ( $n = 10$ ,  $1.016 \pm 0.006$ ,  $1.006 \pm 0.004$ ,  $1.006 \pm 0.005$ ,  $1.009 \pm 0.007$ , for baseline, 60, 120, and 180 minutes, respectively,  $p < 0.05$ ), while USG at 120 minutes was significantly lower than 180 minutes ( $p < 0.05$ ).

#### HEART RATE, CORE TEMPERATURE, PHYSIOLOGIC STRAIN INDEX, AND SKIN TEMPERATURE

At 5 minutes, HR in MCE was significantly higher than MW (time x trial interaction,  $p < 0.05$ ), while at 60 minutes HR in the CE trials were significantly higher than the W trials (time x trial interaction,  $p < 0.05$ ) (Figure 1). HR at 60 and 120 minutes was significantly higher than 5 minutes within each trial (BCE, BW, MCE, MW) ( $p < 0.05$ ) (Figure 1), while HR at 120 minutes was significantly higher than 60 minutes within each trial (BCE, BW, MCE, MW) ( $p < 0.05$ ) (Figure 1).  $T_c$  at 60 and 120 minutes was significantly higher than  $T_c$  at 5 minutes ( $p < 0.05$ ), while  $T_c$  at 120 minutes was significantly higher than 60 minutes ( $p < 0.05$ ) (Figure 2), there were no differences across trials. PSI at 120 minutes was significantly higher than 60 minutes ( $p < 0.05$ ) (Figure 3), however there were no differences across trials.  $T_s$  at 60 and 120 minutes were significantly higher than 5 minutes ( $p < 0.05$ ) (Figure 4). There were no differences across trials.

#### CARBOHYDRATE OXIDATION

At 60 and 120 minutes the BCE and MCE trials demonstrated significantly higher rates of total CHO oxidation than baseline and the BW and MW trials (time x trial interaction,  $p < 0.05$ )

(Figure 5). CHO oxidation between dosing styles of identical composition was not significantly different ( $p > 0.05$ ) (Figure 5).

## **Discussion**

The purpose of this study was to evaluate the influence of altered fluid delivery schedules of identical total hourly volume on fluid retention, heat stress, and CHO use during extended exercise in the heat. We hypothesized that, regardless of composition, a micro-dosed fluid administration would improve fluid retention, while the inclusion of a CE solution would provide further fluid retention benefits. We also hypothesized that a micro-dosed oral delivery of CHO would maintain a higher CHO oxidation rate than a bolus-dosed oral delivery. Our findings did not confirm our hypotheses.

Our findings demonstrate similar physiologic outcomes when consuming fluids as either a micro- or bolus-dose ( $22 \text{ doses}\cdot\text{h}^{-1}$  or  $1 \text{ dose}\cdot\text{h}^{-1}$ , respectively) designed to approximate body weight loss during low-moderate intensity ( $22.0 \pm 1.3 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ,  $40.2 \pm 4.5 \%$  of peak  $\text{VO}_2$ ) extended exercise in the heat. Altered fluid delivery schedules were not differentially advantageous in maintaining euhydration status when evaluating body weight loss or urine volume output across trials. However, despite a lower sweat rate during the MCE trial when compared to the BCE, BW, and MW trials, PSI was not different across trials and suggests no thermoregulatory advantage among interventions. Similarly, total CHO oxidation during the BCE and MCE trials were not different, indicating a comparable digestion and subsequent delivery rate to working muscle. These findings do not directly contest previous research examining post-exercise fluid recovery strategies that manipulate fluid volume, composition, or delivery interval<sup>18-22</sup>. The present data instead propose that manipulation and implementation of

commonly supported post-exercise fluid recovery relationships are not necessarily maintained during exercise.

The rationale for this project developed from our previous field research with WLFF that demonstrates the physiologic and fluid delivery demands during wildfire suppression. The use of stable isotope tracers on wildfire assignments have elucidated water turnover rates as high as  $10 \text{ L}\cdot\text{day}^{-1}$  <sup>1,2,6</sup>. These field studies outline the necessity of adequate fluid provision during extended workshifts. Similarly, digital flowmeter drinking technology examining *ad libitum* drinking behavior during wildfire suppression has shown both bolus- and micro-dose type fluid consumption strategies as a function of workshift time and ambient temperature <sup>7</sup>. Fluid intake interval throughout a fireline workshift can average approximately  $10 \text{ drinks}\cdot\text{h}^{-1}$ , with reported hourly water intakes of  $504 \pm 472 \text{ ml}\cdot\text{h}^{-1}$  that reach peak volume at hours 6-13 of the workshift (approximately  $800 \text{ ml}\cdot\text{h}^{-1}$ ) <sup>7</sup>. Consequently, diverse and flexible fluid delivery strategies/schedules can be deployed on fireline assignments, if total hourly volume needs are provided.

Some subjects expressed sensations of low-moderate stomach discomfort during the bolus-dose trials, while the micro-dose trials appeared to attenuate these concerns. However, it is important to note that the subject who vomited immediately following exercise did so at the end of his third experimental visit, an MW trial, without any indication of gastrointestinal discomfort during exercise. He had previously completed both a BCE and MCE trial. Despite this, the advantage to the micro-dose administration strategy is that it could be practically implemented with a reservoir type hydration apparatus but unless automated, would require closer attention to the regularity of dose delivery.

At rest, following a dehydrating bout of exercise, intermittent delivery of CE or higher energy density beverages have been shown to improve fluid balance more effectively than plain water<sup>19,21,22</sup>. Fluids administered in bolus type doses that match fluid losses, but have low sodium compositions, provoke increased rates of urinary output at rest and result in a net negative fluid balance<sup>18-21</sup>. The premise behind intermittent fluid delivery is to attenuate large volume dependent gastric emptying rates and subsequent alterations to plasma volume and osmolality, which would promote higher urine production rates. The present study implemented a micro- and bolus-dosing intervention concurrently with exercise to combine elements of fluid volume, composition, and delivery interval that often work in tandem during fluid recovery. Despite differences in fluid composition and delivery interval, there were no differences in total body weight loss ( $0.6 \pm 0.3$  kg) and cumulative ( $677 \pm 440$  ml), exercise ( $442 \pm 328$  ml), and post-exercise ( $205 \pm 147$  ml) urine outputs across trials.

Antidiuretic hormones increase in response to exercise intensity<sup>36-38</sup>, exercise duration<sup>39-41</sup>, and heat<sup>36</sup>, which can carry over to post-exercise recovery periods between 30 and 120 minutes<sup>20,40</sup>. The present study occurred in a 33°C environment at a %  $\text{VO}_2$  peak of  $40.2 \pm 4.5$  for 120 minutes. This represents previously measured ambient temperature<sup>1,2,7</sup> and work intensities<sup>5</sup> during significant portions of a workshift on the fireline. Delivery of adequate hourly fluid volume concurrently with exercise appeared to diminish any differences that often occur with different fluid delivery schedules and compositions seen at rest.

Fluids in the present study were delivered to accommodate 100% of expected fluid loss as determined from a pre-experimental familiarization trial ( $1005 \pm 245$  ml·h<sup>-1</sup>). The subjects demonstrated a  $0.34 \pm 0.21$  % body weight loss as a result of exercise, which increased to  $0.71 \pm 0.32$  % body weight loss after an additional 60 minutes of recovery with corresponding USG



values of  $1.006 \pm 0.005$  and  $1.009 \pm 0.007$ , respectively. This adheres to current recommendations for fluid balance during exercise<sup>15-17</sup>. Fluids delivered to avoid dehydration by  $> 2\%$  of body weight are often shown to attenuate unfavorable cardiovascular and core temperature responses to exercise<sup>8-13,42</sup>. The MCE trial resulted in, at most, a 70 ml discrepancy in sweat rate. This could have altered the ability to offload heat during exercise, as sweat is a primary driver of heat loss during exercise. However, PSI increased significantly from 60 to 120 minutes across all trials ( $3.6 \pm 0.7$  and  $4.5 \pm 0.9$ , respectively), characterized by increases in both HR ( $114 \pm 16$  and  $119 \pm 19$  beats·min<sup>-1</sup> at 60 and 120 minutes, respectively) and core temperature ( $37.7 \pm 0.4$  and  $38.1 \pm 0.4$  °C at 60 and 120 minutes, respectively). This time dependent drift in both measures occurred at a constant workload despite  $< 1\%$  of fluid loss by body weight. These findings demonstrate that although fluid delivery can be accurately provided in an attempt to optimize fluid balance, it is not solely capable of alleviating accumulated thermal load when the exercise persists at the same absolute intensity in the heat.

A case study captured by Cuddy and Ruby<sup>43</sup> describes the thermal load, drinking behaviors, and movement patterns for a WLFF that experienced heat exhaustion and medical evacuation from the fireline. Despite aggressive hydration strategies at an average rate of 24 doses·h<sup>-1</sup> and an average volume of 840 ml·h<sup>-1</sup>, heat exhaustion occurred approximately 7 hours into the workshift. However, his self-selected work rates (accelerometry derived) were considerably higher than previously measured in nearly 300 WLFF days on assignment. WLFF on assignment modulate total energy expenditure, work rates and/or work:rest ratios to mitigate thermal strain<sup>2,14</sup>. This documented relationship underscores that overemphasizing fluid provision may diminish the safety concerns associated with safe work intensities and work:rest ratios.

Hydration advocates must also recognize that proper management of the environmental condition and work intensity interaction necessitates consideration of altered work:rest ratios. The present results clearly demonstrate that varied fluid delivery methods provided to approximate fluid losses do not showcase an advantage in offloading heat during low-moderate extended exercise. Moreover, minimizing body weight loss with aggressive fluid intake cannot singularly halt accumulated thermal load as demonstrated by the present study's significant drift in HR and core temperature from 60 minutes to 120 minutes.

The present study was able to deliver fluids with a mean CHO administration rate during the CE trials of  $62 \pm 15 \text{ g}\cdot\text{h}^{-1}$ . At the 60- and 120-minute collection points during exercise, total CHO oxidation rates were not different between BCE and MCE trials ( $1.5 \pm 0.3 \text{ g}\cdot\text{min}^{-1}$ ). These findings suggest similarities in the digestion, absorption and eventual oxidation of exogenous CHO regardless of the oral delivery frequency for the same hourly CHO content. Micro- or bolus-dosing CHO can effectively maintain exogenous CHO oxidation across 120 minutes of moderate intensity exercise in the heat, which is comparable to 15-30-minute CHO delivery intervals <sup>27,44,45</sup>.

Fluid delivery into the small intestine is influenced by glucose concentration of the beverage and exercise intensity <sup>23</sup>. The CE solution (Gatorade Endurance Thirst Quencher, PepsiCo, Chicago, IL) used in this study is formulated with a 6% CHO combination of maltodextrin and fructose, which is generally accepted as a more optimal beverage formula <sup>15</sup>. Ingestion of a beverage with a glucose and fructose combination can increase peak rate of exogenous CHO oxidation <sup>26</sup> by mitigating sodium glucose transporter saturation <sup>46</sup>. The exercise intensity of the present study ( $40.2 \pm 4.5 \%$   $\text{VO}_2$  peak) also falls beneath the 70 %  $\text{VO}_2$  peak threshold at which gastric emptying is altered <sup>23</sup>. Together, the beverage formula and exercise

intensity likely allowed CHO to empty and reach working muscle. Supplemental oral CHO feedings during wildfire suppression at a rate of  $40 \text{ g}\cdot\text{h}^{-1}$  have shown increased self-selected work output (accelerometry derived) during latter stages of the day, which may translate to improved vigilance on the fireline <sup>47</sup>. The present findings then indicate that higher amounts of oral ingested CHO could be reasonably implemented and oxidized during higher intensity segments of a prolonged workshift.

#### LIMITATIONS AND CONSIDERATIONS

The current study delivered fluids at a temperature equivalent to the ambient environment ( $33^{\circ}\text{C}$ ) to both control and closely mimic the temperature of fluids regularly ingested on the fireline. However, the temperature of ingested fluids coupled with dosing interval could influence the gastric emptying of the beverage, subsequently altering fluid retention and downstream thermoregulation <sup>48</sup>. The modality, intensity, and work:rest ratio of exercise that an individual is participating in may also alter gastric emptying rates and subsequent fluid retention and thermoregulation. The present study may have benefitted from added hematic measures of plasma osmolality and plasma volume, as well as evaluation of hormonal alterations to provide insight into hydration status and urine volume production. Gastric aspiration techniques coupled with stable isotope labeling of CHO could have ascertained how much fluid was emptied and how it related to the amount of CHO delivered and oxidized. Together, these metrics may give further insight into the mechanisms underlying the physiologic outcomes. Regardless, the main outcome measures clearly demonstrate no significant differences in fluid balance, fluid retention, or thermoregulation across the varied fluid delivery strategies.

## Conclusions

These data demonstrate that fluid retention, physiologic strain, and CHO oxidation during continuous work in the heat are unaffected by varied fluid delivery schedules of equal volume.

However, administration of fluids to closely match fluid losses does not protect against accumulated thermal load at constant work rates. This key finding suggests that attention to work:rest ratios should be prioritized over aggressive hydration to mitigate thermal stress and heat related injury risk. This is well understood and practiced by experienced WLFF<sup>14</sup>.

Regardless, individuals participating in extended, moderate work in the heat can rely on widely varied and flexible fluid delivery strategies as long as fluid volume accommodates individual sweat rates.

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**FIGURE AND TABLE LEGENDS**

**Figure 1:** Heart rate during 120 minutes of extended walking in the heat ( $n = 12$ ). **A** – At 5 minutes MCE is different than MW,  $p < 0.05$ . **B** – At 60 minutes BCE is different than BW and MW,  $p < 0.05$ . **C** – At 60 minutes MCE is different than BW and MW,  $p < 0.05$ . **D** – 60 and 120 minutes are different than 5 minutes within each trial (BCE, BW, MCE, MW),  $p < 0.05$ . **E** – 120 minutes is different than 60 minutes within each trial (BCE, BW, MCE, MW),  $p < 0.05$ .

**Figure 2:** Core temperature during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – Core temperature at 60 and 120 minutes is different than at 5 minutes,  $p < 0.05$ . **B** – Core temperature at 120 minutes is different than at 60 minutes,  $p < 0.05$ .

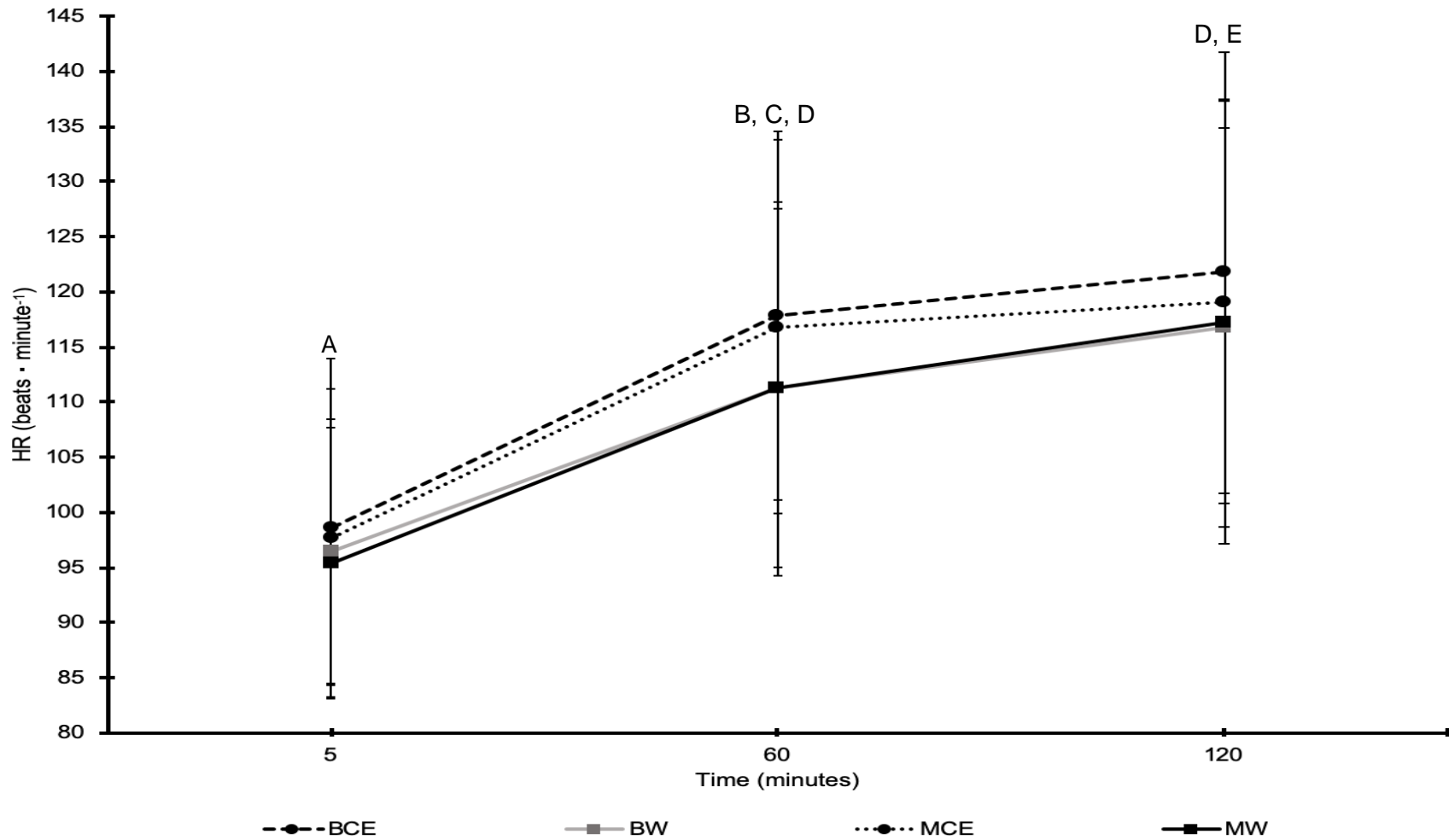
**Figure 3:** Physiologic strain index (PSI) during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – PSI at 120 minutes is different than 60 minutes,  $p < 0.05$ .

**Figure 4:** Skin temperature during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – Skin temperature at 60 and 120 minutes are different than at 5 minutes,  $p < 0.05$ .

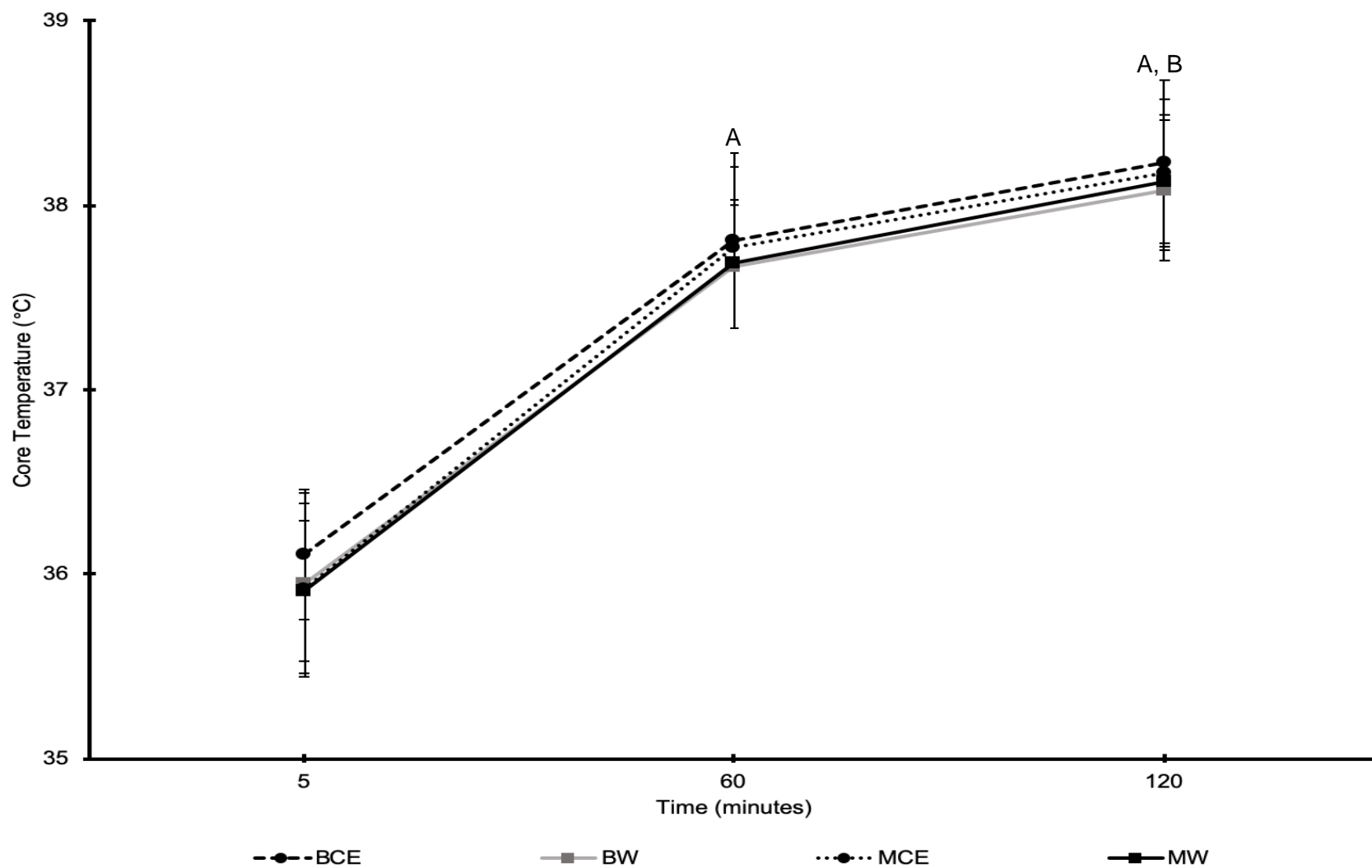
**Figure 5:** Carbohydrate oxidation during 120 minutes of extended walking in the heat ( $n = 12$ ). BCE: bolus-dosing carbohydrate-electrolyte, BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – At 60 minutes and 120 minutes BCE and MCE are different than 5 minutes,  $p < 0.05$ . **B** – At 60 minutes and 120 minutes BCE and MCE are different than BW and MW,  $p < 0.05$ .

**Table 1:** Relative intensity of exercise, body weight pre, post, and 60 minutes post exercise, urine output, respiratory water loss (RWL), and sweat rate (*mean ± SD*)

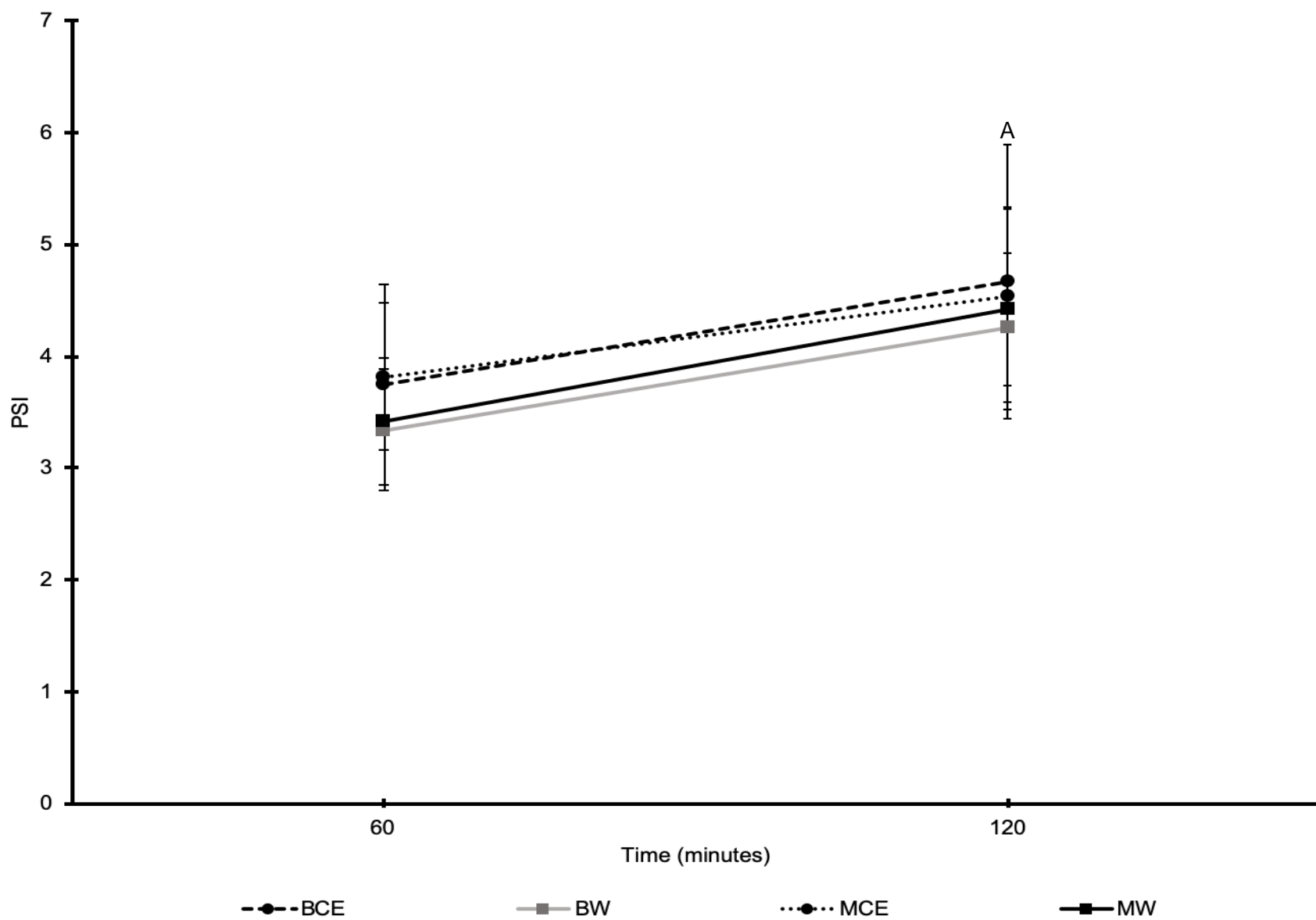
Figures and Tables



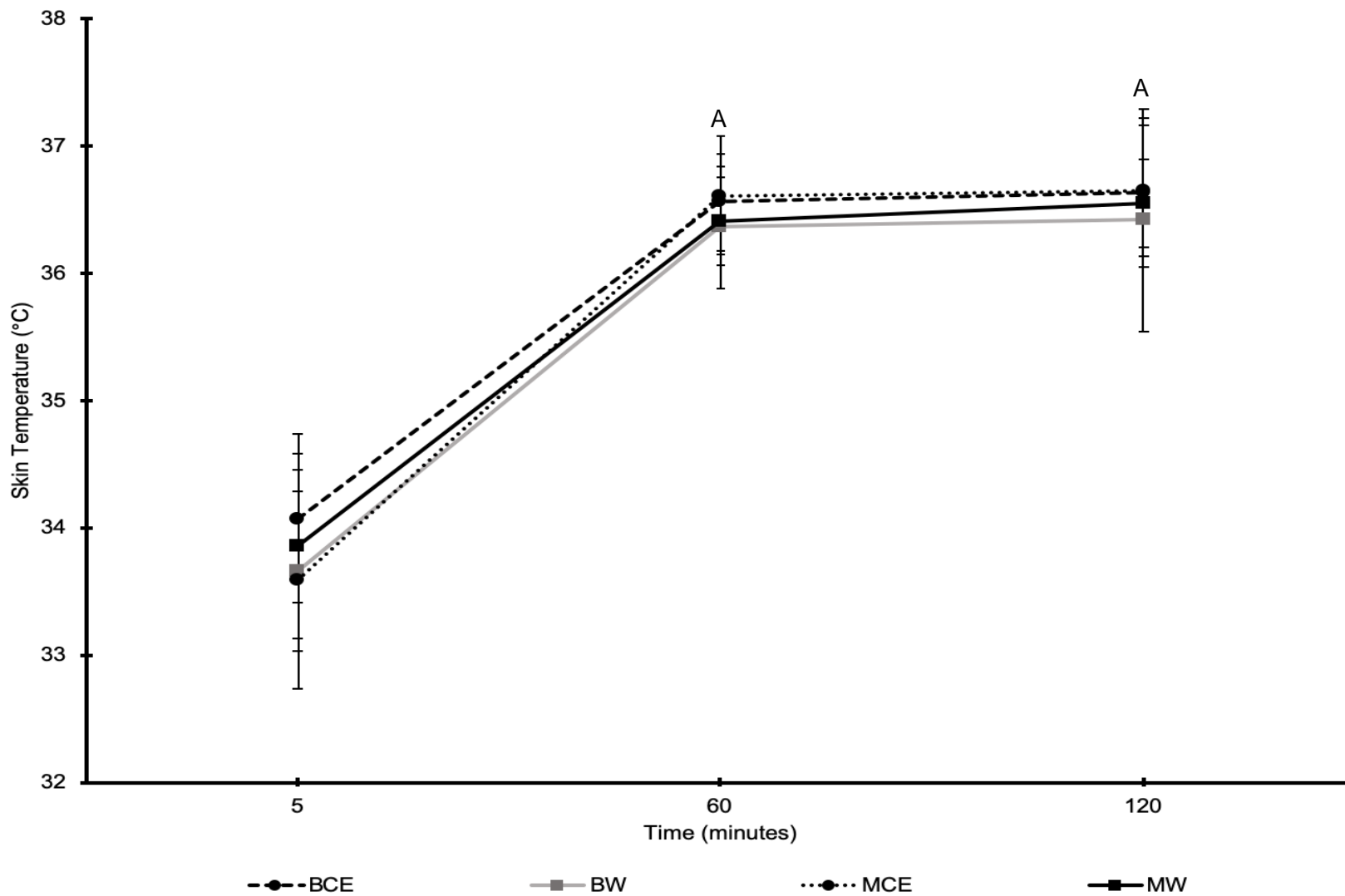
**Figure 1:** Heart rate during 120 minutes of extended walking in the heat ( $n = 12$ ). **A** – At 5 minutes MCE is different than MW,  $p < 0.05$ . **B** – At 60 minutes BCE is different than BW and MW,  $p < 0.05$ . **C** – At 60 minutes MCE is different than BW and MW,  $p < 0.05$ . **D** – 60 and 120 minutes are different than 5 minutes within each trial (BCE, BW, MCE, MW),  $p < 0.05$ . **E** – 120 minutes is different than 60 minutes within each trial (BCE, BW, MCE, MW),  $p < 0.05$ .



**Figure 2:** Core temperature during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – Core temperature at 60 and 120 minutes is different than at 5 minutes,  $p < 0.05$ . **B** – Core temperature at 120 minutes is different than at 60 minutes,  $p < 0.05$ .

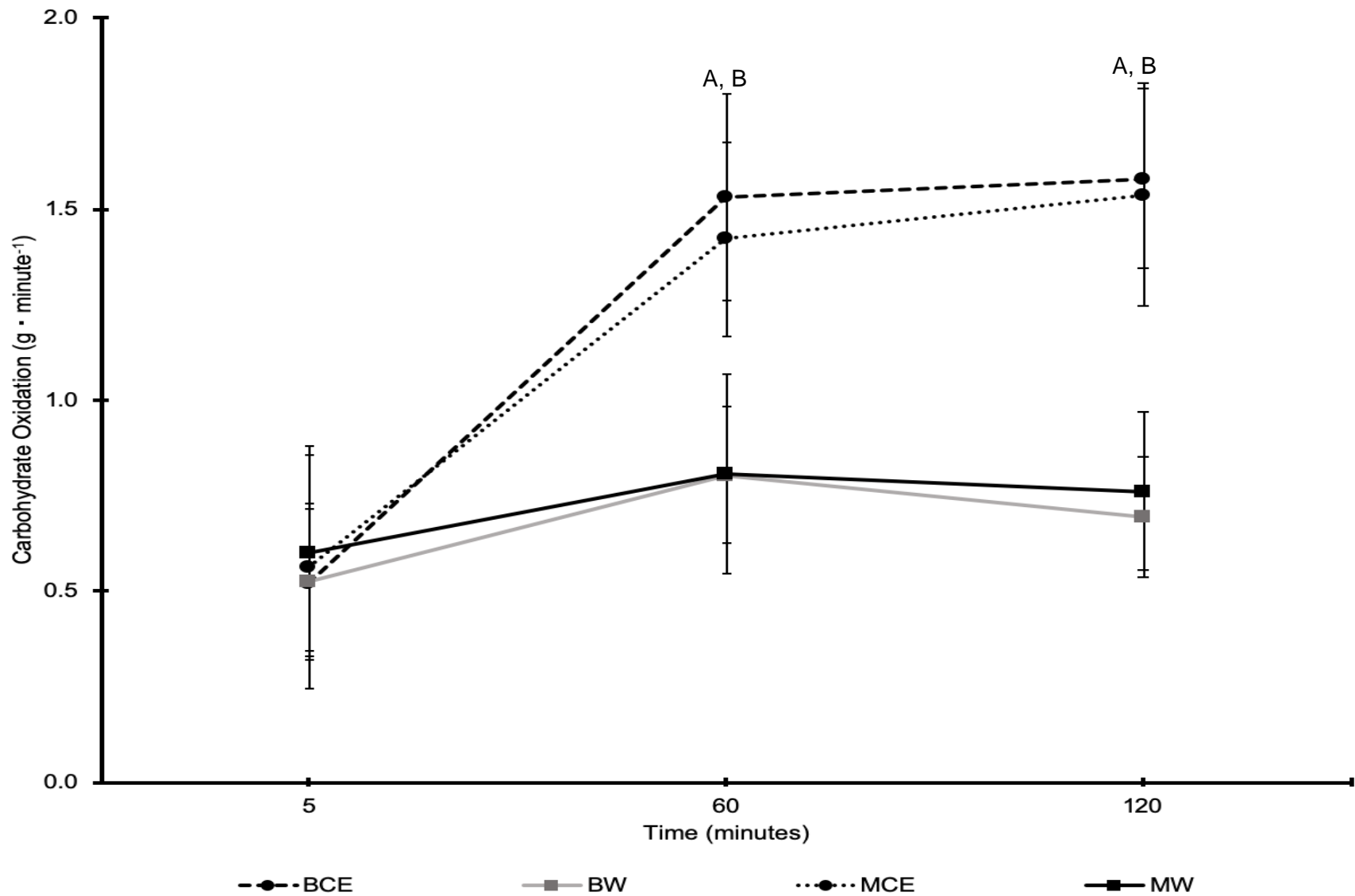


**Figure 3:** Physiologic strain index (PSI) during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – PSI at 120 minutes is different than 60 minutes,  $p < 0.05$ .



**Figure 4:** Skin temperature during 120 minutes of extended walking in the heat ( $n = 12$ ). BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – Skin temperature at 60 and 120 minutes are different than at 5 minutes,  $p < 0.05$ .





**Figure 5:** Carbohydrate oxidation during 2 hours of extended walking in the heat ( $n = 12$ ). BCE: bolus-dosing carbohydrate-electrolyte, BW: bolus-dosing water, MCE: micro-dosing carbohydrate-electrolyte, and MW: micro-dosing water. **A** – At 60 minutes and 120 minutes BCE and MCE are different than 5 minutes,  $p < 0.05$ . **B** – At 60 minutes and 120 minutes BCE and MCE are different than BW and MW,  $p < 0.05$ .

**Table 1:** Relative intensity of exercise, body weight pre, post, and 60 minutes post exercise, urine output, respiratory water loss (RWL), and sweat rate.

	<b>BCE</b>	<b>BW</b>	<b>MCE</b>	<b>MW</b>	<b>Grand Mean</b>
Relative Intensity ( $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ )	22.1 $\pm$ 1.3	21.9 $\pm$ 1.3	22.0 $\pm$ 1.3	21.8 $\pm$ 1.5	22.0 $\pm$ 1.3
Body Weight (kg) ( $n = 11$ )					
<i>Pre</i>	79.4 $\pm$ 11.3	79.6 $\pm$ 10.7	79.5 $\pm$ 11.2	79.2 $\pm$ 10.8	79.4 $\pm$ 10.6
<i>Post</i>	79.1 $\pm$ 11.2	79.3 $\pm$ 10.8	79.3 $\pm$ 11.2	79.1 $\pm$ 10.8	79.2 $\pm$ 10.6 <sup>A</sup>
<i>60 Minutes Post</i>	78.8 $\pm$ 11.2	78.9 $\pm$ 10.70	79.0 $\pm$ 11.2	78.7 $\pm$ 10.8	78.9 $\pm$ 10.6 <sup>A,B</sup>
<i>Pre to 60 Minutes Post Total Loss</i>	-0.6 $\pm$ 0.2	-0.6 $\pm$ 0.3	-0.5 $\pm$ 0.2	-0.5 $\pm$ 0.3	-0.6 $\pm$ 0.3
Cumulative Urine Output (ml) ( $n = 11$ )	634 $\pm$ 455	724 $\pm$ 462	692 $\pm$ 497	659 $\pm$ 399	677 $\pm$ 440
Exercise Sweat Rate ( $\text{L}\cdot\text{h}^{-1}$ ) ( $n = 11$ )	0.93 $\pm$ 0.21	0.91 $\pm$ 0.23	0.86 $\pm$ 0.22*	0.89 $\pm$ 0.22	0.90 $\pm$ 0.21
Exercise RWL ( $\text{g}\cdot\text{min}^{-1}$ ) ( $n = 11$ )	0.196 $\pm$ 0.028	0.193 $\pm$ 0.025	0.193 $\pm$ 0.025	0.192 $\pm$ 0.021	0.193 $\pm$ 0.024
Exercise Urine Output (ml) ( $n = 12$ )	431 $\pm$ 355	469 $\pm$ 338	448 $\pm$ 365	421 $\pm$ 289	442 $\pm$ 328
Post Exercise Sweat Rate ( $\text{L}\cdot\text{h}^{-1}$ ) ( $n = 11$ )	0.12 $\pm$ 0.05	0.12 $\pm$ 0.05	0.13 $\pm$ 0.04	0.13 $\pm$ 0.04	0.12 $\pm$ 0.04
Post Exercise RWL ( $\text{g}\cdot\text{min}^{-1}$ ) ( $n = 11$ )			<i>*Estimated: 0.002</i>		
Post Exercise Urine Output (ml) ( $n = 11$ )	169 $\pm$ 131	225 $\pm$ 166	211 $\pm$ 175	216 $\pm$ 127	205 $\pm$ 147

A – Body weight post and 60 minutes post is different than pre (time effect,  $p < 0.05$ ), B – Body weight at 60 minutes post different than post (time effect,  $p < 0.05$ ), \* – Sweat rate at MCE is different than BCE, BW, and MW (trial effect,  $p < 0.05$ ).