

**The effect of beverage composition on exercise-induced voluntary
dehydration in adolescents**

by

Michaela Caroline Carlson

A thesis submitted to the graduate faculty
in partial fulfillment of the requirements for the degree of
MASTER OF SCIENCE

Major: Exercise and Sport Science
(Biological Basis of Physical Activity)

Program of Study Committee:
Rick Sharp, Major Professor
Joey C. Eisenmann
Douglas King
Kevin Schalinske

Iowa State University

Ames, Iowa

2006

Copyright © Michaela Caroline Carlson, 2006. All rights reserved

UMI Number: 1439850



UMI Microform 1439850

Copyright 2007 by ProQuest Information and Learning Company.
All rights reserved. This microform edition is protected against
unauthorized copying under Title 17, United States Code.

ProQuest Information and Learning Company
300 North Zeeb Road
P.O. Box 1346
Ann Arbor, MI 48106-1346

TABLE OF CONTENTS

ACKNOWLEDGEMENTS	iii
ABSTRACT	v
CHAPTER 1. INTRODUCTION AND PURPOSE	1
Introduction	1
Purpose	4
CHAPTER 2. REVIEW OF LITERATURE	5
Introduction	5
Temperature Regulation in Children	5
Voluntary Dehydration	7
Beverage Preference in Children	8
Sodium and Rehydration Beverages	9
Intestinal Absorption and Gastric Emptying	11
Summary and Conclusions	13
Purpose	14
CHAPTER 3. MATERIALS AND METHODS	15
Study Design and Subjects	15
Experimental Protocol	15
Data Analyses	18
Statistical Analyses	18
CHAPTER 4: RESULTS	20
Subject Characteristics	20
Body Mass/Temperature/Heart Rate/Blood Pressure	20
Psychological/Thirst/Fullness/Hunger Scales	20
Urine	21
<i>Ad libitum</i> Drinking	21
Fluid Balance/% Dehydration/Retention	22
CHAPTER 5: DISCUSSION	23
REFERENCES	27

ACKNOWLEDGEMENTS

I would like to thank my program of study committee members for all of the time and effort that they put into my master's career the past two years. Their support and guidance has brought me much farther in school and in life than I ever thought possible. Dr. Sharp, my major professor, has taught me a lot about patience and independence not only in research but in life; these are characteristics I didn't practice much before, but are now a part of my personality. Dr. King strongly supported me academically and always required just a little more from me than I thought I could accomplish; because of that I have much more confidence in myself. Dr. Eisenmann has been a wonderful mentor in editorial revisions of my thesis as well as mentally in the challenge of completing my thesis. He has supported me a lot from the beginning. Dr. Schalinske has been a great help as he stepped in as a committee member following the unforeseen loss of Dr. Paul Flakoll; therefore, I appreciate his willingness to help in a difficult situation.

I would like to thank my family for all of their support and patience with me. My mother and father were a constant support and ear for me when school became overwhelming and I didn't think that I could do it. My brother and sister were always there to remind me that there were other important aspects to life beyond my master's degree. My husband, Shane, was there for me everyday no matter how difficult I was being. He put up with more late nights and bad moods than I will ever want to admit. I also want to thank my newly born daughter, Makenna, as even though she did not get here until the end she was a constant reminder of what is really important in life and she is what kept me focused the last few months. Lastly, I want to thank all of the friends

that put up with me throughout my master's career. Neil, Megan, Doug, Andrés, Filippo and Brett were daily supporters of mine.

ABSTRACT

This study examined the effects of beverages containing various concentrations of sodium on fluid balance in children during exercise in a thermoneutral environment (22°C). Hypotheses were that the ingestion of chicken noodle soup prior to exercise would cause: 1) increases in *ad libitum* drinking throughout exercise, 2) increases in urine osmolalities, and 3) decreases in urine volumes. Forty-five minutes after 12 children ingested water (0 mmol/L Na⁺), a carbohydrate-electrolyte beverage (16 mmol/L Na⁺), condensed chicken noodle soup (167 mmol/L Na⁺), or dehydrated chicken noodle soup (148 mmol/L Na⁺) the subjects completed 4 bouts of cycling at ~55% VO₂ peak for 20 minutes followed by 10 minute rests. Urine volumes were greater in water and carbohydrate-electrolyte trials. Urine osmolalities were greater in soup trials. Ingestion of chicken noodle soup prior to exercise-induced dehydration led to decreases in urine volumes and increases in urine osmolality, suggesting tendencies of improved water retention.

CHAPTER 1. INTRODUCTION AND PURPOSE

Introduction

Preventing dehydration during exercise is critical to maintain temperature regulation and optimize physical performance and minimize heat related illnesses (27, 36). Dehydration can be avoided if the volume of water intake is equal to the volume of water lost due to sweating, urination, and respiration. Adults and children often may not drink enough fluids during exercise, this is a process known as voluntary dehydration (1, 2, 17). Voluntary dehydration can lead to reductions in plasma and blood volumes which can impair body temperature regulation by radiative, convective, or evaporative cooling mechanisms and compromise physiological function. Given the negative consequences of dehydration, better understanding the role of proper hydration, including the composition of beverages consumed prior to, during, and following exercise, is an important topic.

One important component of a rehydration beverage is sodium because sweating results in the loss of water and sodium. When the body is dehydrated, the kidneys increase sodium reabsorption to create an osmotic drive to retain water. Thus, the addition of sodium in a rehydration beverage increases the restoration of the extracellular space and, subsequently, decreases the plasma volume losses associated with prolonged exercise (8, 35). The maintenance of plasma volumes does not necessarily occur in beverages with sodium concentrations at or below the sodium concentrations of commercially sold carbohydrate-electrolyte beverages (6, 9, 16, 24, 26, 34, 37, 40, 41). The ingestion of beverages with higher sodium concentrations than commercially sold

carbohydrate-electrolyte beverages throughout exercise induced dehydration in adults has demonstrated a decrease of voluntary dehydration (Sharp et. al, unpublished).

Although several studies have examined the process of temperature regulation and fluid balance during exercise in adults (42), few investigations on this topic have been conducted in children (1, 10, 12, 18, 44). There are special concerns with the process of temperature regulation and fluid balance in children due to the differences in the body surface area to mass ratio and sweat rates in children compared to adults. Since children have a higher surface area to mass ratio compared to adults the process of radiative and convective cooling causes the absorption of heat if the ambient temperature is higher than the skin temperature. Also, because children sweat less than adults children often have less heat loss due to evaporative cooling than adults (10, 12, 18, 44). Therefore, temperature regulation is more difficult in children, especially in hot environments. Research on fluid balance in children has shown that thirst is an inadequate method to match fluid intake with fluid loss resulting in voluntary dehydration in children (1, 2, 17). Previous research on children has shown that voluntary dehydration can be attenuated but not prevented with beverages that have acceptable tastes and colors (1, 31, 38, 45, 46). Research on the ingestion of sodium and carbohydrate beverages during exercise in children has demonstrated the prevention of voluntary dehydration due to the increase in *ad libitum* fluid intake (45).

Research on rehydration beverages, especially carbohydrate-electrolyte beverages, have been extensively studied; however, a recent study in adults tested chicken noodle soup as a beverage to enhance rehydration following exercise (37). Chicken noodle soup contains 167 mmol/L Na , which is approximately 10 times more

than commercially sold carbohydrate-electrolyte beverages. The ingestion of chicken noodle soup and chicken broth following a ~2.5% body mass loss resulted in the complete restoration of plasma volume in adults in two hours; whereas the ingestion of water and a carbohydrate-electrolyte beverage resulted in significantly lower plasma volumes (37). Subsequent research in our laboratory suggests that the sodium and carbohydrate contents found in chicken noodle soup may also provide a more efficient method of preventing voluntary dehydration by increasing *ad libitum* fluid intake during exercise in adults (Sharp et. al., unpublished). The ingestion of sodium after exercise/thermal induced dehydration results in the increased release of antidiuretic hormone leading to a retention of water and aldosterone leading to a retention of sodium; therefore, the ingestion of sodium prior to exercise-induced dehydration could result in the retaining of water prior to becoming dehydrated (Sharp et al., unpublished).

A consideration regarding chicken noodle soup is that chicken noodle soup contains more fat, protein, and carbohydrate than previously studied rehydration beverages. Chicken noodle soup is a high carbohydrate meal with approximately 33.3 g/L carbohydrate, 6.25 g/L fat, and 12.5 g/L protein. Therefore, the rate of gastric emptying and intestinal absorption associated with the chicken noodle soup may be important factors to consider. The rate of gastric emptying has been determined to be associated with the caloric density of the meal and not with the volume of the meal (5, 21). High carbohydrate meals have been demonstrated to have a faster gastric emptying rate than high fat meals, and protein has been demonstrated to effect gastric emptying at the same rate as carbohydrates when given isocalorically (4, 7). Previous research on high glucose meals compared to high fat and high protein meals demonstrated that the

high protein and high fat meals had gastric emptying half times of approximately 26 min compared to the 1-2 min for the high glucose meal (5). Intestinal absorption of water has been demonstrated to increase following the ingestion of sodium, carbohydrate, and proteins (14, 15, 19, 36). Therefore, the effects of the soup should increase hydration status based on intestinal absorption reactions; however, the fat, carbohydrate, and protein content could slower gastric emptying enough to negatively effect hydration status.

Purpose

The purpose of this study was to examine the effects of beverages containing various concentrations of sodium, carbohydrates, protein and fats on fluid balance in children when ingested 45 min prior to exercise. It was hypothesized that compared to water the ingestion of chicken noodle soup prior to exercise would: 1) increase *ad libitum* drinking throughout exercise, 2) increase urine osmolalities, and 3) and decrease urine volumes.

CHAPTER 2. REVIEW OF LITERATURE

Introduction

Exercise in adults and children could result in a state of voluntary dehydration that can compromise both athletic performance and result in heat related illnesses. Therefore, understanding rehydration methods during exercise is important in order to prevent these situations. Creating the optimal rehydration beverage for adults and children involves understanding temperature regulation during exercise as well as understanding the effects of different hydration beverages on both adults and children.

The purpose of the following review is to summarize the information pertaining to temperature regulation in children and adults as well as information pertaining to the effect of hydration beverages prior to and following exercise induced dehydration. Topics that will be discussed in this review are: 1) temperature regulation in children, 2) voluntary dehydration, 3) prevention of voluntary dehydration, 4) sodium and rehydration beverages, and 5) intestinal absorption and gastric emptying.

Temperature Regulation in Children

During exercise in all environments, temperature regulation is governed by convective, radiative, and evaporative heat loss. The nature of temperature regulation in children is different from adults due to the greater body surface area to mass ratio in children than in adults, a lower sweating rate in children than in adults, and an increase in convective heat lost in children. In a thermoneutral environment the greater surface area to mass ratio allows for convective heat loss and an increase in evaporative cooling which could, however, lead to greater dehydration. In a hot environment, convective heat loss is ineffective and the greater surface area to mass ratio could have a tendency to generate

heat within the body. A generation of heat production within the body would reduce the rate of evaporative cooling and lead to less water lost in hot environments and less effective temperature regulation (10, 12, 18, 44).

Research on young pre-pubertal boys (8-14 years) and adult men in hot environments has demonstrated lower sweat rates during exercise in young boys compared to adult men (10, 12, 13, 30, 44). The lower sweat rates that occur in young boys compared to men occurs due to a lower sweat production per gland (10, 13, 30, 44). However, the finding is not as apparent in young girls. For example in a previous study, 5 girls and 5 women were subjected to exercise in three environmental conditions (28°C, 35°C, 48°C) and the percentage of total thermal load lost through sweating was demonstrated to be similar in young girls and women. Therefore, puberty does not appear to influence sweat rate in women as it does in men (12).

Reduced sweating rates have also been seen in conjunction with a reduction of *ad libitum* fluid ingestion with children exercising in thermoneutral environments (20). Together with the decrease in sweating rate and a reduction in the concentration of sodium lost in sweat, young boys are losing less water and less sodium than adults during exercise (30). The reduction in sweat rates puts an increased reliance on convective heat loss for temperature regulation during exercise,, an increase in convective heat loss occurs effectively and at greater rates in children in a thermoneutral environment (10, 12, 18, 44). Although temperature regulation methods may be different in children, the tendency for dehydration during exercise in a thermoneutral environment may be less than that of an adult due to less water and sodium lost. However, young children in a hot environment are demonstrating dehydration during exercise.

Cardiovascular responses serve important roles in temperature regulation in children. Because children rely heavily on convection for temperature regulation, (10, 12, 18, 44) both cardiac output and skin blood flow would be expected to increase during exercise in children more than adults. However, children exercising in a thermoneutral environment have demonstrated a lower cardiac output than adults at a given oxygen uptake. The lower cardiac output observed is a result of the reduction of stroke volume at a given O_2 uptake (3, 43). The lower cardiac output, though, has been demonstrated to be compensated with an increase in (a-v) O_2 difference (3, 43).

One explanation for the decrease in stroke volume resulting in a decrease in cardiac output is the associated increase in skin blood flow demonstrated in children compared to adults (22). Increased skin blood flow with a lower cardiac output could result in less blood flow to the internal blood flow leading to the lower level of tolerance, especially with exercise in the heat. However, Rowland et al. (39) demonstrated that there are not significant differences in the cardiac function of adults and children (11-12 years) during exercise with the exception of a higher maximal heart rate in children.

Voluntary Dehydration

Voluntary dehydration is a negative body water balance achieved when water is lost due to urination, sweating, and respiration without adequate replacement. Voluntary dehydration occurs in both adults and children. Unflavored water drunk *ad libitum* by both adults and children during exercise in hot environments has resulted in voluntary dehydration (1, 2, 17). Bar-Or et al. (1) demonstrated voluntary dehydration with decreases in body mass in 10-12 year old boys during exercise in a heat chamber at 39°C. The subjects not only experienced voluntary dehydration but they also experienced

difficulties in temperature regulation with a mean increase of 0.28°C for every 1% body mass lost.

Exercise induced dehydration can cause difficulties in temperature regulation due to reductions in blood volume (plasma volume) induced by sweating. To maintain adequate temperature regulation sufficient blood volume must be sustained to allow for radiative, convective, and evaporative cooling. Therefore, the body adjusts to reductions in plasma volume by mobilizing fluid from the intracellular spaces (36, 27). The mobilization of fluid from the intracellular space is directly related to the sodium concentrations lost during sweating in adults (36). Likewise, Dill et al. (11) found that the amount of voluntary fluid replacement for individuals exercising in desert conditions was directly correlated with the concentration of chloride in sweat. However, the concentration of sodium in sweat tends to be lower in children than the concentration of sodium in adult sweat (30). Therefore, because prepubertal boys have lower sweat rates, total sodium lost due to sweating could be less in children than in adults (30).

Beverage Preference in Children

One method of preventing voluntary dehydration in children is by providing a rehydration beverage that tastes good. Research has demonstrated that when children are offered drinks of their choice during exercise, volume or quantity of ingestion increases (1). Conversely, even though preferred beverages were consumed, voluntary dehydration was not prevented in children (1, 31, 38, 45, 46). Voluntary dehydration was prevented when carbohydrates and NaCl were added to the preferred beverage. In the study by Wilk and Bar-Or (45), the total *ad libitum* drink intake during endurance exercise in the heat was 44.5% higher in the grape flavored water (preferred beverage) trial compared to

the plain water trial among 9-12 year old boys. However, the total drinking intake was 91% higher in the grape flavored water plus 6% carbohydrate and 18mmol/l NaCl than the plain water trial. Therefore, although flavor is important, carbohydrate and NaCl appear to also play a role in preventing voluntary dehydration.

Sodium and Rehydration Beverages

The composition of rehydration beverages is important to allow adequate rehydration. In response to a loss in plasma volume due to dehydration the body produces a hormonal response with aldosterone and vasopressin in order for the kidneys to retain sodium and water. The sodium retention allows for an osmotic drive of water in to the body. Sodium retention leads to a decrease in urine osmolality and an increase in plasma osmolality (36). Therefore, the sodium is one key component of a rehydration beverage.

Previous studies in adults have indicated that rehydration with the addition of sodium leads to improved fluid retention and plasma volume restoration (8, 9, 36). In order to prevent dehydration, the concentration of sodium in rehydration beverages needs to be higher than that of common carbohydrate electrolyte beverages. Common carbohydrate electrolyte beverages contain more sodium than water and, therefore, lead to improvements of dehydration compared to water, but the carbohydrate electrolyte beverages do not completely prevent dehydration (6, 9, 16, 24, 26, 34, 37, 40, 41).

Although previous fluid balance studies have focused on carbohydrate-electrolyte beverages, a more recent study looked at the ingestion of high sodium liquid foods. In a study by Ray et al. (37), subjects exercised until they had lost ~2.5% body mass. The subjects then rehydrated with 350 ml of either water, chicken broth (109.5 mmol/L Na⁺),

a carbohydrate-electrolyte beverage (16 mmol/L Na), or chicken noodle soup (167 mmol/L Na) and then finished rehydration with the ingestion of water at volumes equal to body mass lost. Plasma volumes were restored with chicken broth and the chicken noodle soup. Urine volumes were also higher for the carbohydrate-electrolyte beverage and chicken broth. Therefore, the ingestion of the high sodium chicken noodle soup and chicken broth resulted in the restoration of plasma volume including also a reduction of urine volume (37).

In a previous study by Wilk et al. (46), researchers administered a grape flavored solution with 6% carbohydrate and 18.0 mmol/L NaCl *ad libitum* to twelve, 10-12 year old boys throughout six 70 min intermittent exercise sessions. In 67 out of 72 sessions the subjects maintained a positive fluid balance. Further research on rehydration beverages with the addition of sodium and carbohydrate has demonstrated a decrease in voluntary dehydration in children (38, 45). However, other studies using beverages with sodium levels up to 18.5 mmol/L failed to yield similar results (29, 32). Possibly, the concentrations of sodium were not great enough to show any improvements.

Based on previous research, the ingestion of sodium fortified beverages can lead to a reduction in voluntary dehydration due to increased *ad libitum* drinking when ingested. A pre-exercise ingestion study conducted in our laboratory, demonstrated that the ingestion of sodium fortified beverages (chicken noodle soup) prior to 90 min of steady state exercise and time to work time trial significantly improved fluid balance compared to water and Gatorade due to greater *ad libitum* fluid intake and improved fluid retention (Sharp et. al., unpublished). A resting study done examined the effects of either water, a high sodium and high potassium beverage, chicken noodle soup, or water with

the same sodium concentration as soup from an already existing hydrated state in adults demonstrated an increase in plasma volume with ingestion of the salt water and chicken noodle soup compared to the water or the sodium/potassium beverage. Salt water and chicken noodle soup also resulted in greater thirst responses and reductions in total urine volume through 2 hours of rest (Johannsen et al., unpublished). Therefore, the ingestion of a high sodium beverage prior to exercise could lead to an increase in *ad libitum* water drinking as well as better retention of that water.

Intestinal Absorption and Gastric Emptying

Regardless of sodium, the beverages previously used in rehydration studies did not vary in composition. However, chicken noodle soup involves the ingestion of not only sodium but also the carbohydrates, fats, proteins, vitamins, and minerals that are within the chicken noodle soup. Previous research on the ingestion of meals with beverages has demonstrated the restoration of body mass and plasma volume following dehydration (25, 42). Therefore, understanding the gastric emptying and intestinal absorption associated with the composition of chicken noodle soup is important for understanding the effects of hydration.

The rate of gastric emptying has been demonstrated to have an inversely proportional relationship to the caloric density of the meal ingested. However, the nature of these calories seems to play only a minor role in the rate of gastric emptying (5, 21). Chicken noodle soup contains approximately 33.3 g/L carbohydrate, 6.25 g/L fat, and 12.5 g/L protein. Previous research on the oral ingestion of a high fat soup (68.9% energy from fat, 27.4% from carbohydrate, 2.8% from protein) and a high carbohydrate soup (19.7% energy from fat, 77% energy from carbohydrate, 3.2% from protein)

demonstrated gastric emptying to be significantly faster in the high carbohydrate soup compared to the high fat soup (7). Therefore, chicken noodle soup would be suggested to have a faster gastric emptying rate because of the higher carbohydrate content compared to fat content.

The rate of gastric emptying is also dependent on the composition of the solution ingested and not the volume of the solution ingested. High concentrations of fat have been demonstrated to slow down gastric emptying significantly. A primarily all-glucose solution with no protein and no fat has a gastric emptying half time of 1-2 min. In the same study a solution that contained 18.3 g of protein, 15.0 g of glucose, and 17.6 g fat slowed down but still had a gastric emptying half time of 26.4 min (5). Isocaloric concentrations of protein and carbohydrate have similar effects on gastric emptying because of the hydrolysis of proteins and the stimulation of the osmoreceptors in the duodenum by the by-products of protein (4).

After the hydrolyzed components of soup exit the stomach, intestinal absorption continues the process of digestion. The increased concentration of sodium in the soup increases intestinal fluid absorption and restoration of plasma volume due to the increase in extracellular fluid space (36, 37). Previous research on intestinal absorption following the ingestion of glucose has demonstrated an increase in the absorption of water in the jejunum (14, 15). Free amino acids, like alanine and glycine, have also been demonstrated to stimulate fluid absorption in the intestine and increase water absorption linearly with increasing concentration of amino acids (19).

Therefore, the rate of gastric emptying following the ingestion of chicken noodle soup may decline due to the added components of the soup, especially the small amount

of fat, but intestinal fluid absorption would be expected to increase due to sodium, carbohydrate, and protein contents.

Summary and Conclusions

Despite allowing adults and children the opportunity to drink water *ad libitum* during exercise, humans may enter a state of voluntary dehydration at the end of exercise. Therefore, developing strategies for finding a beverage that can prevent dehydration is important. Previous research conducted on rehydration beverages in adults has demonstrated the need for high concentrations of sodium to be present in the rehydration beverage. High sodium beverages cause a hormonal response of the body to reabsorb the sodium as well as a follow-up response of the body to retain water.

Exercising children exhibit differences in temperature regulation responses that could affect hydration status during exercise as well as modify rehydration methods in children. Children have a larger body surface area to mass ratio that allows for more effective convective heat loss in a thermoneutral environment. However, the larger body surface area to mass ratio also leads to less effective heat loss in hot environments and possibly an increase in the rate of heat gain due to a decrease in sweat evaporation. As long as the child is in a thermoneutral environment there should not be a temperature regulation difference (i.e. increase in core temperature) between adults and children. Young boys also have a lower sweat rate than that of adult males. In a hot environment young boys sweat less and have less effective convective heat loss than adults. Children also show less sodium concentrations in sweat than adults. Therefore, the sodium replacement needs in a rehydration beverage for children may not be as high as for adults.

Rehydration beverages studied on adults following exercise induced-dehydration have demonstrated that commercially sold carbohydrate-electrolyte beverages do not contain enough sodium to retain water and prevent dehydration. However, chicken noodle soup has been demonstrated to contain enough sodium to retain water and prevent dehydration. Another study done at rest where either, water, a sodium/potassium beverage, chicken noodle soup, or salt water was ingested from an already hydrated state demonstrated that chicken noodle soup and salt water resulted in a better maintenance of plasma volume than the water or the sodium/potassium beverage. Therefore, ingesting a sodium fortified beverage prior to exercise-induced dehydration would prepare the body to better maintain a hydrated state throughout exercise. Recent work from our laboratory, demonstrated that the ingestion of high sodium chicken noodle soup prior to a 2-hour exercise bout significantly improved fluid balance at the completion of exercise compared to water and Gatorade.

Purpose

The purpose of this study was to examine the effects of beverages containing various concentrations of sodium, carbohydrates, protein and fats on fluid balance in children when ingested 45 min prior to exercise. It was hypothesized that compared to water the ingestion of chicken noodle soup prior to exercise would: 1) increase *ad libitum* drinking throughout exercise, 2) increase urine osmolality, and 3) and decrease urine volume.

CHAPTER 3. MATERIALS AND METHODS

Study Design and Subjects

The study design consisted of four, 3 hour trials in a randomized format, where the effects of exercise-induced dehydration were examined following ingestion of four different beverages in a thermoneutral environment (~22°C, 27% relative humidity). Twelve healthy children (6 boys, 6 girls) ages 12-16 years participated in this study. Prior to initiation of the study, medical history forms were completed by the participant and a written informed consent and assent was signed by the guardian and participant, respectively. The study was approved by the Iowa State University Institutional Review Board.

One week prior to the experimental protocol, each participant underwent a graded exercise test to exhaustion on an electronically-braked cycle ergometer (Lode BV, Groningen, The Netherlands) to determine peak oxygen consumption ($\text{VO}_{2\text{peak}}$). Before the start of the $\text{VO}_{2\text{peak}}$ test height and mass were measured according to standard methods (23). Respiratory gas measurements (Physio-dyne Instrument Corp., Quogue, NY), heart rate (HR) (Polar, Electro Oy, Finland), ratings of perceived exertion (RPE), and psychological affect scales were measured at the end of each power output stage. Following the $\text{VO}_{2\text{peak}}$ test, the participants were familiarized with the protocol of the experimental trials.

Experimental Protocol

The experimental protocol is outlined in Figure 1. Participants were asked to fast for 8 hours from food and beverage (except water) prior to each of the 4 experimental trials. Each trial was separated by one week to allow for adequate rest. Two hours prior

to reporting to the laboratory the participants ingested a VitalSense core temperature pill (Minimitter) with 8oz of water. The VitalSense core pill has been shown to be a reliable and valid when compared with rectal temperature ($R^2=0.80$) (28). When the participants entered the lab a pre-ingestion (Pre-I) urine sample was collected. The participant's nude body mass was recorded (Pre-I) and a HR monitor (Polar, Electro Oy, Finland) was fitted on the participant. The participant was then seated for 5 min and Pre-I HR, blood pressure, and temperature were recorded. Pre-I ratings of perceived thirst (sliding scale; 0=not thirsty at all, 10=very very thirsty), fullness/hunger (likert scale; 0 = No, not at all, 10 = Yes, very much), and psychological feeling and arousal (likert scale; +5 = feel very good, -5 = feel very bad and 1 = low arousal, 5 = high arousal) (PA, PF) were also measured. These scales gave an overall impression of how thirsty, hungry, and full the subjects were, as well as their overall feeling and how excited or aroused they were. The participants then ingested 355mL of either water (W), carbohydrate-electrolyte beverage (CE), condensed chicken noodle soup (CS), or dehydrated chicken noodle soup (DS). The beverage compositions are shown in Table 1. After ingestion of the experimental beverage the participant sat for 35 min; after which, pre-exercise (Pre-Ex) body mass, HR, blood pressure, temperature, and ratings on the thirst, fullness, and PA/PF were measured. Exactly 45 min post-ingestion, the exercise trial began on the cycle ergometer (Lode BV, Groningen, The Netherlands). The exercise protocol consisted of four, 20-min cycling bouts separated by 10-min rest periods. Each 20-min cycling bout ranged from 40%-70% $\text{VO}_{2\text{peak}}$ intensity and was completed using the protocol in Figure 2. The average intensity of the cycling bout was calculated to be ~55% $\text{VO}_{2\text{peak}}$. During the last min of the 20-min exercise bout, HR, blood pressure, temperature, and ratings on

the thirst, fullness/hunger, RPE, and PA/PF were measured (Ex-0, Ex-30, Ex-60, Ex-90). At the completion of the 20 min of cycling, a nude body mass was taken and the subject then rested for 10 min (Ex-0, Ex-30, Ex-60, Ex-90). Following the measurements of the final cycling bout, a post exercise urine sample and volume were collected (Post-Ex) after which a Post-Ex body mass was measured. Throughout the exercise and rest periods, participants were allowed to drink water *ad libitum*. The participant's water bottles were weighed prior to and after each drink throughout the trial without the subjects' knowledge. On the final trial, standing height and sitting height were also measured to estimate the biological maturity status using a non-invasive maturity offset equation (33). The maturity offset equation estimates how far the subject is from the age when their maximum velocity in statural growth occurs (peak height velocity). The approximation of biological maturity status was used as a covariate given the age range of the subjects and the influence of puberty on temperature regulation and other physiological functions considered here.

Equation 1 (Male maturity offset)

$$\text{Maturity Offset} = -9.236 + 0.0002708 (\text{Leg Length} \times \text{Sitting Height}) - 0.001663 (\text{Age} \times \text{Leg Length}) + 0.007216 (\text{Age} \times \text{Sitting Height}) + 0.02292 (\text{Mass} / \text{Height}) \quad (33)$$

Equation 2 (Female maturity offset)

$$\text{Maturity Offset} = -9.376 + 0.0001882 (\text{Leg Length} \times \text{Sitting Height}) + 0.0022 (\text{Age} \times \text{Leg Length}) + 0.005841 (\text{Age} \times \text{Sitting Height}) - 0.002658 (\text{Age} \times \text{Mass}) + 0.07693 (\text{Mass} / \text{Height}) \quad (33)$$

Data Analyses

Drinking behavior was determined by the volume of each drink and total volume as well as number of drinks per stage and total time. Urine samples were analyzed for specific gravity using spectral refractometry (Leica TS 400, Leica Microsystems, Inc., New York) and for osmolality using freezing point depression (Westcor, Inc., Utah). Finally, the urine samples were analyzed for electrolyte concentrations of Na⁺ and K⁺ using digital flame photometry (Cole-Parmer, Chicago, Ill.).

The main outcome variables were the volume of water ingested *ad libitum*, the volume of urine lost, and the urine osmolality. Fluid balance was calculated using the following equation: ((PostEx BW (kg) - PI BW (kg))). Percent dehydration was calculated using the following equation: $[(\text{PostEx BW (kg)} - \text{Pre-Ex BW (kg)}) / \text{Pre-Ex BW (kg)}] * 100$. Percent retention was calculated using the following equation: $[(\text{water intake (ml)} - \text{urine volume (ml)}) / \text{water intake (ml)}] * 100$.

Statistical Analyses

The data were analyzed using JMP 5.1 (SAS, SAS Institute, Inc., Cary, North Carolina) statistical software with various two-way (trial and time) repeated measures analyses of co-variance (ANCOVA) tests for urine osmolality, body mass, HR, blood pressure, temperature, thirst, PA/PF, hunger/fullness, and urinary Na⁺/K⁺ concentration. A one-way repeated measures ANCOVA was used for urine volume, total volume of water ingested, total number of drinks taken, percent dehydration, and fluid balance. Percent retention was analyzed using a Wilcoxon nonparametric test. Maturity offset was used as a covariate in all analyses. Significant time and trial differences were declared when $P \leq 0.05$. The order of the trials was also used as a covariate when analyzing

temperature, RPE, and total water intake because of a main effect of trial order found in both of these variables. A post hoc Tukey test was completed when a significant effect was rated by ANOVA.

CHAPTER 4. RESULTS

Subject Characteristics

Table 2 provides the descriptive statistics for the study sample. Mean height, mass, and BMI for girls and boys approximated the 50th percentile on the CDC growth chart (data not shown). There were no significant differences between males and females in this study and the genders were combined for statistical analyses.

Body Mass/Temperature/Heart Rate/Blood Pressure

Figure 3 shows the time related changes in body mass to be similar during all four trials. There were also no differences in the body mass among the different times points. Core temperatures were analyzed and are illustrated in Figure 3. There were significant main effects for trial and time. The main effects for trial showed that the core temperatures for trials CE and CS were significantly greater than W ($P=0.0048$). The time effects showed that the temperature significantly increased from Pre-I and Pre-Ex to Ex-0 and then again to Ex-30, Ex-60, and Ex-90 (Figure 1; $P<0.0001$). HR and mean arterial blood pressure also increased from Pre-I and Pre-Ex to Ex-0, Ex-30, Ex-60, and Ex-90 but did not vary by trial (Figure 3; $P<0.0001$).

Psychological/Thirst/Fullness/Hunger Scales

Psychological, thirst, and fullness/hunger responses are shown in Figure 4. Feeling and arousal did not change within or between the trials. Subjects reported greater fullness during CS and DS compared to W ($P<0.0001$). All trials demonstrated that Pre-Ex values for fullness were significantly greater than the Pre-I, Ex-60 and Ex-90 values. Subjects were significantly more hungry in W and CE than DS ($P<0.0001$). Also, subjects were more hungry in Ex-90 than all other time points and subjects were more

hungry in Ex-60 than Pre-Ex and Ex-0 ($P<0.0001$). The thirst scales showed that the participants were thirstier during Ex-30, Ex-60, and Ex-90 than Pre-I and Pre-Ex ($P<0.0001$). Significant time effects in RPE were found where RPE values significantly increased with each exercise time period but there were no differences among the beverage trials ($P<0.0001$).

Urine

Table 3 shows the results of the composition of the urine samples. There were significant main effects for time as the osmolality, specific gravity, and sodium values were all significantly different from Pre-I to Post-Ex ($P<0.0001$). Furthermore, trial effects were shown for osmolality, specific gravity, and sodium concentration with trial CS having significantly greater values than W ($P=0.0166$, $P=0.0358$, $P=0.0103$). CS sodium concentration was also significantly greater than CE sodium concentration ($P<0.0001$). Post-Ex osmolality and specific gravity values were significantly greater in CS and DS than W and CE ($P<0.0001$). Post-Ex sodium concentration was significantly greater in CS and DS compared to W and CE ($P<0.0001$). Also, the Post-Ex sodium concentration in CS was significantly greater than DS ($P<0.0001$). The Post-Ex potassium concentration was significantly greater in CS and DS than W and CE ($P<0.0001$). The Post-Ex potassium concentration in CE was also significantly greater than Post-Ex CS ($P<0.0001$). Total urine volume was greater in W and CE compared to CS and DS (Figure 5; $P<0.0001$).

Ad libitum Drinking

There were no significant main effects for trial found in the total volume of water intake (Figure 6; $P=0.9480$). Likewise, no significant differences were found for the total

volume ingested or the number of drinks between trials (Table 4). Significant differences were found in time points ($P < 0.0001$). Significantly lower volumes of water and number of drinks were ingested in the first hour of the trials compared to the second and third hours (Tables 4 and 5). Significantly lower volumes of water and number of drinks were ingested Pre-Ex than compared to during the exercise/rest time periods (Tables 4 and 5; $P = 0.0012$).

Fluid Balance/%Dehydration/Retention

There were no differences in trials found in fluid balance (Figure 7; $P = 0.5324$). There were also no differences in percent dehydration (Figure 8; $P = 0.4189$). There were no differences in the absolute volume of retained fluid (Figure 9; $P = 0.3751$).

CHAPTER 5. DISCUSSION

The results of this study indicate that the ingestion of chicken noodle soup before exercise did not stimulate *ad libitum* fluid intake during subsequent exercise and had no effect on body fluid balance. The chicken noodle soup did, however, result in reduced urine volume and increased urine osmolality throughout the exercise regimen.

The only hypothesis not supported in the current study dealt with no significant differences found in *ad libitum* drinking between trials. Previous research on pre-hydration beverages in adults has demonstrated differences in *ad libitum* drinking throughout the trials depending on the beverage consumed prior to the exercise bout (Johannsen et. al., unpublished). However, this previous study had a larger sample size (n=20) as well as a different exercise protocol. The exercise protocol in the previous study was a higher intensity including 90 min of steady state exercise at ~57% $\text{VO}_{2\text{peak}}$ followed by a 30 min time trial. The exercise bout and the ingestion of chicken noodle soup also resulted in higher plasma osmolality (Johannsen et. al., unpublished). Previous research has suggested that an increase in plasma osmolality can influence the thirst behavior (36). Plasma osmolality was not measured in the current study.

A previous study by Rivera-Brown et al. (38) in children demonstrated an increase in *ad libitum* drinking with a flavored water that contained 18mmol Na^+ compared to unflavored plain water. The increase in *ad libitum* drinking led the subjects to a state of euhydration following exercise where as the plain water lead to a state of dehydration. The subjects in the current study did not demonstrate significant levels of dehydration following any of the trials. However, in the current study the subjects exercised in a thermoneutral environment and at 55% $\text{VO}_{2\text{peak}}$ where as the subjects in

the Rivera-Brown et al. study exercised at 60% VO_2 peak in a heated environment (30°C) (38). Therefore, the exercise protocol of the current study may not have been intense enough with too cool of an environment to elicit negative fluid balances and, consequently, different drinking behaviors. Also, in the study by Rivera-Brown et al., the subjects drank the test beverage *ad libitum* throughout the exercise whereas in the current study the subjects drank the test beverage only prior to exercise and then plain water throughout the exercise. Perhaps a flavored beverage may have caused a larger *ad libitum* drinking response.

The condensed chicken noodle soup (167 mmol/L Na^+ ; 6.9 mmol/L K^+) and the dehydrated chicken noodle soup (148 mmol/L Na^+ ; N/A mmol/L K^+) contained higher concentrations of electrolytes than the carbohydrate-electrolyte beverage (16 mmol/L Na^+ ; 3.3 mmol/L K^+) or the water (0 mmol/L Na^+ ; 0 mmol/L K^+). The results of the urine concentration of sodium and potassium as well as the urine osmolality and specific gravity following the ingestion of the condensed chicken noodle soup were similar to the results from an unpublished study in our laboratory on adults that ingested chicken noodle soup prior to exercise (Johannsen et. al., unpublished). However, in the present study there were also differences in the urine sodium concentration between the two soups, with the condensed chicken noodle soup resulting in a higher sodium concentration than the dehydrated chicken noodle soup.

Assessing total water intake throughout each trial as well as total urine volume for each trial is one indicator of water retention. In the present study the total urine volume was greatest following the water and carbohydrate-electrolyte trials. The urine volume results are in accordance with those found in a previous study done on chicken noodle

soup versus water and carbohydrate-electrolyte beverages (Johannsen et. al., unpublished). Although the *ad libitum* water intake was not different among the trials, the urine volumes were less in the chicken noodle soup trials compared to the water and carbohydrate-electrolyte trials. These results suggest that the water and the carbohydrate-electrolyte beverage respond similarly. The results also show a tendency for better retention with the chicken noodle soup trials. Although greater retention was not found in the current study a previous pre-hydration study done in adults demonstrated that percent retention was significantly greater in the chicken noodle soup trial compared to a water trial (Johannsen et. al., unpublished). However, in the Sharp et al. study the soup trial resulted in greater *ad libitum* drinking and less urine volume.

There were no differences found among trials for fluid balance and percent dehydration. Previous research on adults with pre-hydration beverages has shown significantly better fluid balance and less dehydration following the ingestion of chicken noodle soup compared to water (Johannsen et. al., unpublished). However, the previous study had more subjects (20) and a higher intensity exercise protocol coupled with a thermal load.

Previous research on children and *ad libitum* drinking has demonstrated that children have a tendency to voluntarily dehydrate during exercise (1, 2). The results from the current study did not demonstrate large dehydration in the children following exercise; in fact, the fluid balance and percent dehydration were not different from zero (Figure 5 and Figure 6). However, previous studies were conducted in hot environments and the current study was performed in a thermoneutral environment. Therefore, the difference in the ambient temperature among studies may have led to differences in the

ad libitum drinking which in turn resulted in differences in fluid balance and percent dehydration. Many of the subjects that experienced dehydration in the previous studies also exhibited temperature increases of 0.8-1.2°C during exercise; the same changes were exhibited in the current study, however the corresponding weight changes of 1-2.5% body mass were not observed (1, 38).

In conclusion, the ingestion of condensed chicken noodle soup and dehydrated chicken noodle soup prior to exercise in a thermoneutral environment may be more advantageous in decreasing urine volume and possibly improving the retention of fluids throughout exercise while increasing urine osmolality and specific gravity. However, further research is required prior to concluding whether or not these results will lead to a better state of hydration and fluid balance following and throughout exercise.

REFERENCES

1. Bar-Or O, Dotan R, Inbar O, Rotchtein A, and Zonder H. Voluntary hypohydration in 10- to 12-year-old boys. *Journal of Applied Physiology*. 48(1):104-108, 1980.
2. Bar-Or O, Hay JA, Ward DS, Blimkie CJR, MacDougall JD, and Wilson WM. Voluntary dehydration and heat intolerance in cystic fibrosis. *The Lancet*. 339(8795):696-699, 1992.
3. Bar-Or O, Shephard RJ, and Allen CL. Cardiac output of 10- to 13-year-old boys and girls during submaximal exercise. *Journal of Applied Physiology*. 30(2):219-223, 1971.
4. Burn-Murdoch RA, Fisher MA, and Hunt JN. The slowing of gastric emptying by proteins in test meals. *Journal of Physiology*. 274:477-485, 1978.
5. Calbet JAL and MacLean DA. Role of caloric content on gastric emptying in humans. *Journal of Physiology*. 498(2): 553-559, 1997.
6. Candas V, Libert JP, Brandenberger G, Sagot JC, Amoros C, and Kahn JM. Hydration during exercise. *European Journal of Applied Physiology*. 55:113-122, 1986.
7. Cecil JE, Francis J, and Read NW. Comparison of the effects of a high-fat and high-carbohydrate soup delivered orally and intragastrically on gastric emptying, appetite, and eating behavior. *Physiology and Behavior*. 67(2): 299-306, 1999.
8. Costill DL, Fink BW, and Nelson R. Exercise induced sodium conservation: changes in plasma renin and aldosterone. *Medicine and Science in Sports*. 8(4):209-213, 1976.
9. Costill DL and Sparks KE. Rapid fluid replacement following thermal dehydration. *Journal of Applied Physiology*. 34(3):299-303, 1973.
10. Delmarche P, Bittel J, Lacour JR, and Flandrois R. Thermoregulation at rest and during exercise in prepubertal boys. *European Journal of Applied Physiology*. 60:436-440, 1990.
11. Dill DB, Soholt LF, and Oddershede IB. Physiological adjustments of young men to five-hour desert walk. *Journal of Applied Physiology*. 40(2):236-242, 1976.
12. Drinkwater BL, Kupprat IC, Denton JE, Crist JL, and Horvath SM. Response of prepubertal girls and college women to work in the heat. *Journal of Applied Physiology*. 43(6):1046-1053, 1977.

13. Falk B, Bar-Or O, Calvert R, and MacDougall D. Sweat gland response to exercise in the heat among pre-, mid-, and late-pubertal boys. *Medicine and Science in Sports and Exercise*. 24(3):313-319, 1992.
14. Fordtran JS. Stimulation of active and passive sodium absorption by sugars in the human jejunum. *The Journal of Clinical Investigation*. 55:728-737, 1975.
15. Gisolfi CV, Summers RD, Schedl HP, and Bleiler TL. Effect of sodium concentration in a carbohydrate-electrolyte solution on intestinal absorption. *Medicine and Science in Sports and Exercise*. 27(10): 1414-1420, 1995.
16. González-Alonso J, Heaps CL, and Coyle EF. Rehydration after exercise with common beverages and water. *International Journal of Sports Medicine*. 13(5):399-406, 1992.
17. Greenleaf JE and Sargent II F. Voluntary dehydration in man. *Journal of Applied Physiology*. 20(4):719-724, 1965.
18. Haymes EM, Buskirk ER, Hodgson JL, Lundegren HM, and Nicholas WC. Heat tolerance of exercising lean and heavy prepubertal girls. *Journal of Applied Physiology*. 36(5):566-571, 1974.
19. Hellier MD, Thirumalai C, and Holdsworth CD. The effect of amino acids and dipeptides on sodium and water absorption in man. *Gut*. 14: 41-45, 1973.
20. Horswill CA, Passe DH, Stofan JR, Horn MK, and Murray R. Adequacy of fluid ingestion in adolescents and adults during moderate-intensity exercise. *Pediatric Exercise Science*. 17:41-50, 2005.
21. Hunt JN and Stubbs DF. The volume and energy content of meals as determinants of gastric emptying. *Journal of Physiology*. 245: 209-225, 1975.
22. Koch G. Muscle blood flow in prepubertal boys. *Med. Sport Sci*. 11:39-46, 1978.
23. Malina RM. Anthropometry. Physiological Assessment of Human Fitness. Champaign, IL: Human Kinetics; 1995. p. 205-219.
24. Maughan RJ and Leiper JB. Sodium intake and post-exercise rehydration in man. *European Journal of Applied Physiology*. 71:311-319, 1995.
25. Maughan RJ, Leiper JB, and Shirreffs SM. Restoration of fluid balance after exercise-induced dehydration: effects of food and fluid intake. *European Journal of Applied Physiology*. 73: 317-325, 1996.

26. Maughan RJ, Owen JH, Shirreffs SM, and Leiper JB. Post-exercise rehydration in man: effects of electrolyte addition to ingested fluids. *European Journal of Applied Physiology*. 69:209-215, 1994.
27. Maw MJ, MacKenzie IL, and Taylor NAS. Human body-fluid distribution during exercise in hot, temperature and cool environments. *Acta Physiologica Scandinavica*. 163:297-304, 1998.
28. McKenzie JE, Osgood DW. Validation of a new telemetric core temperature monitor. *Journal of Thermal Biology*. 29:605-611, 2004.
29. Meyer F, Bar-Or O, MacDougall D, and Heigenhauser GJF. Drink composition and the electrolyte balance of children exercising in the heat. *Medicine and Science in Sports and Exercise*. 27(6):882-887, 1995.
30. Meyer F, Bar-Or O, MacDougall D, and Heigenhauser GJF. Sweat electrolyte loss during exercise in the heat: effects of gender and maturation. *Medicine and Science in Sports and Exercise*. 24(7):776-781, 1992.
31. Meyer F, Bar-Or O, Salsberg A, and Passe D. Hypohydration during exercise in children: effect on thirst, drink preferences, and rehydration. *International Journal of Sport Nutrition*. 4:22-35, 1994.
32. Meyer F, Bar-Or O, Wilk B. Children's perceptual responses to ingesting drinks of different compositions during and following exercise in the heat. *International Journal of Sport Nutrition*. 5:13-24, 1995.
33. Mirwald RL, Baxter-Jones ADG, Bailey DA, and Beunen GP. An assessment of maturity from anthropometric measurements. *Medicine and Science in Sports and Exercise*. 34(4):689-694, 2002.
34. Mitchell JB, Phillips MD, Mercer SP, Baylies HL, and Pizza FX. Postexercise rehydration: effect of Na⁺ and volume on restoration of fluid spaces and cardiovascular function. *Journal of Applied Physiology*. 89:1302-1309, 2000.
35. Nose H, Mack GW, Shi X, and Nadel ER. Involvement of sodium retention hormones during rehydration in humans. *Journal of Applied Physiology*. 65(1):332-336, 1988.
36. Nose H, Mack GW, Shi X, and Nadel ER. Role of osmolality and plasma volume during rehydration in humans. *Journal of Applied Physiology*. 65(1):325-331, 1988.
37. Ray ML, Bryan MW, Ruden TM, Baier SM, Sharp RL, and King DK. Effect of sodium in a rehydration beverage when consumed as a fluid or meal. *Journal of Applied Physiology*. 85(4):1329-1336, 1998.

38. Rivera-Brown AM, Gutiérrez R, Gutiérrez JC, Frontera WR, and Bar-Or O. Drink composition, voluntary drinking, and fluid balance in exercising, trained, heat-acclimatized boys. *Journal of Applied Physiology*. 86(1):78-84, 1999.
39. Rowland T, Potts J, Potts T, Sandor G, Goff D, and Ferrone L. Cardiac responses to progressive exercise in normal children: a synthesis. *Medicine and Science in Sports and Exercise*. 32(2): 253-259, 2000.
40. Shirreffs SM and Maughan RJ. Volume repletion after exercise-induced volume depletion in humans: replacement of water and sodium losses. *American Journal of Physiology*. 271(Renal Physiology 43):F868-F875, 1998.
41. Shirreffs SM, Taylor AJ, Leiper JB, and Maughan RJ. Post-exercise rehydration in man: effects of volume consumed and drink sodium content. *Medicine and Science in Sports and Exercise*. 28(10):1260-1271, 1996.
42. Sproles CB, Smith DP, Byrd RJ, and Allen TE. Circulatory responses to submaximal exercise after dehydration and rehydration. *Journal of Sports Medicine*. 16: 98-105, 1976.
43. Turley KR and Wilmore JH. Cardiovascular responses to treadmill and cycle ergometer exercise in children and adults. *Journal of Applied Physiology*. 83(3):948-957, 1997.
44. Wagner JA, Robinson S, Tzankoff SP, and Marino RP. Heat tolerance and acclimatization to work in the heat in relation to age. *Journal of Applied Physiology*. 33(5):616-622, 1972.
45. Wilk B and Bar-Or O. Effect of drink flavor and NaCl on voluntary drinking and hydration in boys exercising in the heat. *Journal of Applied Physiology*. 80(4):1112-1117, 1996.
46. Wilk B, Kriemler S, Keller H, and Bar-Or O. Consistency in preventing voluntary dehydration in boys who drink a flavored carbohydrate-NaCl beverage during exercise in the heat. *International Journal of Sport Nutrition*. 8:1-9, 1998.

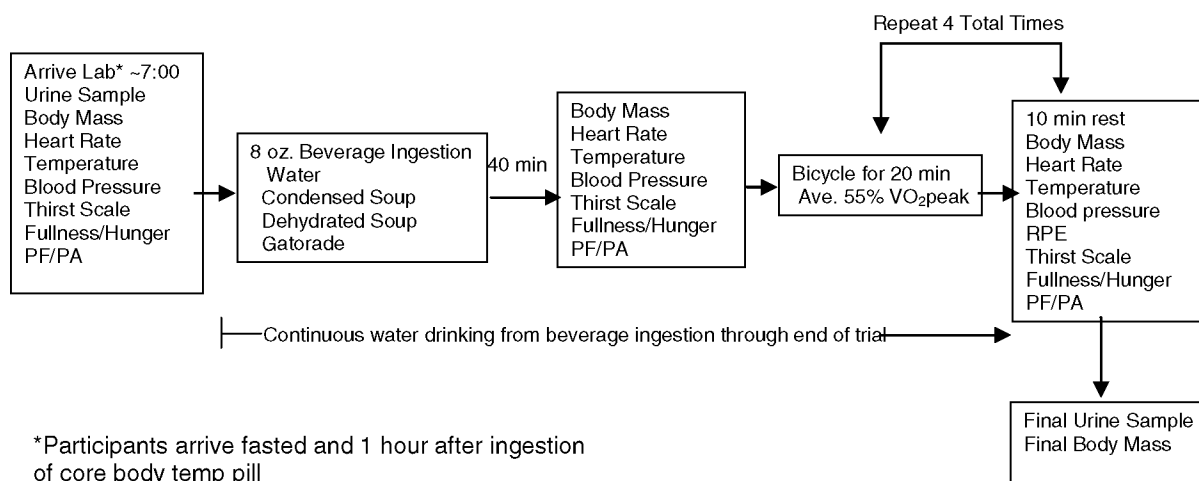


Figure 1. Flow chart of experimental protocol. Subjects completed the experimental protocol 4 times for each experimental beverage. Each trial was separated by a minimum of 1 week and each participant came in at the same time each week.

Table 1. Beverage composition. Osmolality was determined using freezing point depression. All other variables were supplied by the manufacturer.

	W	CE	CS	DS
Osmolality, (mmosm/ kg)	6.5	382.5	283.3	291.0
[Na⁺], (mmol/L)	0.0	20.2	163.6	147.8
[K⁺], (mmol/L)	0.0	3.2	N/A	N/A
Total CHO, (g/L)	0.0	59.2	33.8	71.7
Simple Sugar, (g/L)	0.0	59.2	4.2	8.4
Total Fat, (g/L)	0.0	0.0	8.4	6.3
Total Protein, (g/L)	0.0	0.0	12.7	16.9

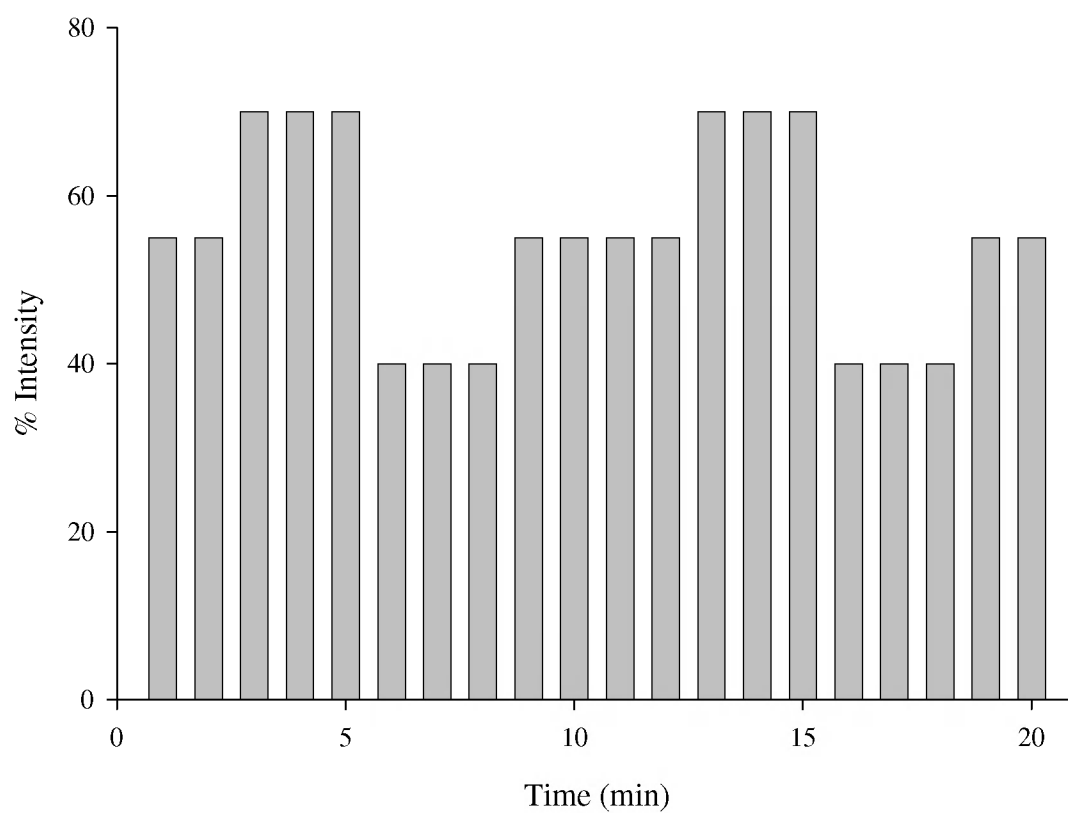


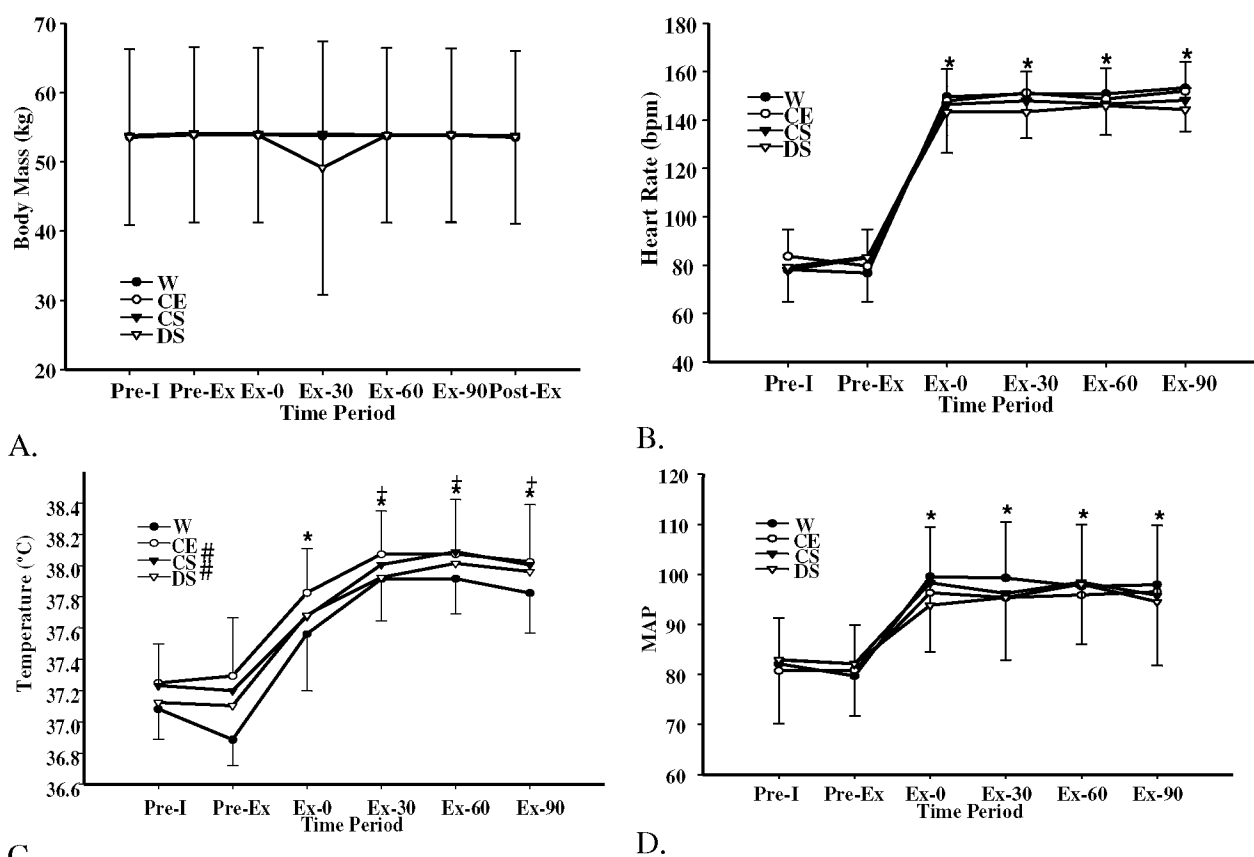
Figure 2. 20-min exercise protocol. The exercise protocol was followed during each exercise bout to allow for an average intensity of 55%.

Table 2. Subject characteristics. Values are mean \pm standard deviation (n=12). There were no differences between gender in any of the categories. Estimated age predicted height (Est. APH) is the estimated age at peak height velocity.

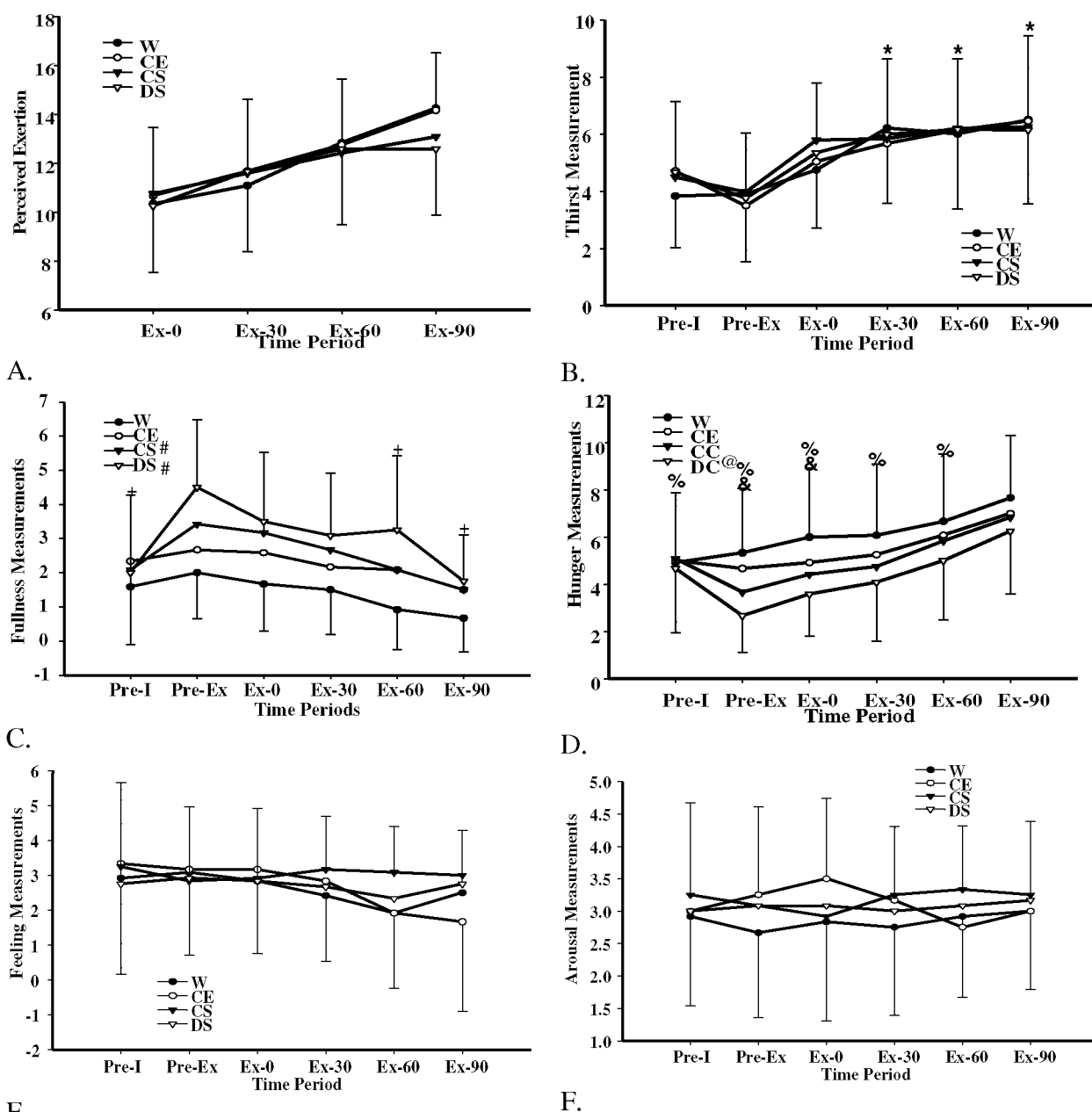
	N	Age (y)	Height (cm)	Mass (kg)	BMI	VO ₂ Peak	Maturity Offset (y)	Est. APH (y)
Females	6	14.1 \pm 0.9	167.6 \pm 15	51.6 \pm 8.0	18.6 \pm 2	43.0 \pm 7	1.9 \pm 0.9	12.2 \pm 0.6
Males	6	14.0 \pm 1.0	165.5 \pm 7.0	56.3 \pm 17	19.5 \pm 4	47.0 \pm 6	0.3 \pm 1.6	13.6 \pm 0.8
Total	12	14.0 \pm 0.9	166.6 \pm 11	54.0 \pm 13	19.1 \pm 3	45.0 \pm 7	1.12 \pm 1.5	12.9 \pm 1.0

Table 3. Urine sample analyses results. Values are mean \pm standard deviation (n=12).* Trials significantly different from W and CE. ⁺Trials significantly different from CS. [#] Trials significantly different from W. $P \leq 0.05$

	Pre-I				Post-Ex			
	W	CE	CC	DC	W	CE	CC	DC
Specific Gravity (g/ml)	1.018 ± 0.007	1.020 ± 0.009	1.019 ± 0.006	1.018 ± 0.008	1.009 ± 0.007	1.011 ± 0.010	1.018* ± 0.007	1.016* ± 0.006
Osmolality (mmosm/kg)	714 ± 254	784 ± 340	763 ± 249	687 ± 263	357 ± 264	437 ± 348	739* ± 260	648* ± 258
[Na⁺] (mmol/L)	184 ± 76	177 ± 65	198 ± 84	143 ± 63	77 ± 62	83 ± 68	179* ± 81	142* ⁺ ± 71
[K⁺] (mmol/L)	51 ± 30	52 ± 40	45 ± 22	42 ± 24	39 ± 21	45 [#] ± 39	77* ± 46	82* ± 36



C. Figure 3. Body mass (A), heart rate (B), temperature (C), and mean arterial pressure (D) responses. Values are mean \pm standard deviation (n=12). * Time points significantly different from Pre-I and Pre-Ex. + Time points significantly different from Ex-0. # Trials significantly different from W. $P \leq 0.05$.



E. Figure 4. Rating of perceived of exertion (A), thirst (B), fullness (C), hunger (D), psychological feeling (E), and psychological arousal (F) responses. Values are mean \pm standard deviation (n=12). * Significantly different from Pre-I and Pre-Ex. + Significantly different from Pre-Ex. # Significantly different from W. @ Significantly different from W and CE. \$Significantly different from Ex-60. % Significantly different from Ex-90. $P \leq 0.05$

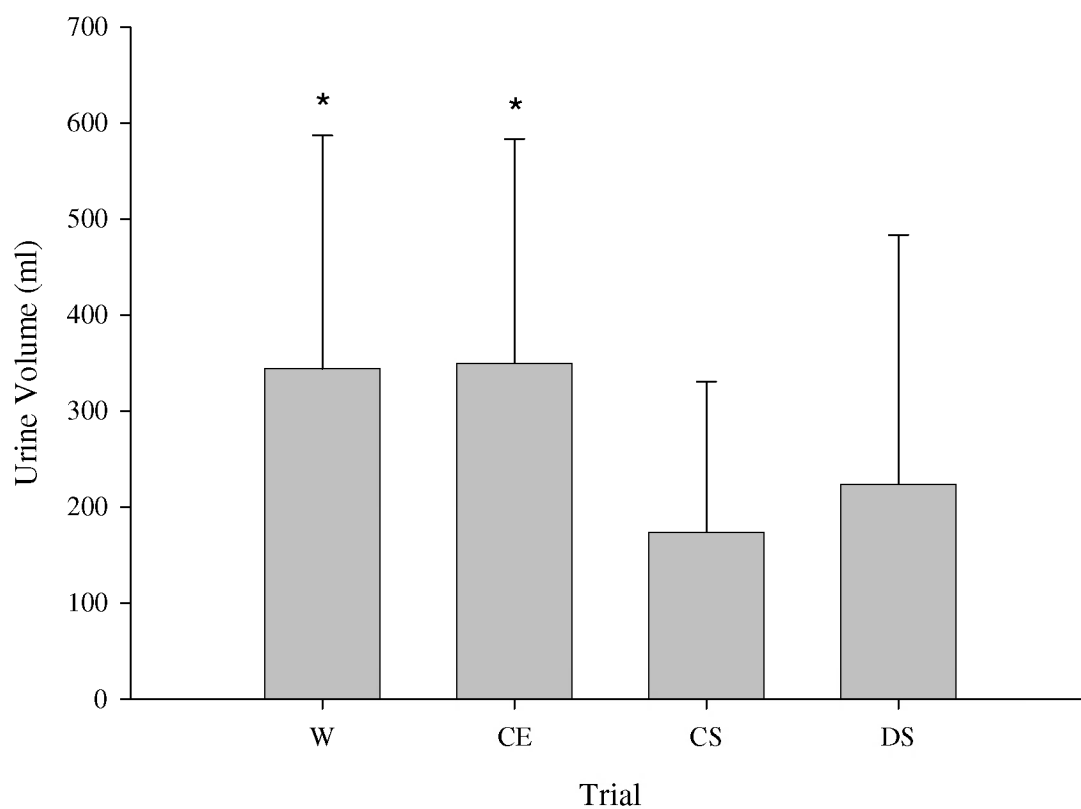


Figure 5. Total volume of urine following ingestion of the test beverage. Values are mean \pm standard deviation (n=12). * Trials CS and DS significantly different from W and CE, $P \leq 0.05$.

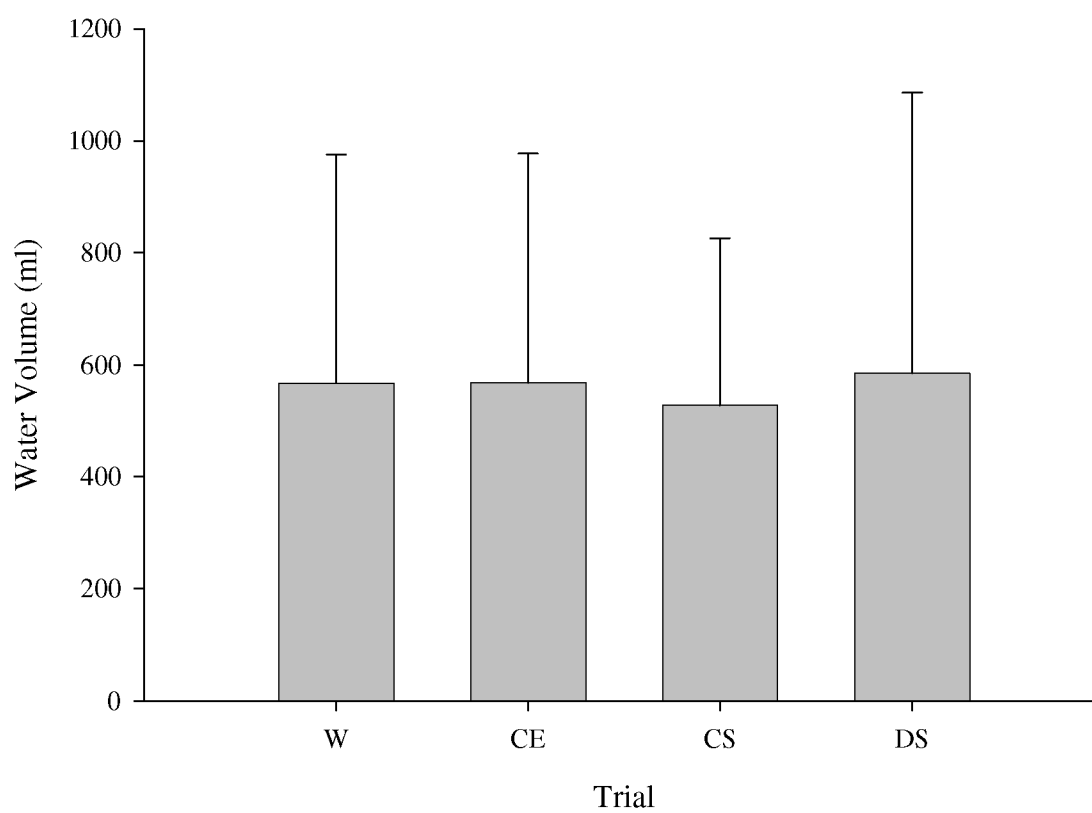


Figure 6. Total water ingested following the ingestion of the test beverage. Values are mean \pm standard deviation (n=12).

Table 4. Total number of drinks taken and volume of water per drink. Number of drinks taken during each hour of the experiment as well as during Pre-Ex and during the exercise/rest periods. Values are mean \pm standard deviation (n=12). *Significantly greater than Hour 1. ⁺Significantly greater than Pre-Ex. $P \leq 0.05$.

Trial	Total Drink #	Volume (ml)/ Drink	Hour 1	Hour 2	Hour 3	Pre-Ex	Ex/Rest
A	2.4 \pm 3.0	141.6 \pm 125.0	0.2 \pm 0.4	1.9 \pm 1.1*	2.9 \pm 1.8*	0.2 \pm 0.4	4.6 \pm 2.9 ⁺
B	2.6 \pm 3.1	121.0 \pm 73.5	0.2 \pm 0.8	2.1 \pm 1.1*	3.3 \pm 2.5*	0.3 \pm 0.9	4.9 \pm 2.8 ⁺
C	2.6 \pm 4.1	121.8 \pm 64.3	0.1 \pm 0.3	2.8 \pm 3.3*	3.3 \pm 2.7*	0.1 \pm 0.3	5.1 \pm 4.6 ⁺
D	2.3 \pm 3.3	145.8 \pm 125.8	0.2 \pm 0.6	2.3 \pm 1.6*	2.5 \pm 2.6*	0.3 \pm 0.6	4.3 \pm 3.7 ⁺

Table 5. Volume of drinks taken during each hour of the experiment as well as during Pre-Ex and during the exercise/rest periods. Values are mean \pm standard deviation (n=12). *Significantly greater than Hour 1. ⁺Significantly greater than Pre-Ex. $P \leq 0.05$.

Trial	Hour 1 (ml)	Hour 2 (ml)	Hour 3 (ml)	Pre-Ex (ml)	Ex/Rest (ml)
A	18.4 \pm 45.1	240.3 \pm 270.0*	287.5 \pm 155.2*	19.9 \pm 46.8	571.5 \pm 393.0 ⁺
B	18.1 \pm 65.3	229.9 \pm 171.8*	328.7 \pm 238.4*	19.6 \pm 68.0	573.6 \pm 368.7 ⁺
C	3.0 \pm 10.9	241.3 \pm 177.6*	308.2 \pm 162.2*	3.3 \pm 11.4	616.3 \pm 317.0 ⁺
D	28.5 \pm 95.9	311.3 \pm 265.7*	248.5 \pm 288.0*	30.9 \pm 99.8	627.7 \pm 550.3 ⁺

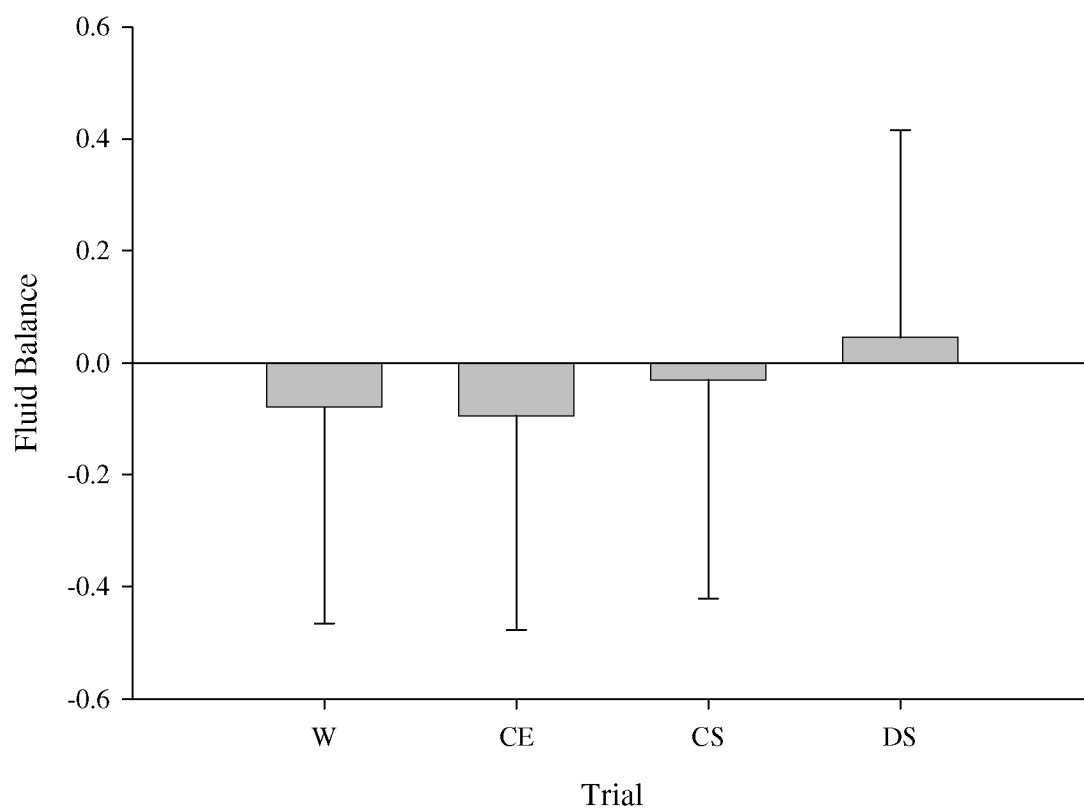


Figure 7. Fluid balance throughout the trial from Pre-I to Post-Ex. Values are mean \pm standard deviation (n=12).

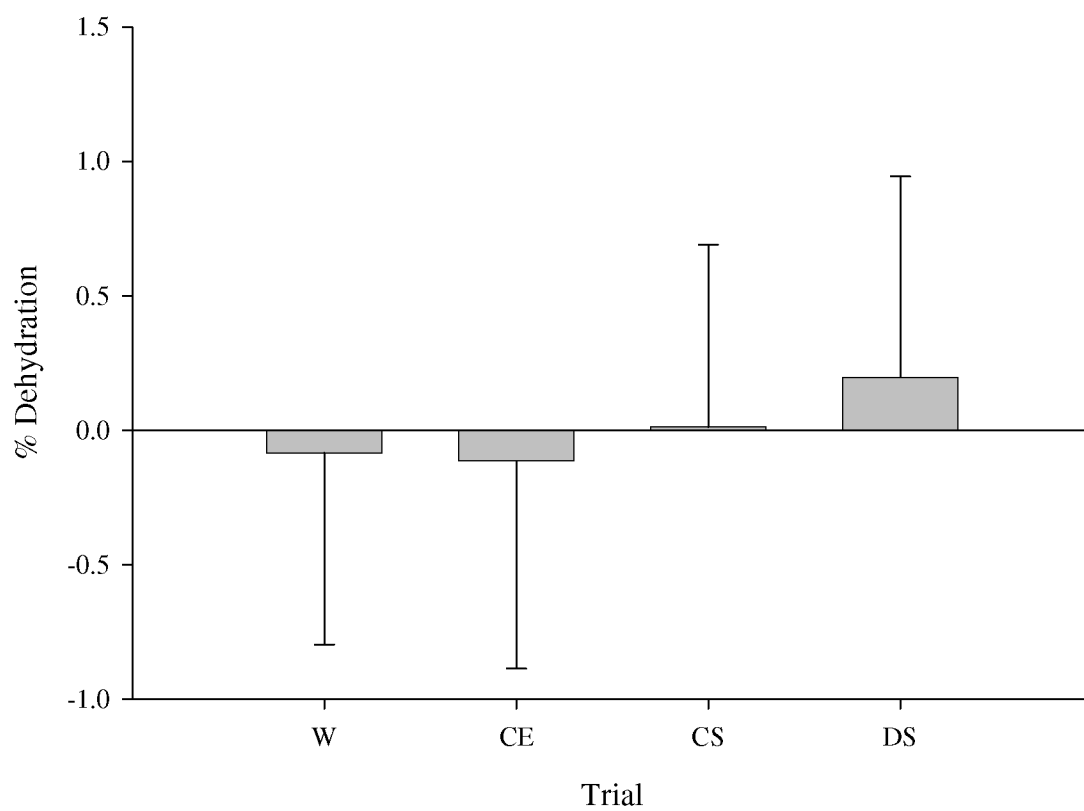


Figure 8. Percent dehydration. Values are \pm standard deviation (n=12).

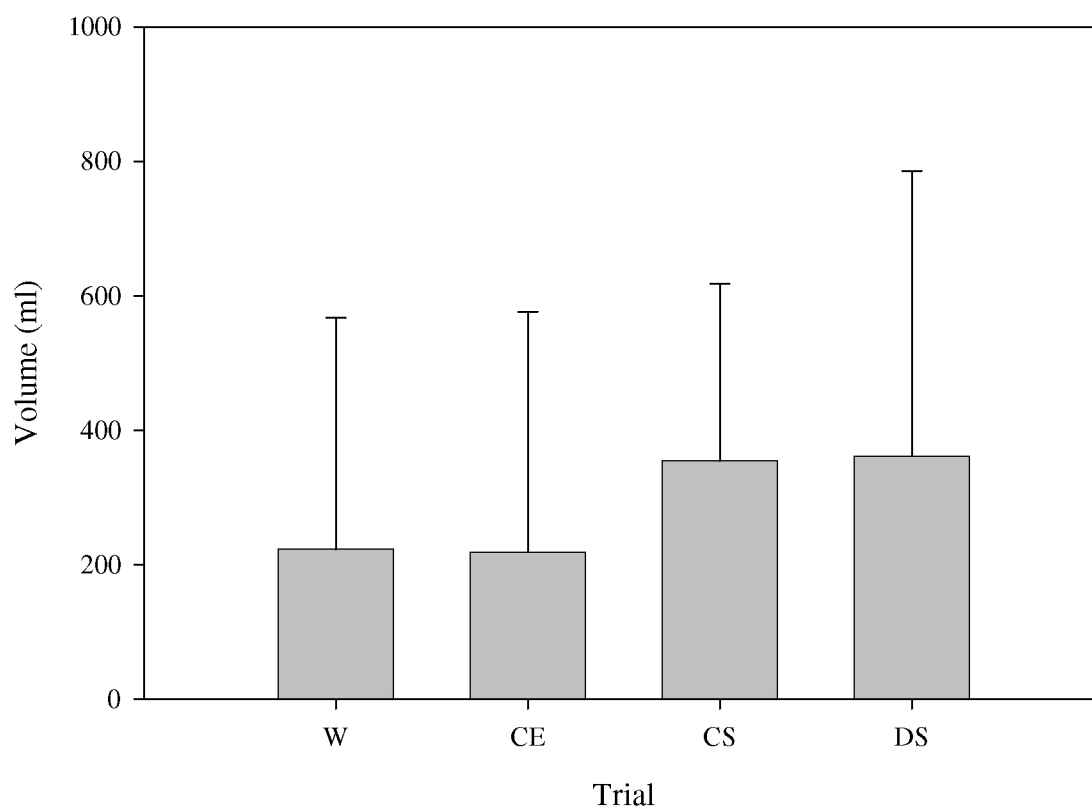


Figure 9. Volume of fluid retained calculated from fluid ingested following the test beverage and the volume lost as urine. Values are mean \pm standard deviation (n=12).